COMPARATIVE STUDY ON WATER QUALITY AND MICROALGAL DIVERSITY FROM THREE DIFFERENT STATIONS OF THOOTHUKUDI COAST

A dissertation submitted to

ST. MARY'S COLLEGE (AUTONOMOUS), THOOTHUKUDI

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MANONMANIAM SUNDARANAR UNIVERSITY, THIRUNELVELI

in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE IN BOTANY

By

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THOOTHUKUDI - 628001

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CERTIFICATE

It is certified that this short term project work entitled **"Comparative Study on Water Quality and Microalgal Diversity from Three Different Stations of Thoothukudi Coast"** submitted to **St. Mary's College (Autonomous)** affiliated to **Manonmaniam Sundaranar University** in partial fulfillment of the requirements for the degree of **Master of Science in Botany** and is a record of work done in the Department of Botany, St. **Mary's College (Autonomous)**, Thoothukudi during the year 2022-2023 by **Poornisha G (19APBO11).**

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DECLARATION

I do hereby declare that this dissertation entitled **"Comparative Study on Water Quality and Microalgal Diversity from Three Different Stations of Thoothukudi Coast"** submitted by me in partial fulfillment for the award of the degree of **Master of Science in Botany**, is the result of my original and independent work carried out under the guidance of **Dr. Sr. A. Arockia Jenecius Alphonse,** Assistant Professor of Botany, St. Mary's College (Autonomous), Thoothukudi and it has not been submitted elsewhere for the award of any other degree.

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CONTENT

INTRODUCTION

INTRODUCTION

The marine system has a huge number of unexplored animals, plants and microorganisms of great interest to researchers. Among these, the marine algae have been extensively studied for various biological and biomedical applications. Marine microalgae are typically found in the marine system and are explored for industrial applications. Among the various marine ecosystem, the coastal ecosystem is found most productive ecosystem. Hence, the diversity and wealth of life in the coast have a special attention among the environmentalist.

Microalgae are unicellular species commonly found in marine and freshwater with the size ranging from a few micro meters to a few hundreds of micro meters. It has been estimated that 2×10^5 to 8×10^5 species exist. The genetic and phenotypic diversity of microalgae is obvious in their ubiquitous distribution in the biosphere. Green microalgae usually grow in freshwater and seawater whereas several other species o microalgae grow in extremely in saline environments. Within these aqueous habits, some algae grow inside a few hundred micro meters of the water layer, others populate the subsurface water column and a few grow at the limits of the photic zone that is 200 – 300m below the water surface.

Richness and productivity of marine ecosystems can be assessed by investing the hydrographical features **(Rajasekar, 2003).** Microalgae, the predominant primary producers are restricted to the neritic zone due to the abundance of nutrients, light and favourable physicochemical variables like temperature, pH and salinity **(Trombettta *et al*., 2020).** Several important nutrients such as nitrate, urea, ammonium, vitamins,

phosphorous, nitrogen, iron, manganese, selenium, cobalt, nickel and zinc are required for the production of any microalgae species.

The relationship between distribution of plankton communities and physical and chemical variables has been studied in several estuaries and frequently salinity and temperature have been shown to be the most important parameters affecting the distribution and abundance of plankton **(Marques *et al*., 2006).** Diversity, distribution and variation in the biotic parameters provide a good indication of energy turnover in aquatic environments.

Microalgae are a promising source for several bioactive compounds. They contain proteins, carbohydrates and lipids. The lipids can be converted into biodiesel, carbohydrates into ethanol and H_2 and proteins into the raw material of biofertilizers. Microalgae have been widely used for various applications including human and animal nutrition, cosmetics, pharmaceuticals, CO_2 capture, bioenergy production and nutrient removal from waste water. Currently, algae have been widely used for fuel production because of their high photosynthetic efficiency, high biomass production and fast growth.

Microalgae have been identified as a potential resource for a number of value-added products as well as feedstock for future green fuels. Their wide diversity makes them an easily cultivable resource that does not interfere with the food, fodder or other products from terrestrial crops. They require much fewer resources as compared to other crops. They are easily modifiable by simple genetic engineering methods to enhance the yields of desired products.

SCOPE AND OBJECTIVE

SCOPE AND OBJECTIVES

Microalgal abundance and distribution critically depending on various physical, chemical and biological factors **(Kimmel *et al*., 1999)** and their ability to assimilate sufficient carbon, nitrogen and phosphorous, as well as minor nutrients, to ensure replication. Availability of nutrients, especially nitrogen and phosphorus, is frequently referred as a key factor regulating microalgal growth and species composition **(Domingues *et al*., 2011).** Besides the essential nutrients, salinity is also an important factor affecting phytoplankton communities in coastal aquatic ecosystems. Therefore, the phytoplanktonic communities display a potential for change that may lead to the substitution, removal or addition of species. Alterations in species richness are mainly due to the variability of abiotic factors, such as short-term climatic variations **(Cody, 1996).** The present study was undertaken

- To analyse the phytoplankton in water samples collected from three different stations of Thoothukudi Coastal region of Southeast Coast of Tamil Nadu

- To assess the distribution of microalgae as influenced by the environmental factors like temperature, salinity, pH, dissolved oxygen and also nutrients (nitrate, nitrite, silicate, ammonia and inorganic phosphate)

- To identify the phytoplankton species by means of Light Microscopic studies

REVIEW OF LITERATURE

HYDROLOGICAL PARAMETERS

The marine environment, as a complex system is mainly influenced by various physical, chemical and biological processes. The open ocean is more stable compared to the near shore waters, where the interaction with the terrestrial zone is more effective in bringing about variations in different physico-chemical parameters. Hence a thorough knowledge of hydrography is indispensable to estimate the quality of the environment and its influence on biological fertility **(Poonam and Rahul, 2012)**.

Physical parameters play an important role in the biochemistry of the water body. Important physical and chemical parameters influencing the aquatic environment are temperature, rainfall, pH, salinity, Dissolved Oxygen and carbon dioxide. Others are total suspended and dissolved solids, total alkalinity and acidity and heavy metal contaminants. These parameters are the limiting factors for the survival of aquatic organisms (flora and fauna) **(Lawson, 2011).** The physical and chemical properties of water immensely influence the uses of a water body for the distribution and richness of biota **(Unanam and Akpan, 2006)** and also influence both vertical and horizontal migration of aquatic organisms. It affects their distribution, diversity and feeding **(Imam and Balarabe, 2012).**

Diversity, distribution and variation in the biotic parameters provide a good indication of energy turnover in aquatic environments. Within these environments, phytoplankton are located at the base level and are represented as a major source of organic carbon **(Gaikwad *et al.*, 2004). Macedo *et al.* (2001)** investigated the annual variation of environmental variables, phytoplankton species composition and photosynthetic parameters in a coastal lagoon. **Collos *et al.* (2003)** examined the

seasonal variations of nitrogen uptake, regeneration and response of coastal phytoplankton to ammonium and nitrate. **Raghunathan *et al*. (2004)** recorded the physico-chemical parameters of sea water in the intertidal zone of Saurashtra coast of Gujarat.

Vijayakumaran (2005) made observations on the productivity parameters in relation to hydrography on the inshore surface waters of the Bay of Bengal. **Abdo (2005)** reported that salinity and temperature were useful parameters describing the chemical constituents of the water and can be considered as general of edaphically relations that contribute to productivity within the water body. **Priyadarshani (2007)** investigated the baseline data on chemical, biological and physical oceanographic parameters in the Gulf of Mannar and Palk bay strait. The physico-chemical parameters and quantity of nutrients in water play significant role in the diversity and ecology of plankton **(Ponmanickam *et al.,* 2007)**.

Ashok Prabu *et al.* (2008) recorded the physico-chemical parameters such as temperature, salinity, pH, dissolved oxygen, total dissolved solids and nutrients like nitrate, nitrite, reactive silicate, ammonia and inorganic phosphate in Vellar and Arasalar estuary, southeast coastal region of Tamil Nadu, for a period of one year (January 2018 to December 2018). Water temperature varied from 25.7 to 33.06 and 26.03 to 33.1. The pH ranged between 7.7 to 8.4. Salinity recorded from 24.16 to 34.0%. Variations in dissolved oxygen content was from 3.4 to 6.1 (mg/L) and total dissolved solids varied from 31.66 to 98.5 (mg/L).

The seasonal variation of physico-chemical characteristics in Point Calimere coastal waters exhibited considerable seasonal and spatial variation in distribution of nutrients in the study area **(Damotharan *et al.,* 2010). Solai *et al*. (2010)** investigated

the implications of physical parameters and trace elements in surface waters of Pondicherry coast. **Damotharan *et al.* (2010)** studied the physico-chemical variability of Kodiakarai, East Coast of India. **Choudhury and Pal (2010)** recorded the nutrient dynamics of Kolkotta coastal waters of Bay of Bengal. **Rao *et al.* (2011)** examined the seasonal variation on physico-chemical parameters of Bheemunipatanam Coast. The hydrographical variations in Kalpakkam Coast, East coast of India were recorded by **Sahu *et al.* (2012). Pati *et al.* (2013)** made an investigation on the physicochemical parameters in Visakhapatnam harbour waters.

The essential parameters for the growth of microalgae are nutrients, temperature, salinity, pH, and mixing of the culture **(Jalal *et al.*, 2013). Archana and Babu (2013)** examined the physico-chemical characteristics of coastal waters along the Visakhapatnam Coast. The multivariate analysis for the physicochemical parameters at Parangipettai, East Coast of India was recorded by **Gnanamoorthy *et al.* (2013). Anjusha *et al.* (2013)** made an observation on physico-chemical parameters and chlorophyll 'a' concentration in the Gulf of Mannar and the Palk Bay, Southeast Coast of India.

Gopinath *et al.* (2013) determined the physicochemical variations along the Southeast coastal waters during January 2018 to December 2018. According to them variations of physico-chemical parameters viz., atmospheric temperature (˚C), water temperature (˚C), salinity (%), pH, dissolved oxygen (mg/L) and total dissolved solids (mg/L) from (26.8 to 34.13), (25.7 to 33.1), (24.16), (7.7 to 8.4), (3.4 to 6.1) and (31.66 to 98.5) respectively. The nutrient concentration viz., nitrate, nitrite, reactive silicate, ammonia and inorganic phosphate varied from (0.848 to 3.312), (0.132 to 0.930), (20.012 to 46.013), (0.118 to 0.581) (0.072 to 5. 054µm/L) respectively.

Nisha and Achyuthan (2014) studied the physico-chemical parameters along the continental slope, Bay of Bengal. These parameters control the growth and yield of lipids and carotenoids **(Kalla and Khan, 2016).**

ISOLATION AND IDENTIFICATION OF MICROALGAE

Phytoplankton act as an important component of the marine ecosystem, as they liberate oxygen during photosynthesis and aid in energy exchange process **(Khan, 2003).** Isolation is a necessary process to obtain pure cultures and presents the first step towards the selection of microalgae strains. Traditional isolation techniques include the use of a micropipette for isolation under a microscope or cell dilution followed by cultivation in liquid media or agar plates. Single cell isolation using micropipettes (e.g., a glass capillary) carried out under inverted microscope is time-consuming, but results in identifiable pure cultures

Another approach in the laboratory includes the enrichment of some microalgae strains by adding nutrients for algal growth. The most important nutrient sources for algal growth are nitrogen and phosphate. Some particular algae species may require trace minerals for their growth (e.g., silicon for diatoms) **(Andersen, 2005).**

Microalgal species have traditionally been discriminated by morphological observations and pigment profiles **(Bast, 2012).** Algal taxonomy is a key discipline in phycology and is critical for physiology, ecology, algal genetics, applied phycology, and bio-assessment **(Manoylov, 2014).** Most of them are distinguished based upon their plastid and flagellar characteristics. Microscope-based microalgal cell identification methods are usually the standard procedures used in laboratories for the

rapid screening of algal samples. **(Godhe *et al.*, 2002; Bertozzini *et al.*, 2005). Lee *et al.* (1998), Grima *et al.* (2003) and Uduman *et al.* (2010)** demonstrated that aluminium sulfate was more efficient in harvesting cells of *Chlorella* sp. compared to pH adjustment using NaOH.

Bharati *et al.* (2001) investigated the planktonic flagellates in relation to pollution in Visakhapatnam harbour, East Coast of India. **Sarojini and Nittala (2001)** examined the vertical distribution in phytoplankton around the Andaman Islands. **Labry *et al.* (2002)** studied the role of phosphorous on planktonic production of the Gironde plume waters in the Bay of Biscay. **Patil (2003)** recorded 128 phytoplankton (83 and 44 species corresponds to diatoms, dinoflagellates) species from the West Coast of India. **Joseph and Saramma (2004)** studied the seasonal and spatial distribution of cyanobacteria and recorded 44 species in Cochin backwater and near shore water. **Madhav and Kondalarao (2004)** made observations on the distribution of phytoplankton with regional and seasonal variations in the offshore waters of Bay of Bengal.

Panigrahi *et al.* (2004) examined the distribution of diatoms and dinoflagellates in tropical waters of Orissa and West Bengal with emphasis on neritic assemblages. **Madhu *et al.* (2006)** investigated the distribution of phytoplankton in western Bay of Bengal during summer, winter and spring inter monsoon periods and found that there was a lack of pronounced seasonal variation in the chlorophyll 'a' and primary production. **Chowdhury and Pal (2008)** studied the diversity of planktonic diatoms from West Bengal Coast with special reference to taxonomic accounts.

The phytoplankton diversity was studied in Pichavaram marine waters, South East coast of India and was identified 94 species of phytoplankton **by Raj Kumar *et al.* (2009)**. Among which, the diatoms formed the predominant group. Maximum phytoplankton diversity and density were observed during summer season which possesses stable hydrographic conditions. **Naik (2010)** investigated the spatial and temporal distribution of dinoflagellates from Chennai to Port Blair and Port Blair to Kolkatta.

Bhavani *et al*. (2013) recorded 57 species in the study of the diversity of cyanobacteria at Tamilnadu. The distribution and abundance of phytoplanktons and recorded 90 species of phytoplankton in the inshore waters of Nizampatnam, East Coast of India. **(Pandiyarajan *et al*., 2014)**. The phytoplankton diversity and cell volume were noticed between the western and central Indian Sundarbans, East Coast of India. (**Mitra *et al.*, 2014).**

RELATION BETWEEN PHYSICOCHEMICAL PARAMETERS AND MARINE MICROALGAE

A marine phytoplankton community is mostly dependent on nutrients and physical parameters in a coastal environment. The nutrient availability is frequently considered as a key factor regulating the phytoplankton abundance, growth and metabolism. Significant work has been done in relation to seasonal variation in phytoplankton species composition in the different coastal ecosystem of India **(Sridhar *et al*., 2006; Sahu *et al*., 2012; Siva Sankar and Padmavathi, 2012). Saravanan *et al*. (2000)** noticed the change in plankton population in response to altered wind direction at Kalapakkam coast of south India.

Phytoplankton abundance and composition in an aquatic ecosystem are regulated by various abiotic or physicochemical factors such as pH, light, temperature, salinity, turbidity and nutrients **(Lewis, 2000).** Increasing nutrient runoff during the past century has provoked increases of phytoplankton production supporting 3-8-fold increases of fish biomass in the Baltic Sea, Japan Seto Inland Sea, northern Adriatic Sea, shelf waters of the Black Sea, and the Nile Delta **(Nixon and Buckley, 2002; Caddy, 2000).** The primary producers in food webs and ensuring ecological balance, phytoplankton are useful indicators of water quality **(Kitner and Poulickova, 2003; Rey *et al.*, 2004).**

Phytoplankton species shows wide variation in distribution due to changes in factor like hydro-chemical and physical parameters. These dramatic changes in physico-chemical parameters, exhibit differential effect in distribution and abundance of many phytoplankton species, ultimately indicating the quality of water **(Liu *et al.*, 2005; Shashi Shekhar *et al.*, 2008). Sridhar *et al*. (2006)** examined the water quality in terms of physicochemical characteristics and plankton distribution in Palk Bay. The physico-chemical parameters and quantity of nutrients in water play significant role in the diversity and ecology of plankton **(Ponmanickam *et al.*, 2007).**

Paul *et al.* (2008) studied the seasonal variation of nutrient and their impact on the phytoplankton distribution in coastal and off shore of Bay of Bengal. They observed declines in eight out of ten ocean regions and estimated a global rate of decline. The analyses further revealed interannual to decadal phytoplankton fluctuations super-imposed on long term trends. **Castillo and Vazquez (2008)** reported the relationship between the phytoplankton variation and nutrient concentration in Gulf of Mexico. **Panda *et al.* (2012)** examined the phytoplankton

diversity response to abiotic factors along Orissa Coast, Bay of Bengal.

Jyothibabu *et al.* (2013) studied ecology and tropic preference of picoplankton and nanoplankton in the Gulf of Mannar and the Palk Bay, Southeast Coast of India. **Dogiparti *et al.* (2013)** studied the distribution and diversity of phytoplankton in relation to physico-chemical parameters Bhavanapadu creek, East Coast of India. **Ayajuddin *et al.* (2014)** studied the phytoplankton distribution and species composition on a salinity gradient at surface and subsurface layers and reported 52 species at Kakinada, East Coast of India. Use of multivariate procedures (PCA, MDS, CCA, etc.) are increasingly being used to analyze water chemistry and phytoplankton characteristics in coastal aquatic habitats **(Philippart *et al*., 2000; Nasrollahzadeh, 2008).**

MATERIALS AND METHODS

DESCRIPTION OF STUDY AREA

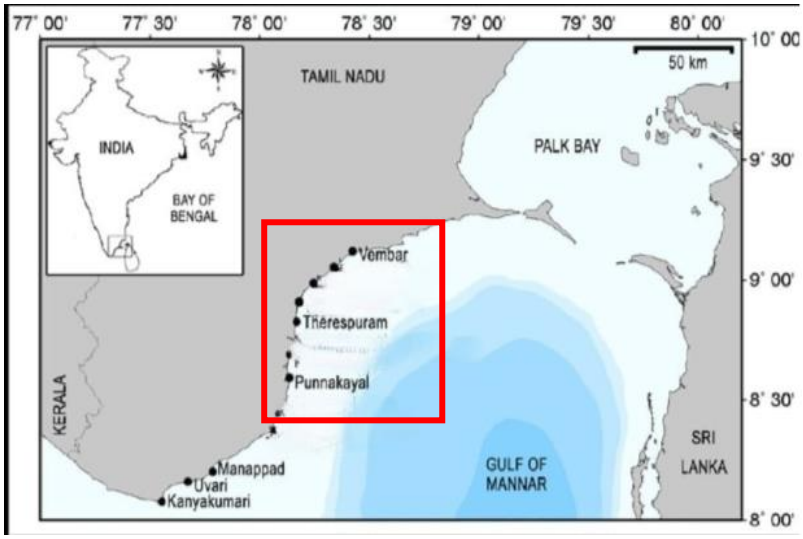
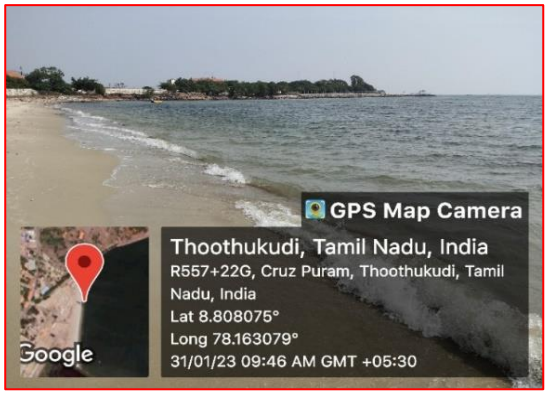


Figure 1: Map showing the study area

Thoothukudi is a port city, a municipal corporation and an industrial area of Tamil Nadu, India. The city lies in the Coromandel Coast of Bay of Bengal. Thoothukudi is the capital and headquarters of Thoothukudi district. It is located about 590 kilometres (367 miles) southwest of Chennai, 190 kilometres (118 miles) northeast of Thiruvananthapuram and 580 kilometres (360 miles) southeast of Bengaluru. Thoothukudi consists of 21 islands between Thoothukudi and Rameswaram shores in the Gulf of Mannar and are notified as the first Marine Biosphere Reserve of India. The region around the Thoothukudi shores are home to rare marine flora and fauna. For the present study, three locations of Thoothukudi district were chosen namely, Vembar, Threspuram and Punnakayal **(Plate 1).**

PLATE 1: SAMPLING STATIONS OF THOOTHUKUDI COAST

STATION I: VEMBAR



STATION II: THERESPURAM

STATION III: PUNNAKAYAL

STATION I: VEMBAR

It lies on the latitudes 8.996223^{o} and longitude 78.255771^{o} and total shoreline length is about 12 km. This coastline is formed by sediments and carried out by small rivers and recycled sediments of waves and currents. A pocket of beach rocks is noticed here and there in the coastline. Southwest and northeast monsoonal waves and littoral currents have played major role on changes in the shoreline configuration

STATION II: THRESPURAM

The study area (latitude 8.808075^{o} and longitude 78.163079^{o}) is a coastal village of Thoothukudi located on 8 km away from Thoothukudi Thermal power station and 5 to 6 km distance from Thoothukudi fishing harbour. Hereby load of untreated sewage, human and animal faecal matter are mixed with coastal water in this station. Salt pans and small fish processing industries located around this station release fish wastes and salt pan effluents in this area.

STATION III: PUNNAKAYAL

The study area is a part of the eastern coast of Tamilnadu namely Tuticorin. It falls on the latitude 8.637021^{o} and longitude 78.122176 ^{o}. The area is characterized by sandy beach. Apart from this beach ridges, sandy flats and creeks are some of the geomorphic features observed in the area.

METHOD OF COLLECTION

Surface water and samples were collected from three different stations of Thoothukudi coast from December 2022 to March 2023 in polyethylene containers for physico-chemical analysis and were transferred to the laboratory shortly after collection. Various chemical parameters were analysed within a day. Plankton were collected using plankton net (mesh size 20 μm) made up of bolting silk cloth. All the samples were brought to the laboratory and examined under the light microscope and

the micro algae were identified using standard manuals of **(Venkataraman 1939, Subramanyan 1946, Desikachary 1959, Smith 1977, Santhanam *et al.*, 1987 and Al-Kandari *et al.*, 2009).**

PHYSICO-CHEMICAL PROPERTIES OF MARINE WATER

Hydrogen Ion Concentration (pH)

The pH value of marine water collected from three different stations was measured by digital pH meter (AP – 1 PLUS)

TEMPERATURE (°C)

Temperature of water samples collected from study areas was measured using a standard thermometer at the time of water sample collection.

TOTAL DISSOLVED SOLIDS (APHA, 1999)

Hundred milliliters of water samples were taken in a pre-weighed evaporating dish, kept at 180°C ± 2°C for one hour in a hot water bath, cooled it in a desiccator and final weight was recorded using the following formula

$$\text{Dissolved Solids at 180°C} = \frac{\text{Initial weight} - \text{Final weight}}{\text{Volume of sample}} \times 1000$$

TOTAL ALKALINITY (APHA 1998**)**

Reagents:

- **0.02 N H_2SO_4**: Dilute 6 ml of concentrated H_2SO_4 (35 N) with distilled water under cooling and make up to one litre with more water in a volumetric flask. This stock solution gives a 0.2 N H_2SO_4. So dilute 100 mL of this stock

solution to 1 litre with distilled water to get a 0.02 H_2SO_4 solution. Standardize this acid against a standard solution of sodium carbonate.

- **Phenolphthalein**: Dissolve 0.25 g of phenolphthalein powder in 50 mL of 50% alcohol.
- **Methyl orange – bromo cresol green**: Dissolve 0.01 g methyl orange and 0.05 g of bromocresol green in 100 mL of water, colour change at end point (pH 4.3) is sharper than that shown by methyl orange alone.

Procedure:

- Measure 100 mL of the sample and transfer it into a 250 mL conical flask.
- Add 3 – 4 drops of phenolphthalein indicator to the sample and titrate against standard
- (0.02 N) H_2SO_4 from the burette. Note the titre value.
- Then to the same sample add 3 – 4 drops of the mixed indicator and titrate again with H_2SO_4 to the point (blue green to orange) and note the titre value.

HARDNESS (APHA 1999)

Hardness is measured by computing the results of separate determinations of calcium and magnesium.

Hardness, mg equivalent CaCO3/L = 2.497 [Ca, mg/L] + 4.118 [Mg, mg/L]

ESTIMATION OF CALCIUM AND MAGNESIUM (Mackareth, 1963)

Reagents:

- **EDTA disodium salt (0.1 N)**: Dissolve 16.825 g of EDTA in one liter of distilled water. In order to avoid probable decomposition, it is better to keep this solution in a polythene bottle.
    - 1 mL of 0.1 N EDTA = 2 mg of calcium
    - 1 mL of 0.1 N EDTA = 1.2 mg of magnesium

- **Ammonium Buffer (pH 10)**: Dissolve 70 g of ammonium chloride in 570 mL of liquor ammonia and make upto 1 litre.
- **Sodium hydroxide**: Dissolve 80 g of sodium hydroxide in 1 g of distilled water.
- **Eriochrome Black T**: Grind and mix 200 g of sodium chloride and 1g of Eriochrome black T.
- **Ammonium Purpurate**: Make a solid mixture of murexide and sodium chloride in the ratio of 1:100.

Procedure:

- Take 5 mL of the water sample in a conical flask and add 5 mL of ammonium buffer.
- Dilute it to 100 mL with distilled water and add a pinch of Eriochrome Black T.
- Warm the solution to 60°C and titrate it against EDTA from the burette. The end point of the titration is from wine red to blue. This titrate value is taken as A.
- Take another 5 mL of the sample in a conical flask and add 5 mL of sodium hydroxide followed by a pinch of Murexide indicator.
- Dilute the solution to 100 mL with distilled water and titrate it against EDTA until the colour changes from pink to purple. This titrate value is taken as B.

Calculations:

Consumption of EDTA due to calcium and magnesium = A

Consumption of EDTA due to calcium alone = B

Consumption of EDTA due to magnesium = A – B

Factor value (F) for calcium = 2 (mg)

$$\therefore \text{Amount of calcium in the sample} = \frac{F \times B \times 1000}{\text{Sample volume}} \text{ mg/l}$$

$$\text{Factor value for magnesium} = 1.2 \text{ (mg)}$$

$$\therefore \text{ Amount of magnesium in the sample} = \frac{F \times (A - B) \times 1000}{\text{Sample volume}} \text{ mg/l}$$

DETERMINATION OF TOTAL PHOSPHOROUS

Reagents:

- **Double distilled water:** To 1 litre of distilled water in a distillation flask, add 0.5 g sodium hydroxide and 0.5 g potassium per sulphate. Boil for 20 min. Connect the condenser and distil. Discard the first 100 mL. Collect the next 750 mL, leaving around 150 mL in the flask (check for the absence of phosphate).
- **Sulphuric acid (9.0 N):** Add carefully with cooling, 250 mL concentrated sulphuric acid to 750 mL water and dilute to 1000 mL.
- **Ammonium molybdate solution (10%):** Dissolve 12.5 g Ammonium molybdate tetrahydrate in 125 mL water. Store in a plastic bottle.
- **Potassium antimonyl tartarate solution (2.5%):** Dissolve 0.5 g potassium antimonyl tartarate in 20 mL water. Store in a glass bottle.
- **Mixed Reagent:** Add slowly, with stirring 125 mL molybdate solution to 350 mL 9.0 N sulphuric acid. Then add 20 mL antimonyl tartarate solution. Mixed reagent is stable for many months. Store in an amber coloured glass bottle.
- **Ascorbic acid solution (10%):** Dissolve 10 g ascorbic acid in 50 mL water and add 50 mL 9.0 N sulphuric acid. Store in an amber coloured glass bottle

in a refrigerator. The reagent can be used for a week or as long as it remains colourless.

- **Phosphate standard solution:** Weigh accurately 0.136 g potassium dihydrogen phosphate and dissolve in 100 mL water containing 1 mL of 9.0 N sulphuric acid. This solution contains 10 µmol phosphate – P/mL. Prior to weighing, dry potassium dihydrogen phosphate in an oven at 110°C and cool in a desiccator.
- **Phosphate working standard solution:** Transfer 5 mL of phosphate standard solution to a 500 mL volumetric flask and dilute to the mark with water. Transfer 5 mL of this solution to a 500 mL volumetric flask and dilute to 500 mL. This working solution contains 1µmol phosphate-P/mL. Prepare fresh.

Procedure:

Blank and Standard: Measure out three 50 mL portions of water in to three oxidation bottles. Similarly, measure out 50 mL portions of working standard into three other bottles. Add 5.0 mL oxidizing reagent to all the flasks, stopper and keep them in the autoclave / pressure cooker. Heat and maintain under pressure conditions for 10 min. Cool. Take out the flask from the autoclave, swirl to dissolve any precipitate and open them carefully to release any over pressure. Allow the flask to cool. Transfer 25 mL of digested solutions to the glass tubes. Add 0.5 mL of ascorbic acid reagent to each tube and mix well. Add 0.5 mL of mixed reagent, mix and wait for 10 min. to allow for the development of blue complex. Measure the absorbance of blank and standards in a spectrophotometer using 50 mm cells at 880 nm with distilled water as reference.

Sample

Take 50 mL of sea water in the oxidation bottles. Add 5.0 mL oxidizing reagent to all the flasks, stopper and keep them in the autoclave / pressure cooker. Heat and maintain under pressure conditions for 10 min. Cool. Take out the flask from the autoclave, swirl to dissolve any precipitate and open them carefully to release any over pressure. Allow the flask to cool. Transfer 25 mL of digested solutions to the glass tubes. Add 0.5 mL of ascorbic acid reagent to each tube and mix well. Add 0.5 mL of mixed reagent, mix and wait for 10 min. to allow for the development of blue complex. Measure the absorbance of sample using 50 mm cells at 880 nm with distilled water as reference.

DETERMINATION OF NITRITE – NITROGEN (Parsons *et al*. 1984)

Reagents

- **Double distilled water:** To 1 litre of distilled water in a distillation flask, add 0.5 g sodium hydroxide and 0.5 g potassium per sulphate. Boil for 20 min. Connect the condenser and distil. Discard the first 100 mL. Collect the next 750 mL, leaving around 150 mL in the flask (check for the absence of nitrite)
- **Sulfanilamide solution (1%):** Dissolve 2.5 g sulfanilamide in 25 mL conc. Hydrochloric acid and make up to 150 mL with water. Dilute to 250 ml with water. Store in an amber coloured glass bottle.
- **N-(1-naphthyl) – ethylenediamine dihydrochloric acid (0.1 %)** – Dissolve 0.25 g in 250 mL water. Store in an amber coloured glass bottle.
- **Nitrite standard solution**: Dissolve 0.345g anhydrous sodium nitrite (dried at 110°C for 1 hr) in 100 mL and dilute to 1000 mL. The solution contains 10 m mol Nitrite-N/l. Store in an amber coloured glass bottle.

- **Nitrite working standard solution**: Pipette out 10 mL of stock solution into a 500 mL volumetric flask and dilute to 500 mL. Transfer 5 mL of this solution to a 500 mL flask and dilute to mark. This standard has 2µmol Nitrite-N/l.

Procedure

Blank and Standard: Take three 25 mL portions of nitrite free distilled water in three tubes. Measure out three 25 mL portions of working standard solution into three tubes. To each tube add 0.5 mL of sulphanilamide, mix and wait for 4 min. Add 0.5 mL of N-(1-naphthyl)-ethylene diamine dihydrochloride solution. Mix the contents well and allow the reaction to proceed for 10 min. Measure the absorbance of blanks and standards in a 50 mm cell against water as reference at 543 nm.

Sample: Take three 25 mL portions of water samples in three tubes. To each tube add 0.5 mL of sulphanilamide, mix and wait for 4 min. Add 0.5 mL of N-(1-naphthyl)-ethylene diamine dihydrochloride solution. Mix the contents well and allow the reaction to proceed for 10 min. Measure the absorbance of sample in a 50 mm cell against water as reference at 543 nm.

DETERMINATION OF AMMONIA (Parsons *et al.* 1984)

Reagents:

- **Deionized water:** Distilled water is passed through a cation exchange column in the hydrogen form. This water should be prepared fresh for use.
- **Phenol Solution**: Dissolve 10 g of phenol in a consisting of 100 mL of 95% v/v ethyl alcohol.
- **Sodium nitroprusside solution**: Dissolve 1 g of sodium nitroprusside in 200 mL of distilled water. Store in amber bottle.

- **Sodium citrate**: Dissolve 100 g of sodium citrate and 5 g of sodium hydroxide in 500 mL of de-ionized water.
- **Sodium hypochlorite**: Commercially available hypochlorite ("Chlorox") of 1.5 N.
- **Oxidizing solution**: Mix 100 mL of sodium citrate solution and 25 mL of sodium hypochlorite solution just before use.

Procedure:

Transfer 50 mL water into a conical flask and add 2 mL of phenol solution. Mix well and add 2 mL of sodium nitroprusside solution followed by 5 mL of oxidizing solution. Mix thoroughly. Cover the flask with polythene sheet and wait for 1 hour. Measure the extinction (O.D) of the reacted sample against ammonia free distilled water reagent blank at 640 nm using 10 cm cell.

ESTIMATION OF CHLORIDE (Strickland and Parsons, 1968)

Reagents:

- **Potassium chromate indicator solution**: Dissolve 50 g K_2CrO_4 in a little distilled water. Add $AgNO_3$ solution until a definite red precipitate is formed. Let stand 12 hrs, filter, and dilute to 1 L with distilled water.
- **Standard silver nitrate titrant (0.0141 N)**: Dissolve 2.395 g $AgNO_3$ in distilled water and dilute to 1000 mL. Store in a brown bottle.
- **Standard sodium chloride (0.0141 N)**: Dissolve 824.0 mg NaCl (dried at 140°C) in distilled water and dilute to 1000 mL.

Special reagents for removal of interference:

- **Aluminum hydroxide suspension**: Dissolve 125 g aluminum potassium sulfate or aluminum ammonium sulfate, $AlK(SO_4)_2$. $12H_2O$ or $AlNH_4(SO_4)_2.12H_2O$, in 1 L distilled water. Warm to 60°C and add 55 mL

conc. ammonium hydroxide (NH_4OH) slowly with stirring. Let stand about 1 hour, transfer to a large bottle, and wash precipitate by successive additions, with through mixing and decanting with distilled water, until free from chloride. When freshly prepared, the suspension occupies a volume of approximately 1 L.

- **Phenolphthalein indicator solution.**
- **Sodium hydroxide**, NaOH, 1N.
- **Sulfuric acid**, H_2SO_4, 1N.
- **Hydrogen peroxide**, H_2O_2 30%.

Procedure:

Sample preparation: Use a 100 mL sample or a suitable portion diluted to 100 mL. If the sample is highly colored, add 3 mL Al $(OH)_3$ suspension, mix, let settle, and filter. If sulfide, sulfite, or thiosulfate is present, add 1 mL H_2O_2 and stir for 1 min.

Titration: Directly titrate samples in the pH range 7 to 10. Adjust sample pH to 7 to 10 with H_2SO_4 or NaOH if it is not in this range. Add 1.0 mL K_2CrO_4 indicator solution. Titrate with standard $AgNO_3$ titrant to a pinkish yellow end point. Be consistent in end-point recognition. Standardize $AgNO_3$ titrant and establish reagent blank value by the titration method outlined above. A blank of 0.2 to 0.3 mL is usual.

DETERMINATION OF NITRATE (APHA 1998)

Reagents:

- **Standard nitrate solution:** Dissolve 13.7 mg sodium nitrate in 100 mL distilled water.

- **Brucine reagent:** Take 50 mL of water and 3 mL of concentrated HCL in a beaker. Heat this to boiling and add one-gram brucine sulphate and 0.1g sulphanilic acid with stirring. Cool and dilute this solution to 100 mL.
- **Sulphuric acid:** Mix carefully 500 mL concentrated sulphuric acid with 100 mL distilled water

Procedure:

Prepare a series of 50 mL standard solution of the nitrate from the standard nitrate solution to obtain a range of concentration. Pipette 2mL aliquots of the standard solution into different 100 mL beakers provided with glass rods. To each beaker, add 1 mL brucine-sulphanilic acid reagent and 10mL sulphuric acid solution. Stir gently for about 5 minutes. Cover the beakers with watch glasses and keep them in the dark for 10 minutes during which time a yellow colour develops. Add 10 mL distilled water to each of the beakers and incubate in the dark for 30 minutes. The absorbance of each solution is then measured at 410 nm. A graph is drawn by plotting the absorbance values against the nitrate concentration of the standard solutions.

DETERMINATION OF SULPHATE (APHA 1999)

Reagents:

- **NaCl - HCl solution:** 60 g of NaCl was dissolved in 200 mL of distilled water. To this 5 mL of cone. HCl was added and this solution was made upto 250 mL with distilled water.
- **Glycerol - ethanol solution**: 50 mL of glycerol was added to 100 mL of ethyl alcohol and mixed thoroughly.
- **Barium chloride** (AR Grade) dry crystals.

Procedure:

5 ml of the sea water was diluted to 50 mL with distilled water and 50 mL of distilled water (blank) was taken separately in 250 mL conical flask. Add 10 mL of NaCl - HCl solution followed by glycerol - ethanol solution. Then added 0.15 g of barium chloride and mixed for 60 seconds over a magnetic stirrer. Absorbance was measured at 420 nm in spectrophotometer. The amount of sulphate was calculated by using the sulphate content from the standard curve.

ESTIMATION OF SODIUM

The sodium content in the water samples were determined by flame photometer (APHA 1999).

Reagents:

- Sodium chloride standard solution: Dissolve 2.542 g NaCl dried at 140°C to constant weight and dilute to 1000 mL with water; 1.00 mL = 1.00 mg Na.

Procedure:

10 mL of aliquot of sample was diluted with 100 mL distilled water in a volumetric flask and used for analysis. Standard curve was prepared by graded concentrations of NaCl and by noting the readings in flame photometer. The concentration of sodium in the water sample was determined from the standard curve.

ESTIMATION OF POTASSIUM

The potassium content in the water samples were determined by flame photometer (APHA 1999).

Reagents:

- Potassium chloride standard solution: : Dissolve 1.907 g KCl dried at 110°C and dilute to 1000 mL with water; 1 mL = 1.00 mg K .

Procedure:

10 mL of sample was diluted with 100 mL distilled water in a volumetric flask and used for analysis. Standard curve was prepared by graded concentrations of KCl and by noting the readings in flame photometer. The concentration of potassium in the water sample was determined from the standard curve.

STATISTICAL ANALYSIS

Statistical analysis work has done by using data analysis tool package of MS office Excel. The experiments were conducted in three replicates using the same treatments. The data of all values were statistically analyzed and expressed as mean ± standard deviation.

PHYTOPLANKTON ISOLATION AND CULTURE MAINTENANCE

The cultures were grown and maintained in ASN medium. It is widely used for culturing of marine microalgae in laboratory. Seawater used for the preparation of 15lbs pressure lbs pressure 120°C for 15 min. The sterilized ASN medium was prepared, cooled and used for culture. Phytoplankton were isolated by serial dilution and micropipette method adopted. The isolated species of *Chlorella spp., Oscillatoria spp*. and *Synechococcus* were maintained in 250 ml, 2000ml, 5000ml conical flasks containing 100 ml of ASN media (prepared in 25 ppt sea water 27˚C temperature and 4500 ± 500 Lux light intensity with 18:6 hours light : dark cycle for a period of 9 days. Cells at different light intensity cultures were counted every day.

RESULTS AND DISCUSSION

The physico-chemical parameters of the surface water samples collected from three different stations (Station 1: Vembar, Station 2: Therespuram, Station 3: Punnakkayal) of Thoothukudi coast were analyzed for the periods of January 2023 to March 2023 according to the standards of the American Public Health Association (APHA). The average values of the physico-chemical parameters of the water samples were recorded and are tabulated in **Tables 1 and 2.**

pH

The pH changes in the water are governed by the amount of free CO_2, carbonates and bicarbonates. These changes are accompanied by the changes in other physico-chemical aspects that in turn influence the quality of water. During the study period, the average pH range varied from 7.46 to 7.91 for the three stations **(Fig.2A).** The average minimum pH (7.46) was recorded at station 2 and the average maximum pH (7.91) was recorded at station 1. **Boyd and Lichtcoppler (1979)** reported pH range of 6.09 – 8.45 as being ideal for supporting aquatic life including fish. **According to Umavathi *et al*. (2007)** pH range between 5 to 8.5 is best for plankton growth. In this study, pH ranged between 7.47 to 7.91 obtained from three different stations of study area with overall mean value of 7.6. The results show that water from all the three locations were moderately alkaline (pH 7.47 to 7.91) and within the permissible limit (pH 6.5 – 8.5) for plankton growth.

TEMPERATURE (◦C)

Temperature is one of the most important factors in the coastal ecosystem which influence the physico-chemical characters of coastal water and also influence the distribution and abundance of flora and fauna **(Sundaramanickam *et al*.,**

2008). The average value of minimum water temperature was recorded as 28.67◦C in near shore region (TNSR) at station 2 while the average maximum water temperature was recorded as 30.33◦C in near shore region (VNSR) at station 1. Between the two regions (Near Shore Region - NSR and Off Shore Region - OSR) of sea, water temperature is more in OSR at stations 2 and 3 whereas at station 1, the result was vice -versa **(Fig. 2B).** From these observations, it could be understood that the water temperature is governed by atmospheric temperature of the area, however, these variations were not significantly different across the stations.

SALINITY (ppt)

Salinity is a fundamental water quality parameter mentioned by freshwater and marine ecologist because of its influence on the biota. Most aquatic organisms are adapted to only a narrow range of salinity beyond which they cannot maintain their osmotic and ionic balance. Some species tolerate only intermediate levels of salinity while broadly adopted species can acclimate to variable salinity ranging from freshwater to seawater. Generally, the range of salinity in brackish water is from 0.5 ppt to 30ppt **(Karleskint *et al.*, 2009).** In this study, salinity ranged between 30ppt and 32.67ppt was recorded **(Fig. 2C).** The average maximum value of salinity was recorded as 32.67 ppt in NSR at stations 1 and 2 whereas, at station 3, the maximum value of salinity (31ppt) was recorded in POSR. Minimum salinity was recorded as 30 in VOSR and TOSR at stations 1 and 2 respectively. The salinity was higher at stations 1 and 2 compared to station 3. The recorded higher values of salinity could be attributed to the low amount of rainfall, higher rate of evaporation and also due to neritic water dominance **(Balasubramanian and Kannan, 2005; Sridhar *et al.*, 2006).**

TOTAL DISSOLVED SOLID (mg / L)

The total dissolved solids ranged from 597.1 mg / L to 697.9 mg / L in the surface water collected from all the three stations of Thoothukudi coast **(Fig. 2D).** Minimum TDS (597.1 mg / L) was recorded in VOSR at station 1 and maximum was noticed in PNSR at station 3. The values were decreasing with increasing distance from disposal area. High content of dissolved solids elevated density of water, influencing osmoregulation and reducing gas solubility, utility of water for drinking, irrigation and industries **(Manikavasakam, 2003).** The total dissolved solids have been reported to be directly related to biological productivity **(Rawson, 1957).**

HARDNESS (mg / L)

Figure 2E. shows that the total hardness ranged from 5477 mg / L to 6558 mg / L. The highest value (6558.14 mg / L) was recorded in VNSR at station 1 while the lowest value (5477.79 mg / L) was noticed in PNSR at station 3. The increase in the total hardness of the sea water would probably be related to the supply by the oueds and outfalls of the leaching waters of the calcareous soils in the region. However, the decrease of this parameter could be correlated with upwelling currents, which include the deep mineral rich waters responsible for maintaining the ions expressed in the total hardness of the water **(Naciri, 1990).** Water hardness up to 60 mg / L is considered as soft water, from 61 – 121 mg / L as moderately hard water, from 121 – 180 mg / L as hard water and above 180 mg /L as very hard water **(Kannan, 1991).**

Nutrients are considered as one of the most important parameters in marine environment influencing growth, reproduction and metabolic activities of living being. Distribution of nutrients is mainly base on the season, tidal conditions and

fresh water flow from land source. The life supporting processes in the sea requires an array of inorganic substances but the role of nitrogen, phosphorous and silicon are considered vital in marine ecosystem. Among nitrogenous nutrients, nitrite, nitrate and ammonia are the major constituents which play key roles in the phytoplankton growth and proliferation.

AMMONIA (mg / L)

The ammonia concentration ranged from 0.03 to 0.04 mg / L, 0.05 to 0.06 mg / L and 0.01 to 0.02 mg / L at stations 1, 2 and 3 respectively **(Fig. 3A).** The maximum values (0.5 mg / L and 0.6 mg / L) were recorded in TNSR and TOSR at station 2 whereas minimum values (0.012 mg / L and 0.02 mg / L) were recorded in PNSR and POSR at station 3. The higher concentration of ammonia could be partially due to the death and subsequent decomposition of phytoplankton and also due to the excretion of ammonia by planktonic organisms **(Segar and Hariharan, 1989).**

CALCIUM (mg / L)

Figure 3B shows the average values of calcium concentration varied from 346. 67 mg / L to 640 mg / L. The average minimum (346.67mg / L) calcium concentration was recorded in PNSR at station 3 and maximum (640 mg / L) calcium concentration was recorded in TNSR at station 2. Moreover, among the OSR of three different stations, maximum calcium concentration (520 mg / L) was recorded in POSR at station 3 and minimum (413.33 mg / L) was recorded in VOSR at station 1.

CHLORIDE (mg / L)

The maximum and minimum values of chloride were recorded as 24.13 mg / L in VNSR at station 1 and 20.83 mg / L in PNSR at station 3 **(Fig. 3C).**

Similarly, maximum concentration (37.4 mg / L) of chloride was recorded in POSR at station 3 and minimum (25. 87 mg /L) in TOSR at station 2. **Sinha (1986)** recorded that high concentrations of chloride are indicators of large amount of organic matter in the water eutrophic condition. **Sarojini *et al.* (1997)** pointed out that high amount of chloride influences the amount of dissolved oxygen in water.

MAGNESIUM (mg / L)

Magnesium contents of water samples at three different sampling stations were recorded **(Fig. 3D)** and tabulated in **Table 2A** and range was found to be from 1264 mg / L to 1285 mg / L at station 1, 1102.7 mg / L to 1192 mg / L at station 2 and 1120 mg / L to 1128 mg / L at station 3.

DISSOLVED INORGANIC NITRATE (mg / L)

The dissolved inorganic nitrate concentration ranged from 0.17 mg / L to 0.25 mg / L, 0.18 mg / L to 0.19 mg / L and 0.322 mg / L to 0.398 mg / L at stations 1, 2 and 3 respectively **(Fig. 3E).** The maximum values were recorded at station 3 and minimum values were recorded at station 2. The recorded low values at station 2 may be due to its utilization by phytoplankton as evidenced by high photosynthetic activity and also due to the neritic water dominance which contained negligible amount of nitrate **(Govindasamy *et al*., 2000).**

DISSOLVED INORGANIC NITRITE (mg / L)

The dissolved inorganic nitrite concentration ranged from 0.26 mg / L and 0.36 mg / L in station 1, 1.21 mg / L and 1.17 mg /L at station 2 and 0.29 mg / L and 0.57 mg / L at station 3 **(Fig. 4A).** The maximum value was recorded at station 2 and minimum value at station 1. The recorded higher nitrite values could be due to increased pond discharge, oxidation of ammonia and reduction of nitrate and by recycling of nitrogen and also due to bacterial decomposition of planktonic detritus

present in the environment **(Maruthanayagam and Subramanian, 2000).** Further, the denitrification and air-sea interaction exchange of chemicals are also responsible for this increased value **(Chodhury and Penigrahy, 1991).** The recorded low nitrite values may be due to high freshwater inflow and low salinity **(Murugan and Ayyakannu, 1991).** The enrichment of nitrites and nitrates could be attributed to various factors. It is stated that unpolluted waters have nominal quality of nitrates **(Jaji *et al.*, 2007).** However, the enrichment of nutrients viz. nitrates and nitrites are mainly due to sewage outlets rich in protein and poly phosphoric products **(Young – Jin Suh and Rousseaux, 2001).** Also, it enhanced level of these nutrients may be due to mixing of subsurface waters higher in nutrient concentration and terrestrial run off **(Ramachandran *et al.*, 2005).**

TOTAL PHOSPHOROUS (mg / L)

Figure 4B shows total phosphorous concentration ranged from 1.77 mg / L and 2.23 mg / L at station 1, 4.31 mg / L and 4.37 mg/L at station2 and 2mg /L and 2.41 mg / L at station 3 respectively. Total phosphorous level was found high at station 2 and was comparatively low at stations 1 and 3. Elevated level of domestic sewage mixed in water column and fisheries waste, animal and human wastes are also reasons for the higher concentration. Phosphorous level increases due to run off from domestic, municipal and agricultural waste flowing into sea **(Correll, 1998).**

SULPHATE **(mg /L)**

Sulphate content of water samples at three different sampling stations were recorded in **Fig. 4C** and tabulated in **Table 2B.** The sulphate content varied from 1.92 mg / L and 2.41 mg / L at station 1, 1.98 mg / L and 2.03 mg / L at station 2 and 1.99 mg / L and 2.08 mg / L at station 3.

TOTAL ALKALINITY (ppm)

Figure 4D shows the results of the alkalinity measurements ranged from 104.67 ppm and 171 ppm at station 1, 163.33ppm and 165.67 ppm at station 2 and 158.33 ppm and 166.33 ppm at station respectively. The highest alkalinity (166.33 ppm) was recorded in PNSR at station 3 and lowest value (104. 67 ppm) was recorded in VNSR at station 1. On the contrary, the highest alkalinity (171ppm) was recorded in VOSR at station 1 and lowest value (158.33ppm) was recorded in POSR at station 3. According to **Yulfiperinis (2004),** a good alkalinity value for living organisms ranges from 100 – 150 ppm.

SODIUM AND POTASSIUM (ppm)

Figure 4E and 4F show the concentration of sodium and potassium at all the three sites. Maximum concentrations (1636.67ppm and 1403. 33ppm) of sodium and potassium were recorded in VNSR at station 1 and minimum concentration (1380ppm and 1033.33ppm) of sodium and potassium were recorded in PNSR at station 3. Weathering of rocks is very common in sea which results in addition of sodium and potassium in aquatic bodies **(Sharma, 2014).**

DISTRIBUTION OF PHYTOPLANKTON

A list of phytoplankton collected from the study area is presented in **Table 3.** Totally, 44 species of phytoplanktons were recorded during the study period. Among them, 33 species of diatoms (Bacillariophyceae), 2 species of dinoflagellates (Dinophyceae), 1 species of Euglena (Euglenaceae) and 8 species of blue greens (Cyanophyceae) were found **(Plates 2 to 10).** The maximum number of phytoplankton species was recorded in station 2 and 3 **(Table 4).**

In station 2, phytoplankton species was found to be maximum in Bacillariophyceae (23 in TNSR and 11 in TOSR), followed by Cyanophyceae (8 in

TABLE 1: PHYSICAL PROPERTIES OF MARINE WATER COLLECTED FROM THREE SITES

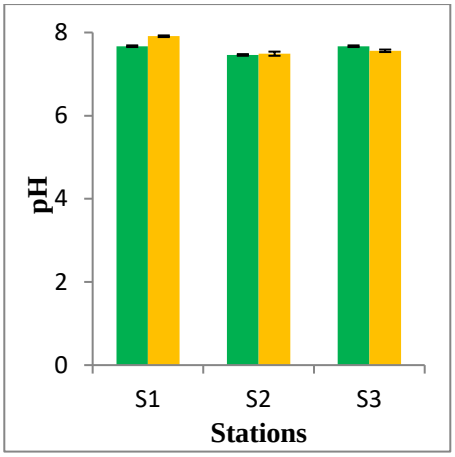
STATION NO.	STATIONS	pH	TEMPERATURE (º C)	SALINITY (ppt)	TDS (mg/L)	HARDNESS (mg/L)
1	VNSR	7.67 ± 0.02	**30.33 ± 0.58**	**32.67 ± 2.52**	652.9 ± 0.93	**6558.14 ± 38.3**
	VOSR	**7.91 ± 0.02**	29.33 ± 0.58	30 ± 1	**597.1 ± 0.76**	6237.2 ± 262.9
2	TNSR	**7.46 ± 0.02**	**28.67 ± 1.15**	**32.67 ± 1.53**	615.1 ± 0.2	6138.86 ± 334.7
	TOSR	7.49 ± 0.05	30.33 ± 0.58	30.33 ± 2.51	622.93 ± 0.3	6173.80 ± 206.1
3	PNSR	7.67 ± 0.02	29.33 ± 1.53	30 ± 2	**697.9 ± 0.4**	**5477.79 ± 316.9**
	POSR	7.56 ± 0.03	30.33 ± 1.53	**31 ± 2.65**	626.8 ± 0.3	5943.54 ± 262.7

TABLE 2A: CHEMICAL PROPERTIES OF MARINE WATER COLLECTED FROM THREE SITES

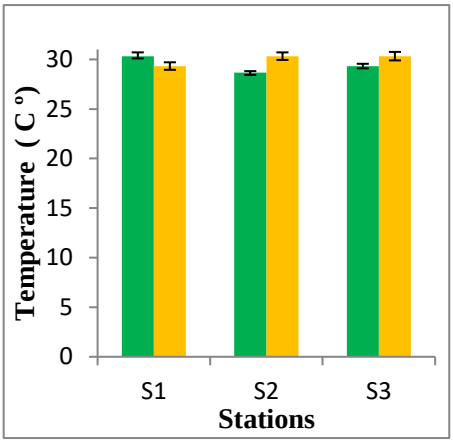
STATION NO.	STATIONS	AMMONIA (mg/L)	CALCIUM (mg/L)	CHLORIDE (mg/L)	MAGNESIUM (mg/L)	NITRATE (mg/L)
1.	VNSR	0.03 ± 0.012	506.67 ± 23.09	**24.13 ± 0.75**	**1285.33 ± 12.22**	0.174 ± 0.02
	VOSR	0.04 ± 0.01	**413.33 ± 59.1**	27.07 ± 0.75	1264 ± 36.67	0.25 ± 0.02
2	TNSR	**0.06 ± 0.012**	**640 ± 51.1**	23.33 ± 1.99	1102.7± 82.01	0.18 ± 0.006
	TOSR	**0.05 ± 0.005**	506.67 ± 46.19	**25.87 ± 1.88**	1192 ± 49.96	0.19 ± 0.007
3.	PNSR	0.012 ± 0.008	**346.67 ± 46.19**	**20.83 ± 0.70**	1120 ± 96.99	0.322 ± 0.016
	POSR	0.02 ± 0.01	**520 ± 69.28**	**37.4 ± 3.41**	1128 ± 48	**0.398 ± 0.02**

TABLE 2B: CHEMICAL PROPERTIES OF MARINE WATER COLLECTED FROM THREE SITES

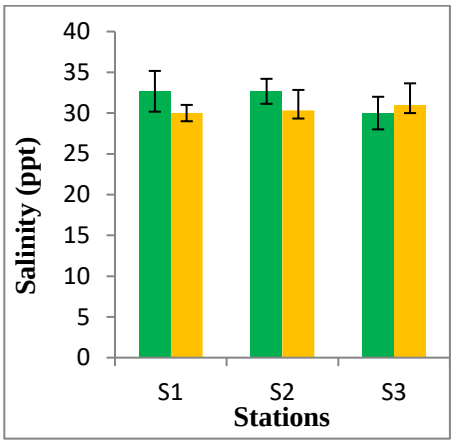
STATION NO.	STATIONS	NITRITE (mg/L)	TOTAL PHOSPHORUS (mg/L)	SULPHATE (mg/L)	TOTAL ALKALINITY (ppm)	POTASSIUM (ppm)	SODIUM (ppm)
S1	VNSR	**0.26 ± 0.02**	2.23 ± 0.16	**2.41 ± 0.15**	**104.67 ± 77.67**	**1403.33 ± 11.55**	**1636.67 ± 30.56**
	VOSR	0.36 ± 0.10	**1.77 ± 0.17**	1.92 ± 0.12	171.33 ± 4.16	1343.33 ± 25.17	1586.67 ± 35.12
S2	TNSR	**1.21 ± 0.04**	4.31 ± 0.11	2.03 ± 0.07	165.67 ± 1.53	1373.33 ± 45.09	1613.33 ± 25.17
	TOSR	1.17 ± 0.03	**4.37 ± 0.11**	1.98 ± 0.09	163.33 ± 1.15	1250 ± 40	1493.33 ± 30.56
S3	PNSR	0.29 ± 0.05	2 ± 0.16	**2.08 ± 0.014**	**166.33 ± 2.08**	**1033.33 ± 15.28**	**1380 ± 10**
	POSR	0.57 ± 0.11	2.41 ± 0.15	1.99 ± 0.103	158.33 ± 6.03	1273.33 ± 35.12	1523.33 ± 30.55



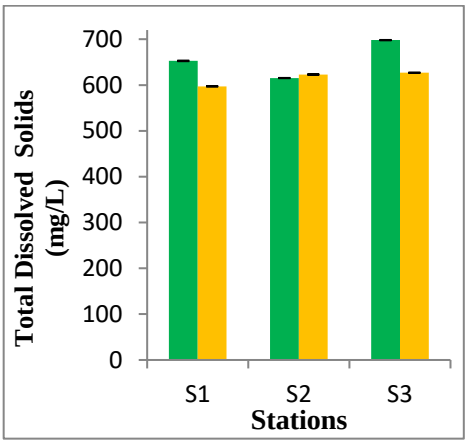
A) pH



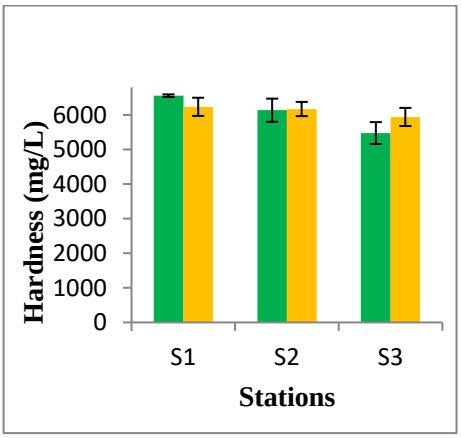
B) Temperature (Cº)



C) Salinity (mg/L)



D) Total Dissolved Solids (mg/L)

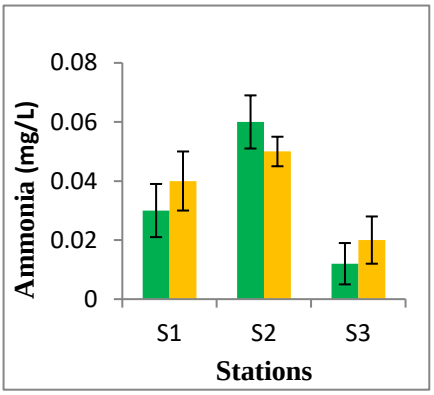


E) Hardness (mg/L)

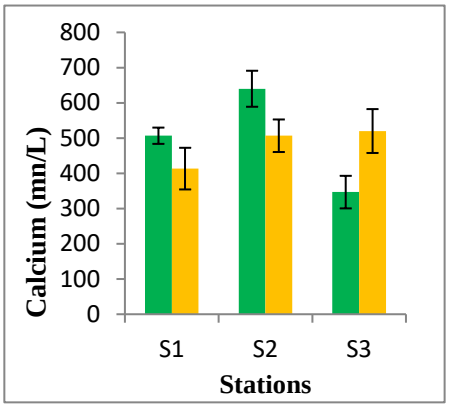
S1 - Vembar, S2 – Therespuram, S3 – Punnakkayal

NSR – Near Shore Region OSR – Off Shore Region

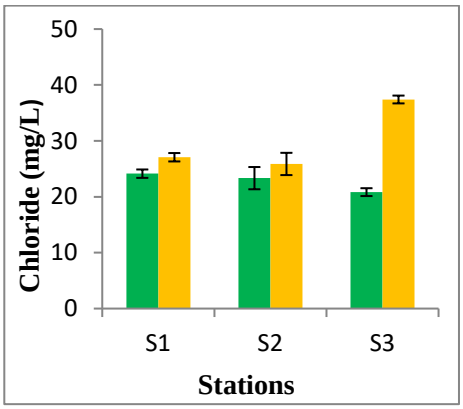
FIGURE 2: PHYSICAL PROPERTIES OF SEAWATER COLLECTED FROM THREE DIFFERENT STATIONS OF THOOTHUKUDI COAST



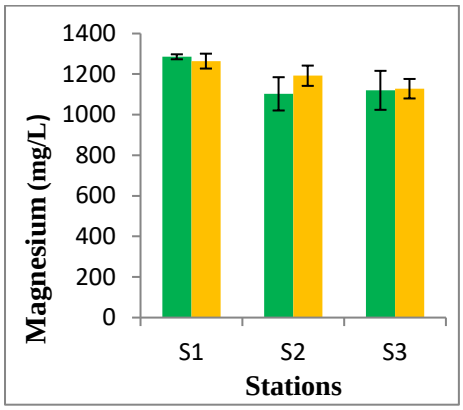
A) Ammonia (mg/L)



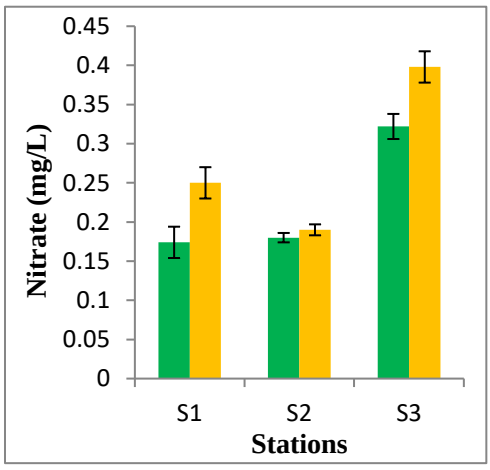
B) Calcium (mg/L)



C) Chloride (mg/L)



D) Magnesium (mg/L)

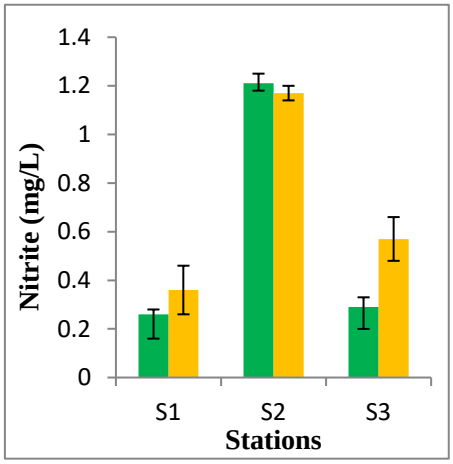


E) Dissolved Inorganic Nitrate (mg/L)

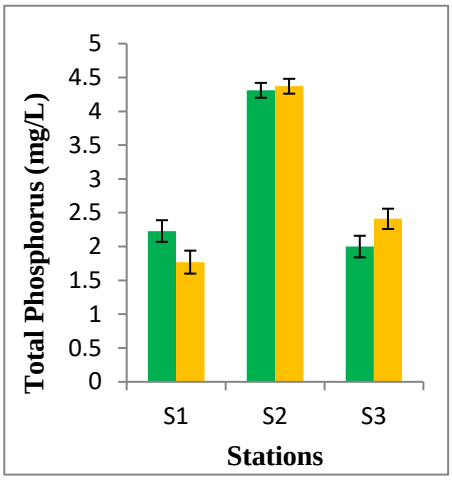
S1 - Vembar, S2 – Therespuram, S3 – Punnakkayal

NSR – Near Shore Region OSR – Off Shore Region

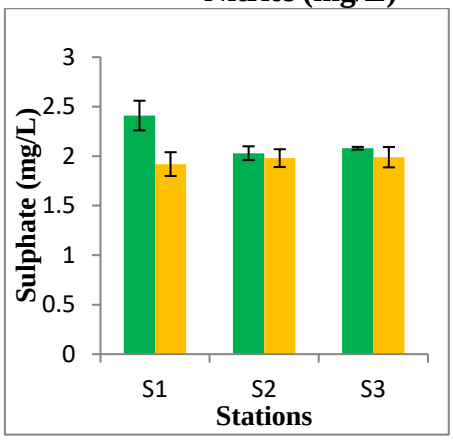
FIGURE 3: CHEMICAL PROPERTIES OF SEAWATER COLLECTED FROM THREE DIFFERENT STATIONS OF THOOTHUKUDI COAST



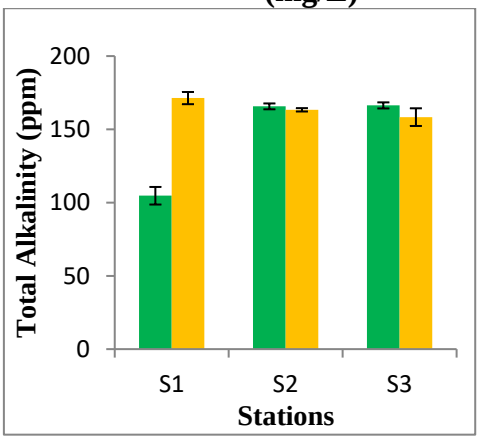
A) Dissolved Inorganic Nitrite (mg/L)



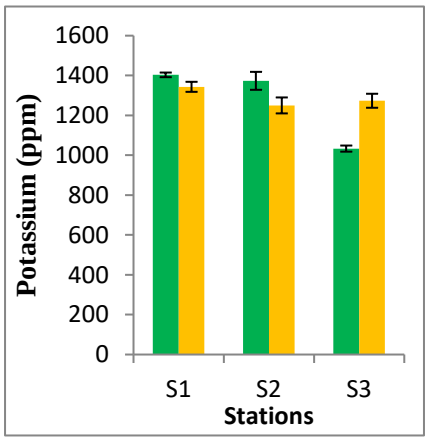
B) Total Phosphorus (mg/L)



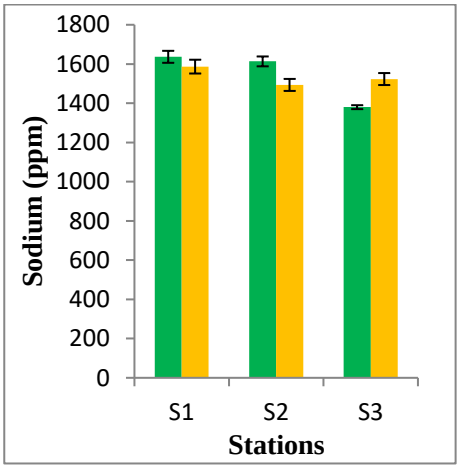
C) Sulphate (mg/L)



D) Total Alkalinity (ppm)



E) Potassium (ppm)



F) Sodium (ppm)

S1 - Vembar, S2 – Therespuram, S3 – Punnakkayal

NSR – Near Shore Region **OSR – Off Shore Region.**

FIGURE 4: CHEMICAL PROPERTIES OF SEAWATER COLLECTED FROM THREE DIFFERENT STATIONS OF THOOTHUKUDI COAST

TNSR and 3 in TOSR), Dinophyceae (2 in TNSR and 1 in TOSR) and Euglenophyceaae (1 in TNSR). In station 3, phytoplankton species was recorded maximum in Bacillariophyceae (25 in PNSR and 11 in POSR) followed by Cyanophyceae (7 in PNSR and 2 in POSR). The result was similar with others research indicating that Bacillariophyceae as the dominant genera on water sample **(Xia *et al.*, 2014; Brazin *et al.*, 2014).** There was no record of Dinophyceaen member in station 3 and 1.

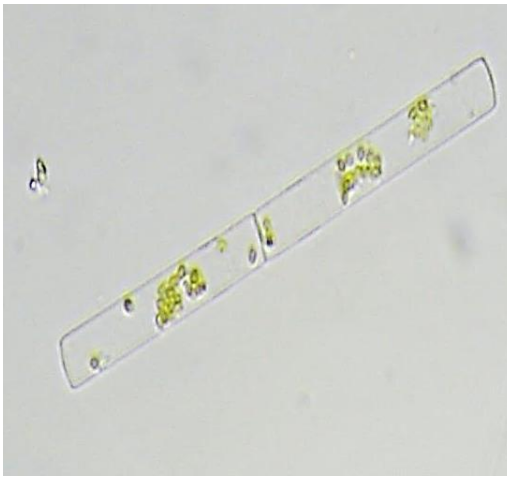
Among various species observed in the study, species of *Gyrosigma, Nitzschia, Closterium, Navicula, Diploneis, Cylindrotheca, Cymbella, Amphora, Achnanthes, Amphiprora, Euglena, Synechococcus, Oscillatoria* were found in all the three stations. *Leptoylindrus danicus, Liamophora abbreviata*, species of *Chaetoceros, Asterionella* and *Rhizosolenia* were found only in station 1 whereas, Species of *Cocconeis* and *Synedra* were found only in station 3. Similarly, species of *Chaetoceros, Tryblionella, Prorocentrum* and *Chlorococcus* were found only in station 2.

The research explained that diatoms diversity is sensitive to a wide range of environmental variable and that their community structure may quickly respond to changing physical, chemical and biological condition in the environment **(Mooser *et al.*, 1996).** Phytoplankton abundance and distribution was varied depending on water depth. **Table 3** showed that phytoplankton abundance and distribution was comparatively lower in OSR in all the 3 stations than in NSR of Southern Bay of Bengal. Difference in phytoplankton abundance and distribution might be varied because of difference in light and nutrients availability.

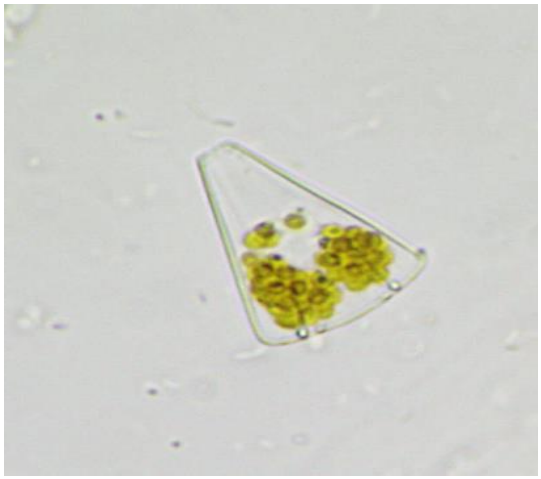
The percentage occurrence of different phytoplankton groups with respect to total phytoplankton taxa in 2 different regions of three stations of Thoothukudi

coast is presented in **Fig. 5.** From this study, it was concluded that, the OSR of three stations had only Bacillariophyceae members among the total phytoplankton and other members were rarely found. Though the percentage occurrence of Bacillariophyceae members were maximum in NSR of three stations among the total phytoplankton but less percentage occurrence in NSR than OSR of three stations of Thoothukudi coast. This may be due to the occurrence of diversity of other members in NSR than OSR of three stations of Thoothukudi coast.

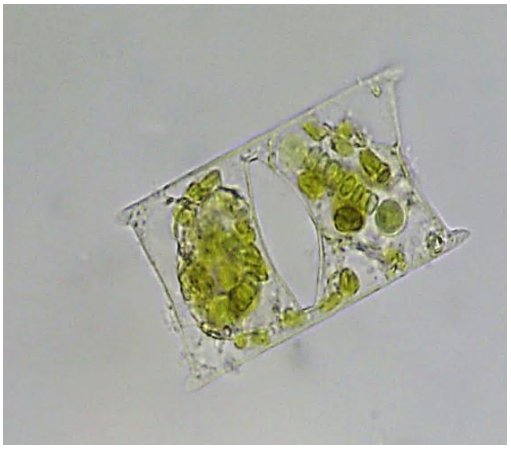
PLATE 2: PHOTOGRAPHS OF PHYTOPLANTON SPECIES BELONGS TO BACILLARIOPHYCEAE



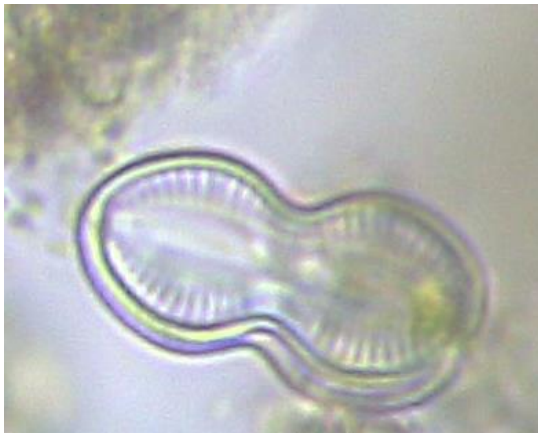
Leptoylindrus danicus



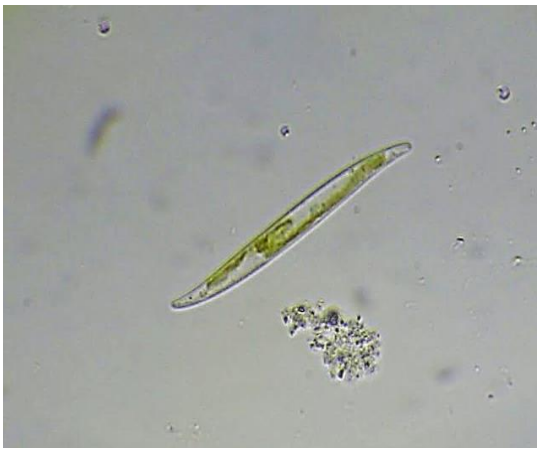
Licmophora abbreviata Agardh



Hemiaulus membranaceus Cleve

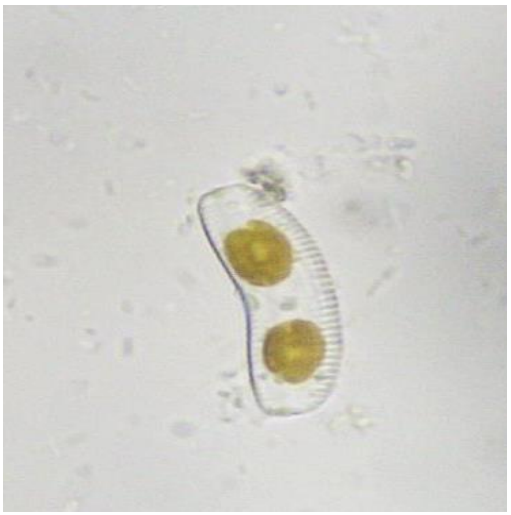


Diploneis weissflogii

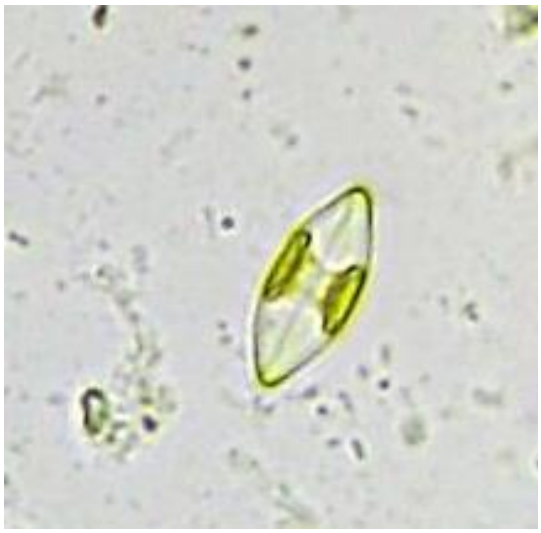


Gyrosigma spp.

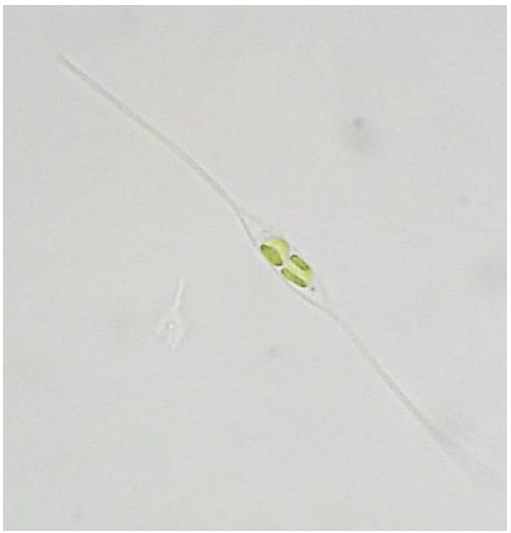
PLATE 3: PHOTOGRAPHS OF PHYTOPLANTON SPECIES BELONGS TO BACILLARIOPHYCEAE



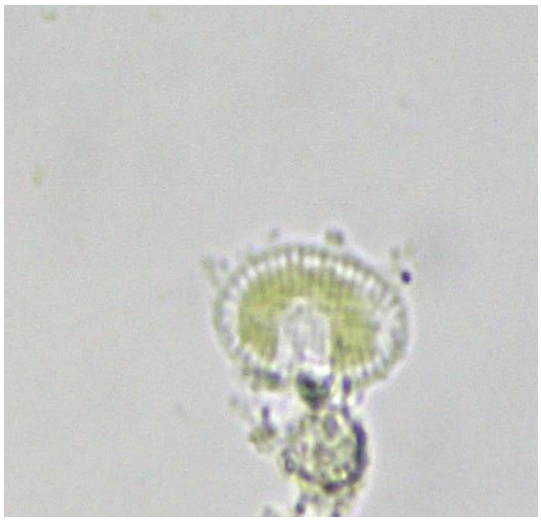
Nitzschia spp.



Nitzschia spp.



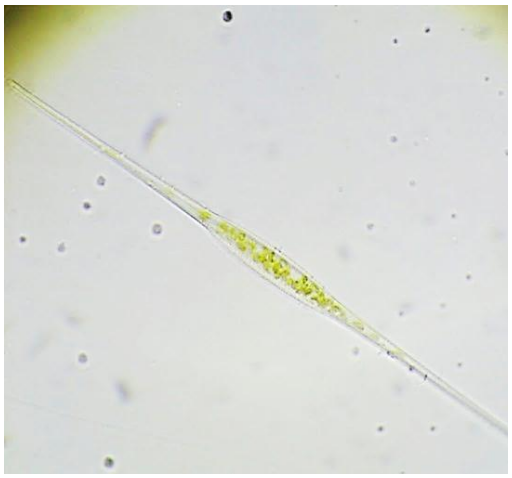
Nitzschia longissima



Cocconeis spp.

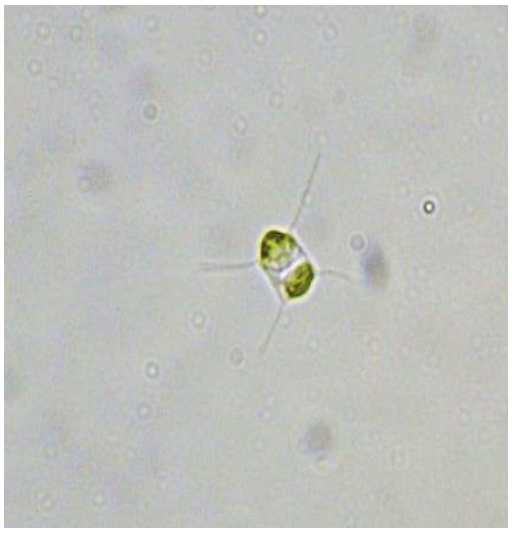
Chaetoceros spp.

PLATE 4: PHOTOGRAPHS OF PHYTOPLANTON SPECIES BELONGS TO BACILLARIOPHYCEAE

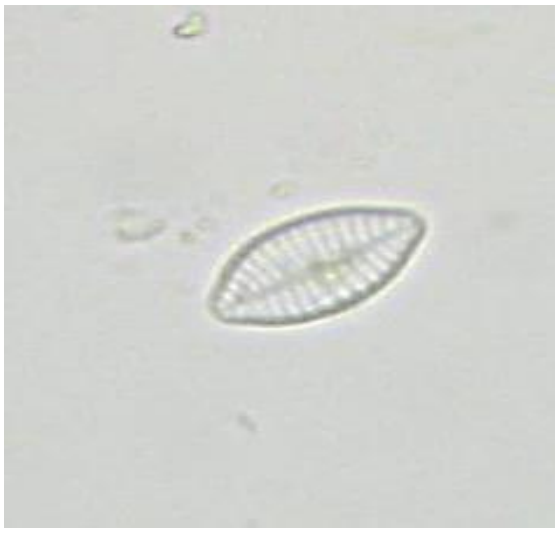


Closterium spp

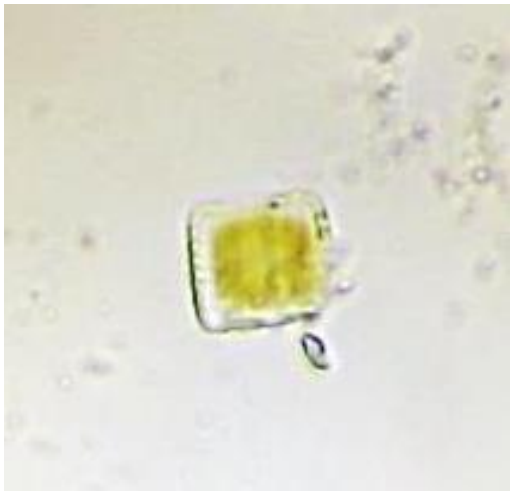
Navicula spp.



Chaetoceros spp.

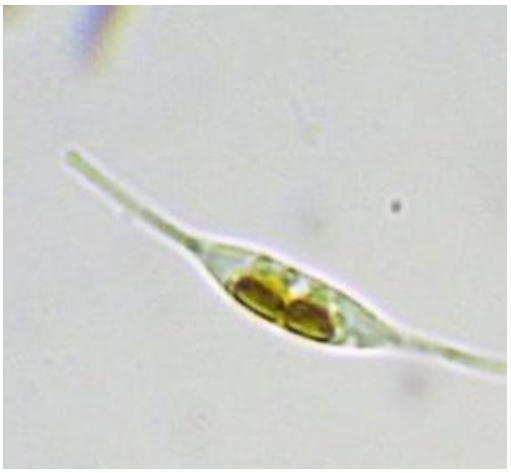


Diploneis spp.

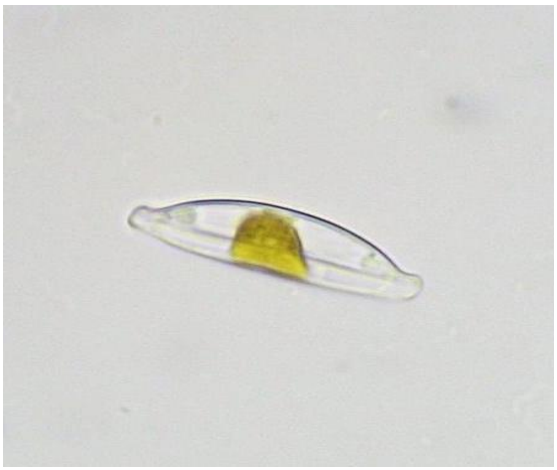


Striatella spp.

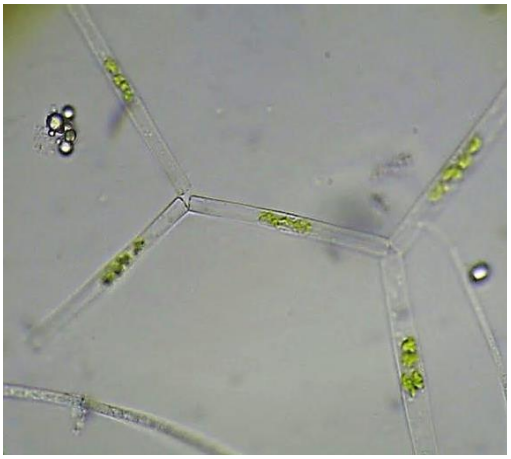
PLATE 5: PHOTOGRAPHS OF PHYTOPLANTON SPECIES BELONGS TO BACILLARIOPHYCEAE



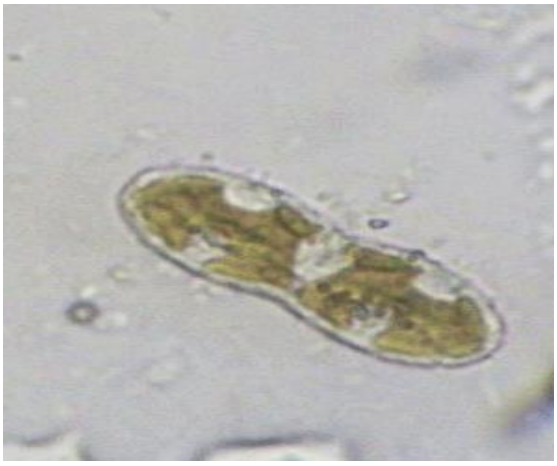
Cylindrotheca closterium



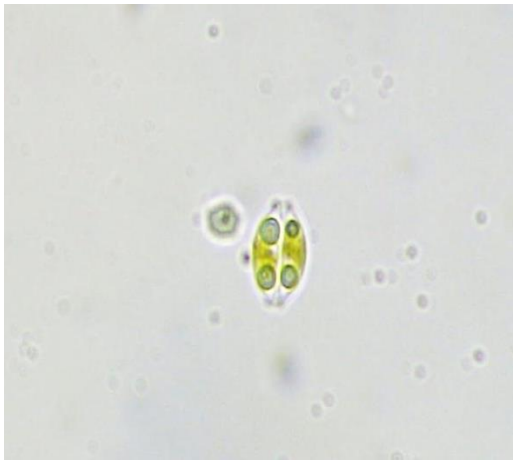
Cymbella spp.



Asterionella spp.

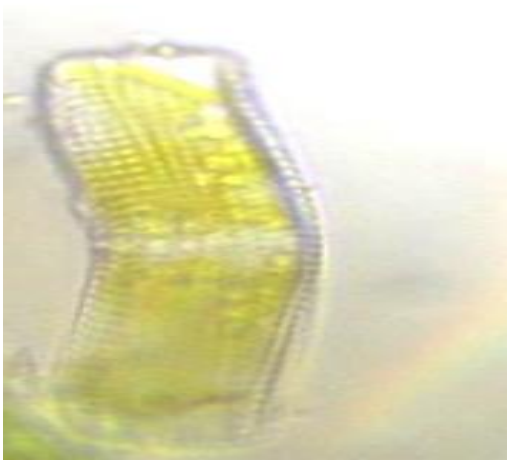


Amphiphora spp.

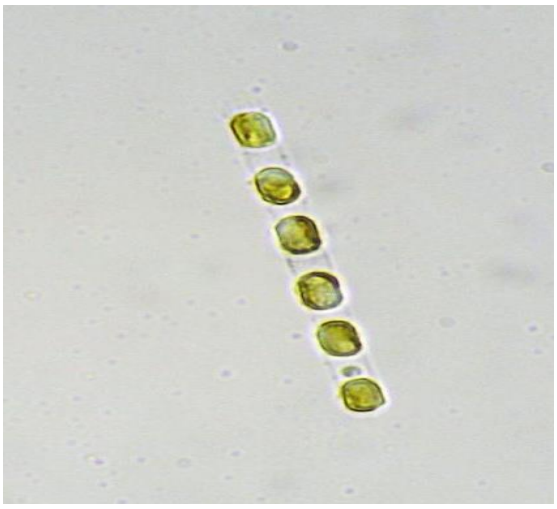


Amphora ovalis

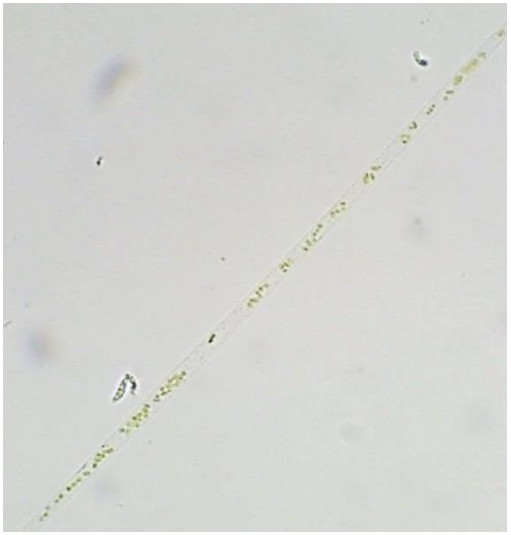
PLATE 6: PHOTOGRAPHS OF PHYTOPLANTON SPECIES BELONGS TO BACILLARIOPHYCEAE



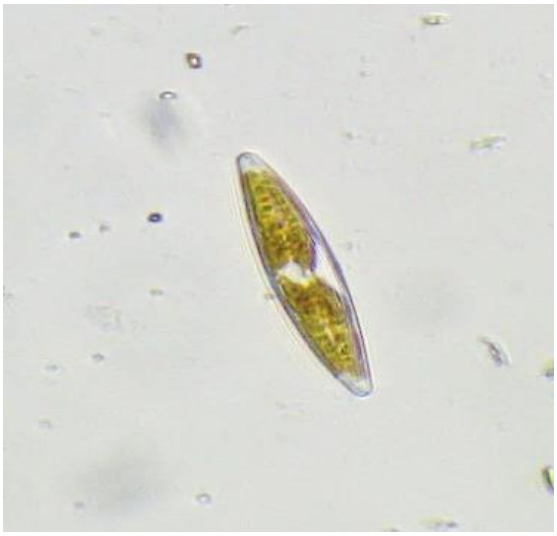
Achnanthes spp.



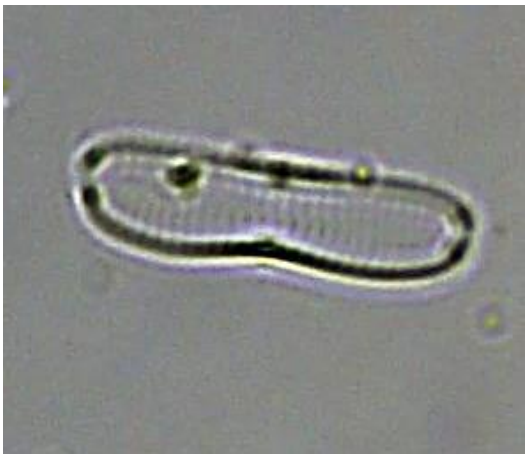
Skeletonema spp.



Rhizosolenia spp.

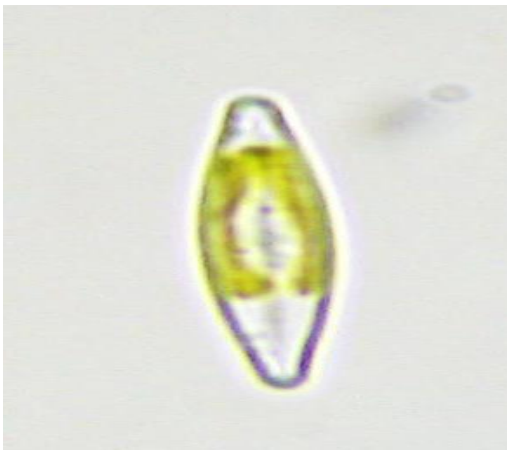


Navicula spp.

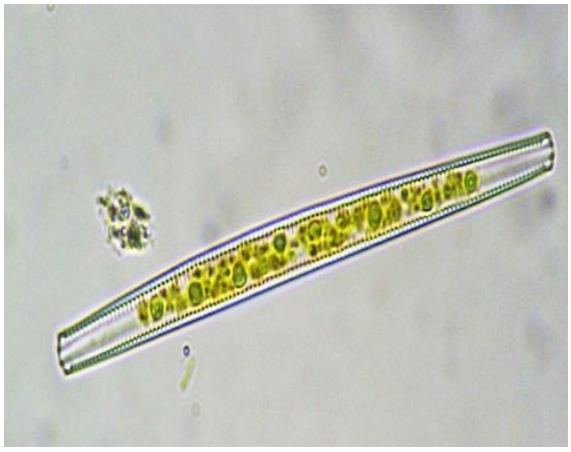


Amphiprora spp

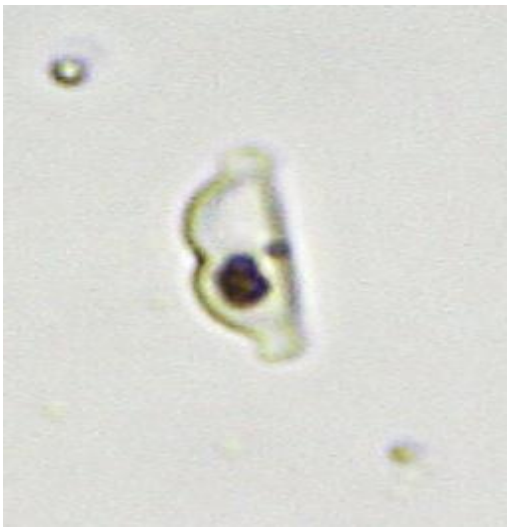
PLATE 7: PHOTOGRAPHS OF PHYTOPLANTON SPECIES BELONGS TO BACILLARIOPHYCEAE



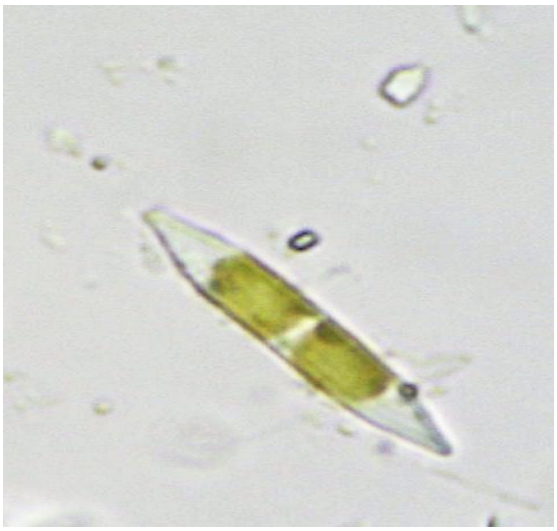
Navicula spp.



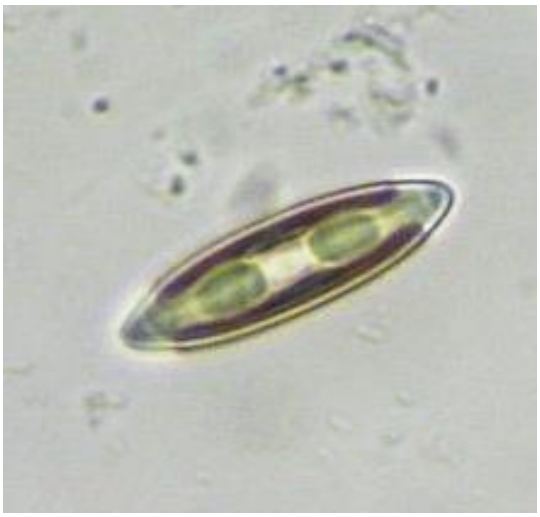
Synedra spp.



Amphora bigibba Grunow

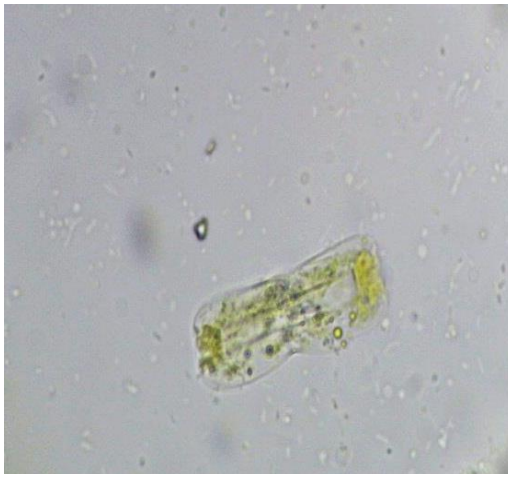


Tryblionella spp.

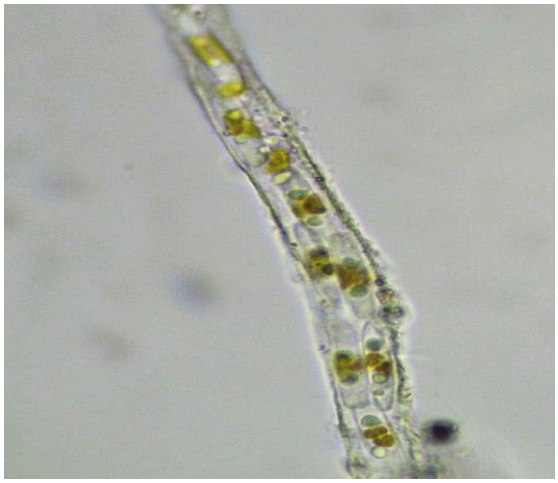


Navicula spp.

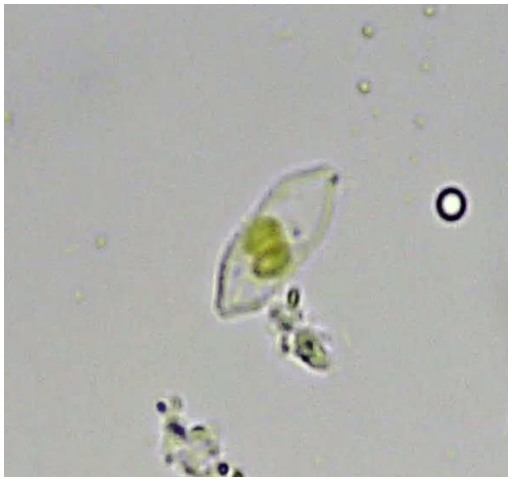
PLATE 8: PHOTOGRAPHS OF PHYTOPLANTON SPECIES BELONGS TO BACILLARIOPHYCEAE AND DINOPHYCEAE



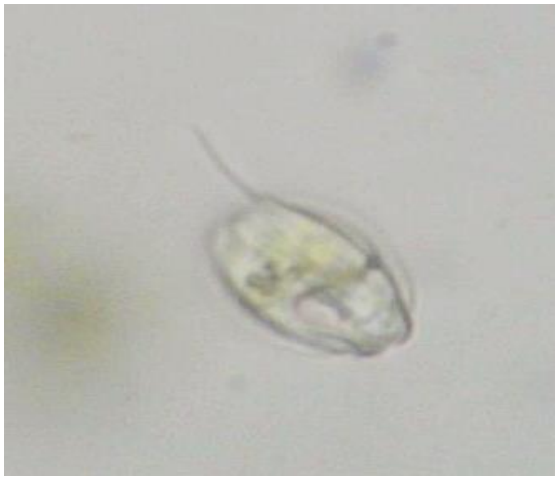
Amphiphora spp



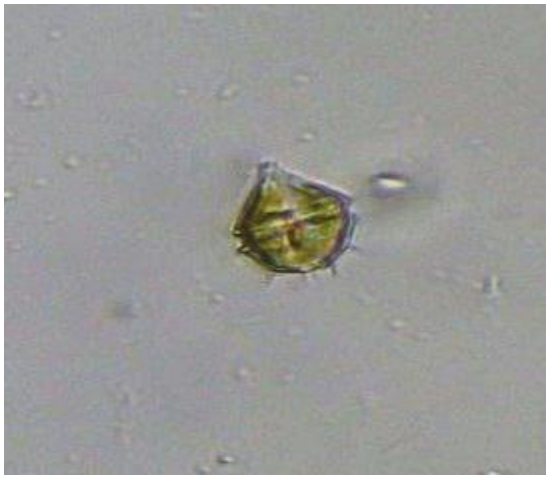
Pseudo-nitzschia



Lyrella spp.

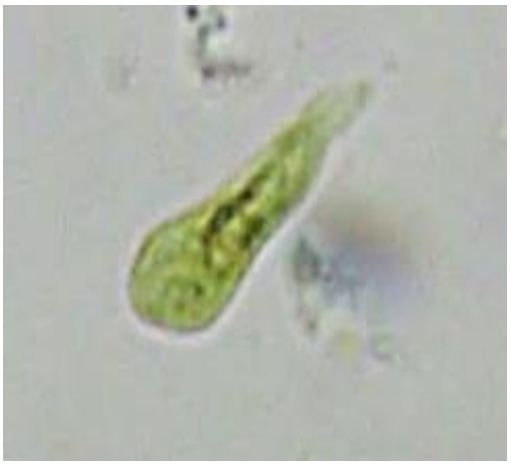


Prorocentrum spp.

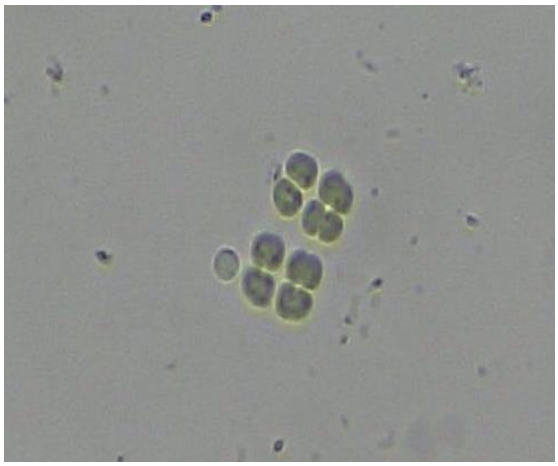


Photoperidinium quinquecorne

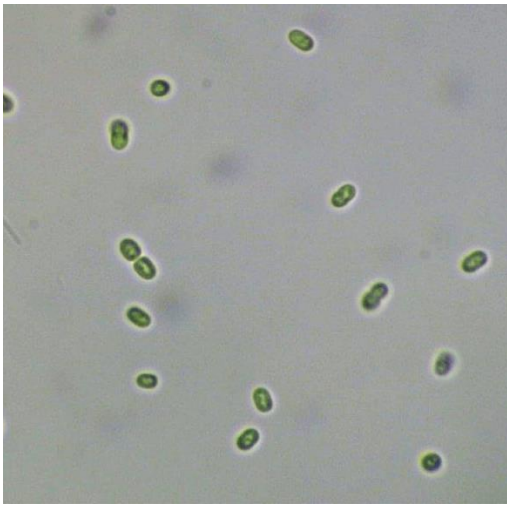
PLATE 9: PHOTOGRAPHS OF PHYTOPLANTON SPECIES BELONGS TO EUGLENACEAE AND CYANOPHYCEAE



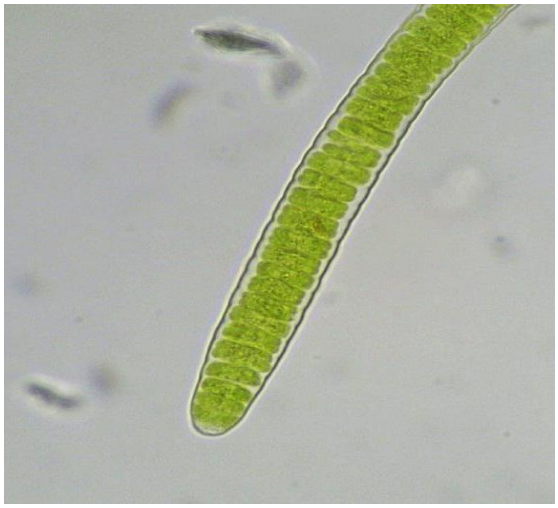
Euglena spp.



Chroococcus spp.



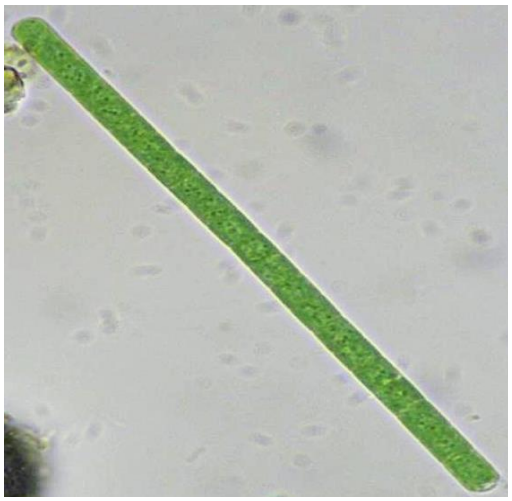
Synechococcus spp.



Plectonema spp.

Oscillatoria princeps

PLATE 10: PHOTOGRAPHS OF PHYTOPLANTON SPECIES BELONGS TO CYANOPHYCEAE



Oscillatoria spp.

Oscillatoria spp.

Phormidium spp.

Oscillatoria spp.

TABLE 3: OCCURRENCE OF MICROALGAE IN THREE DIFFERENT STATIONS OF THOOTHUKUDI COAST

S. NO.	CLASS	NAME OF THE TAXA	STATION I		STATION II		STATION III	
			VNSR	VOSR	TNSR	TOSR	PNSR	POSR
01.	Bacillariophyceae	*Leptoylindrus danicus*	+	+	-	-	-	-
02.		*Licmophora abbreviata* Agardh	-	-	+	-	+	+
03.		*Hemiaulus membranaceus* Cleve	+	+	-	-	-	-
04.		*Diploneis weissflogii*	-	-	+	-	+	+
05.		*Gyrosigma spp.*	+	-	+	-	+	+
06.		*Nitzschia spp.*	+	-	-	-	+	-
07.		*Nitzschia spp.*	+	+	+	+	+	+
08.		*Nitzschia longissima*	+	-	+	+	+	-
09.		*Cocconeis spp.*	-	-	-	-	+	+
10.		*Chaetoceros spp.*	+	+	-	-	-	-
11		*Closterium spp*	+	+	+	-	+	-

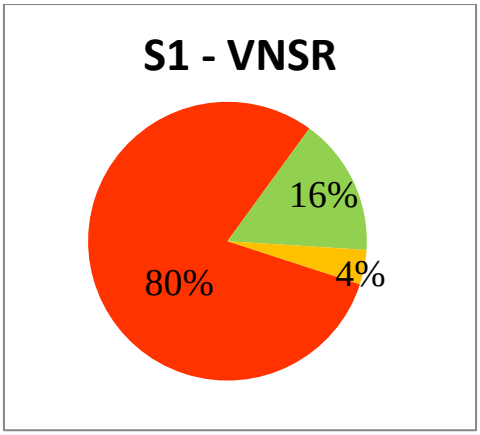
S. NO.	CLASS	NAME OF THE TAXA	STATION I		STATION II		STATION III	
			VNSR	VOSR	TNSR	TOSR	PNSR	POSR
12	Bacillariophyceae	*Navicula spp.*	+	+	+	+	+	+
13		*Chaetoceros spp.*	-	-	+	-	-	-
14		*Diploneis spp.*	+	-	+	+	+	-
15		*Striatella spp.*	-	-	+	-	+	-
16		*Cylindrotheca closterium*	+	-	+	-	+	+
17		*Cymbella spp.*	+	-	+	+	+	+
18		*Asterionella spp.*	+	+	-	-	-	-
19		*Amphiphora spp.*	+	-	-	-	+	+
20		*Amphora ovalis*	+	-	+	+	+	-
21		*Achnanthes* spp.	+	-	+	-	+	-
22		*Skeletonema spp.*	-	+	-	+	-	-

S. NO.	CLASS	NAME OF THE TAXA	STATION I		STATION II		STATION III	
			VNSR	VOSR	TNSR	TOSR	PNSR	POSR
23	Bacillariophyceae	*Rhizosolenia spp.*	-	+	-	-	-	-
24		*Navicula spp.*	+	-	+	+	+	+
25		*Amphiprora spp*	+	-	+	+	+	-
26		*Navicula spp.*	+	+	+	+	+	+
27		*Synedra spp.*	-	-	-	-	+	-
28		*Amphora bigibba* Grunow	-	-	+	-	+	-
29		*Tryblionella spp.*	-	-	+	-	-	-
30		*Navicula spp.*	-	-	+	+	+	+
31.		*Amphiphora spp*	+	-	+	-	+	-
32		*Pseudo-nitzschia*	-	-	+	-	+	-
33.		*Lyrella spp.*	-	-	+	-	+	-

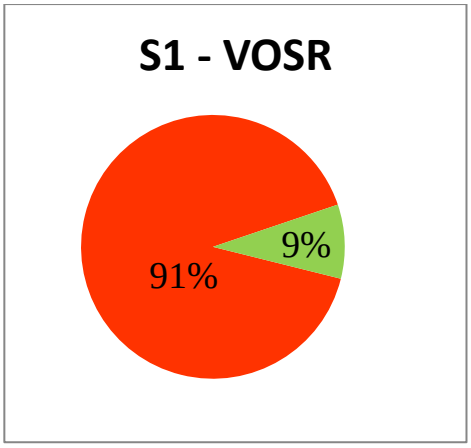
S. No.	Class	Name of the Taxa	Station I		Station II		Station III	
			VNSR	VOSR	TNSR	TOSR	PNSR	POSR
34	Dinophyceae	*Prorocentrum spp.*	-	-	+	-	-	-
35		*Photoperidinium quinquecorne*	-	-	+	+	-	-
36	Euglenaceae	*Euglena spp.*	+	-	+	-	+	-
37	Cyanophyceae	*Chroococcus spp.*	-	-	+	-	-	-
38		Synechococcus spp.	+	-	+	+	+	-
39		*Plectonema spp.*	-	-	+	-	+	-
40		*Oscillatoria princeps*	+	-	+	-	+	-
41		*Oscillatoria spp.*	+	+	+	+	+	+
42		*Oscillatoria spp.*	+	-	+	-	+	+
43		*Phormidium spp.*	-	-	+	-	+	-
44		*Oscillatoria spp.*	-	-	+	+	-	-

TABLE 4: NUMBER OF MICROALGAE SHOWING CLASSWISE DISTRIBUTION

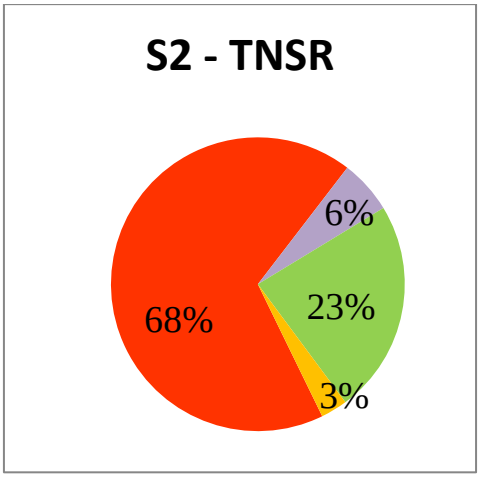
S. NO.	STATIONS	BACILLARIOPHYCEAE	DINOPHYCEAAE	CYANOPHYCEAE	EUGLENACEAE	TOTAL
1	VNSR	20	-	4	1	**25**
	VOSR	10	-	1	-	**11**
2	TNSR	23	2	8	1	**34**
	TOSR	11	1	3	-	**15**
3	PNSR	25	-	7	1	**33**
	POSR	11	-	2	-	**13**



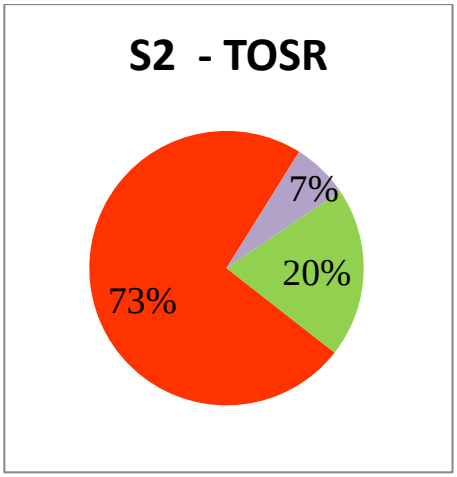
Vembar Near Shore Region



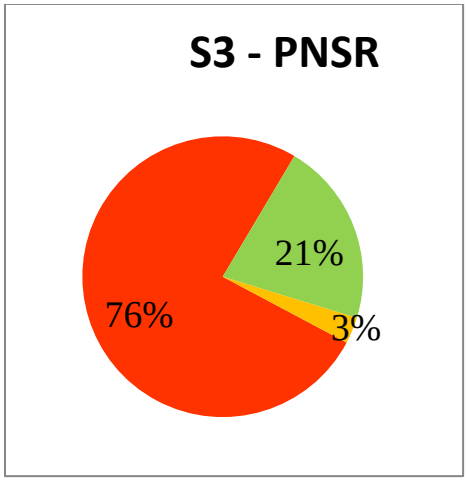
Vembar Off Shore Region



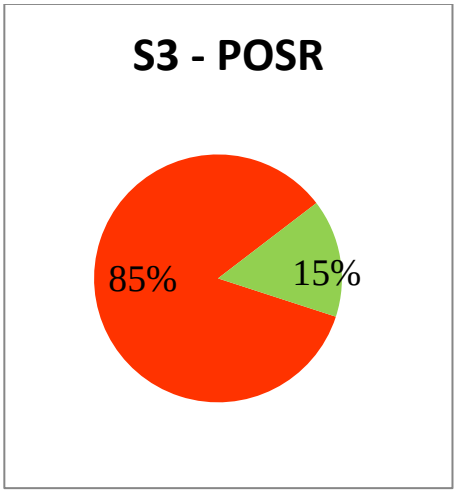
Therespuram Near Shore Region



Therespuram Off Shore Region



Punnakkayal Near Shore Region



Punnakkayal Off Shore Region

Bacillariophyceae, Dinophyceae, Cyanophyceae, Euglenaceae

FIG 5: PERCENTAGE COMPOSITION OF DIFFERENT PLANKTONS CLASSES AT SAMPLING STATION

CONCLUSION

CONCLUSION

In the present study, the physico-chemical factors and phytoplankton community were surveyed. The results of this study contribute with essential information on phytoplankton composition and abundance, their correlation with environmental parameters and environmental characteristics in Thoothukudi coast. These data might be used as criteria for further research in water quality parameters of coastal areas. Besides that, continuous monitoring of other water quality like nutrients and hydrodynamics profiling necessary to achieve for better management of healthy ecosystem in coastal area. It is recommended that the proper maintenance of the water bodies is necessary. Proper sanitation measures and environmental education to public care are essential to keep these water bodies clean and safe. A few efforts like diversion of sewage, presentation of leaching nutrients from catchment area through plantation would definitely yield healthy hygienic and sustainable environment.

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EVALUATION OF PHYTOCHEMICAL, ANTIBACTERIAL ACTIVITY AND SYNTHESIS OF NANOPARTICLES OF PHYSALIS ANGULATA R.Br.

A dissertation submitted to

St. Mary's College (Autonomous)
(Re-Accredited with A^+ Grade by NAAC)

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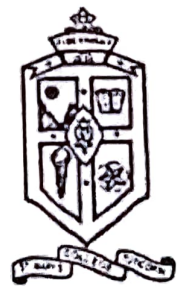
in partial fulfilment of the requirements for the Degree of

Master of Science in Botany.

By R. SANKARA ESWARI

Reg No: 20APBO08



DEPARTMENT OF BOTANY

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2022-2023

DECLARATION

I do here by declare that this dissertation entitled **Evaluation of phytochemical, antibacterial activity and synthesis of nanoparticles *of* *Physalis angulata* R.Br** by me in partial fulfilment for the award of the degree of **Master of Science in Botany**, is the result of my original and independent work carried out under the guidance of **Dr. A.Jacintha Tamil Malar**. Assistant Professor of Botany, St. Mary's College (Autonomous), Thoothukudi and it has not been submitted elsewhere for the award of any degree.

Station: Thoothukudi

Date: 12.04.23

R. Sankara Eswari

(R. SANKARA ESWARI)

CERTIFICATE

This is to certify that this dissertation entitled, **Evaluation of phytochemical, antibacterial activity and synthesis of sliver nanoparticles of *Physalis angulata* submitted by R.SANKARA ESWARI Reg.No.20APBO08,** to ST. MARY'S COLLEGE (Autonomous), THOOTHUKUDI in partial fulfilment for the award of the degree of **"Master of Science in Botany"** is done by her under my supervision. It is further certified that this dissertation or any part of this has not been submitted elsewhere for any other degree.

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CONTENT

INTRODUCTION

INTRODUCTION

. The scientific, technological and social development processes and correspondingly the historical development of the concept of health and disease have led to the emergence of mystic, polypharmacy, aetiology, traditional and modern medical practices, respectively. It is frequently encountered to the traces of the early stages of the historical development process of medical practice today. Especially alternative medicines (natural or traditional medicine) and its practices have caused the many different stages of this developmental period to be conveyed to today's societies.

It is estimated that there is the number of between 750,000 and 1,000,000 plant species on the world. The 500,000 of them have been identified and named. Around 2000 new flowering plant species are identified and named in each year. The number of plants which have been used for treatment since ancient times shows a steady increase. According to a report released by the World Health Organization (WHO), the number of plants used for treatment is estimated to be around 20,000. The studies on medicinal plants and active substances derived from them have increased the interest in these plants in recent years. Plant products are generally preferred and used by patients who have chronic medical conditions including cancers (2%), liver diseases (21%), HIV (22%), asthma (24%) and rheumatologic disorders (26%). Many believe that natural treatment methods are harmless. Whereas, many

recent scientific studies have pointed out the serious consequences of the side effects of herbal products such as giving other damages to patients who seek solutions to problems such as obesity.

Ten percent of all vascular plants are used as medicinal and there are estimated to be between 350,000 and almost half a million species of them. Since ancient times, plants have been used in medicine and are still used today. In the beginning, the trial and error method was used to treat illnesses or even simply to feel better, and in this way, to distinguish useful plants with beneficial effects. The use of these plants has been gradually refined over the generations, and this has become known in many contexts as traditional medicine. The official definition of traditional medicine can be considered as "the sum total of the knowledge, skills and practices based on the theories, beliefs and experiences indigenous to different cultures, whether explicable or not, used in the maintenance of health, as well as in the prevention, diagnosis, improvement or treatment of physical and mental illnesses".

It is a fact that all civilizations have developed this form of medicine based on the plants in their own habitat. There are even authors who claim that this transmitted knowledge is the origin of medicine and pharmacy. Even today, hundreds of higher plants are cultivated worldwide to obtain useful substances in medicine and pharmacy. The therapeutic properties of plants gave rise to medicinal drugs made from certain plants with these benefits

The origin of modern science, especially in the Renaissance, in particular chemical analysis, and the associated instrumentation such as the microscope, was what made it possible to isolate the active principles of medical plants. Since then, these active principles have been obtained synthetically in the laboratory to produce

the medicines later. The use of medicines was gradually expanded. Until today, the direct use of medicinal plants is apparently displaced in modern medicine.

The emphasis on the use of medicinal plants had hitherto been placed on the treatment rather than prevention of diseases. However, there exists in the literature considerable report in recent times on research work on the use of medicinal plants and their constituents in disease prevention. A World Health Organization (WHO) Expert Group defined Traditional Medicine as the sum total of all knowledge and practices, whether explicable or not, used in diagnosis, prevention and elimination of physical, mental, or social imbalance and relying exclusively on experience and observation handed down from generation to generation, whether verbally or in writing (WHO, 1976).

Throughout human history, medicinal plants have always been used as medicine to treat various diseases. Almost 80% who live in developed countries are said to be depended on the practice of traditional medicine A report from the World Health Organization (WHO) comes out with a percentage of 80% of the global population tend to rely on traditional medicines. Most of the therapies use extracts and active compounds of the medicinal plant. Currently, there is a rise in medicinal plant consumption in the world, due to the proven effectiveness of medicinal plants, in curing certain diseases and claims that shows it is safe to be used. Medicinal plants play a major role in medication since the beginning of human civilization and also contribute to the manufacturing of drugs these days.

Phytotherapy which is one of the alternative medicine practices has become basic subjects of many scientific studies about treatment with plants. Alternative medicine, defined in a variety of terms (complementary, supplemental, and non-traditional) includes treatment practices which are not found in traditional medicine.

Traditional medicine is spreading rapidly in all regions of the developing world and in industrialized countries. Phytotherapy in today and the developments in the chemistry and biochemistry sciences during nineteenth and twentieth century gave a great impetus to the pharmaceutical industry, and thus many medicines were developed to respond to the needs of medicine in the laboratories by adopting the principles of efficiency, harmlessness and quality, as a result of analytical, toxicological, pharmacological and clinical studies. The 1/2 of the current medicines are herbal-based, and the active substance to be obtained from the plant in most of these is copied in the laboratory. Due to many factors such as medical and economic problems stemming from serious side effects of synthetic drugs in recent years, ecological approaches and acting which increase environmental pollution and whose creators also include the international pharmaceutical industry and threat posed by many chronic diseases that are not yet curable and the idea that naturalness is always effective and free of side effects, herbal treatment have become popular again.

A good knowledge of the chemical composition of plants leads to a better understanding of its possible medicinal value. Secondary plant metabolites are numerous chemical compounds produced by the plant cell through metabolic pathways derived from the primary metabolic pathways. Secondary metabolites have shown to possess various biological effects, which provide the scientific base for the use of herbs in the traditional medicine in many ancient communities. They have been described as antibiotic, antifungal and antiviral and therefore are able to protect plants from pathogens. metabolites have shown to possess various biological effects, which provide the scientific base for the use of herbs in the traditional medicine in many ancient communities. They have been described as antibiotic, antifungal and antiviral and therefore are able to protect plants from pathogens.

Today's medicine needs the industry producing pharmaceutical medicines, which are largely based on the active principles of plants, and therefore, these are used as raw materials in many cases. Yet, today, the underdeveloped world does not have access to this modern medicine of synthetic origin, and therefore, large areas of the world continue to use traditional medicine based on the direct use of medicinal plants due to their low cost.

The genus *Physalis* of family Solanaceae is among the largest genera in subfamily Solanoideae, with about 100 species, although the estimation of the species within the genus varies considerably – from 75 to 120. The genus is generally recognized by the inflated balloon or lantern-like calyx (husk, fruit basket), which completely envelopes the berry, protecting it against insects, birds, diseases and adverse climatic conditions. Most of *Physalis* species have a long history of ethnomedical use in the treatment of various ailments, including malaria, asthma, hepatitis, liver and kidney problems, dermatitis, and many others, and as immunomodulatory, antitumor, antibacterial or antipyretic agent.

The name *Physalis* is from the Greek, meaning 'bladder', and refers to the inflated calyx. The genus is a member of the Nightshade family, Solanaceae (Mahalakshmi & Nidavani1 2014). *Physalis* species are annuals or perennials, erect or decumbent, sometimes rhizomatous, glabrous or pubescent, and with variously toothed or lobed leaves. The genus *Physalis* is consisting of about 90-100 species (Mahklouf 2016; Sultana et al. 2008). It is thought that it originated in Mexico and presently, it is mainly distributed in tropical, south, and temperate America, although some species have a world-wide distribution. The genus *Physalis* contains many species grown for their ornamental or edible fruits that are eaten raw or cooked.

The last two decades have witnessed a constantly increasing interest in *Physalis* species, with a specific focus on their phytochemistry and pharmacology. As it is well known, medicinal plants are the largest reservoir of secondary metabolites, and the major sources of chemical diversity that has driven many pharmaceutical breakthroughs in the last century (Sang-ngern et al. 2016).

Based on these background information, the present study was justifiably designed with the following objectives:

- With the above background, the present study has undertaken comprehensive approaches to
- Identify the presence of various principal phytochemical
- Explore and compare antimicrobial, properties in various solvent extracts of the leaf and shoot.

REVIEW OF LITERATURE

REVIEW OF LITURATURE

Plants-derived medicines have been the part of traditional health care in most parts of the world for thousands of years and there is an increasing interest in plants as sources of agents to microbial diseases (Chariandy *et al.*, 1999). Natural products discovered from medicinal plants and derivatives have provided numerous clinically used medicines (Balunas and Kinghorn, 2005). Plants are considerably useful and economically essential. They contain active constituents that are used in the treatment of many human diseases. Plants are rich sources of secondary metabolites, which are potential remedies for different ailments. Extreme interest in plants with microbial activity has revived as result of current problems such as resistance associated with the use of antibiotics obtained from microorganisms (Nagendra *et al.*, 2010).

Medicinal plants are the "backbone" of traditional medicine, which means more than 3.3 billion people in the less developed countries utilize medicinal plants on a regular basis (Davidson-Hunt, 2000). It is the nature's gift to human beings to make disease free healthy life. India is one of the most medico-culturally diverse countries in the world, where the medicinal plant sector is a part of time-honored tradition that is respected even today. Here, the main traditional systems of medicine include Ayurveda, Unani and Siddha (Kotnis *et al.*, 2004). Traditional folk remedies from plants have always guided scientists to search for new medications in order to maintain and promote healthy life for human and animals (Achterberg, 2013).

Physalis angulata is used in many parts of the world to treat several diseases, such as anticancer, antibacterial, for diabetes, treatment of malaria, anemia and reducing fever. In Peru, it has been documented that native groups in the Peruvian Amazon, use the decoction of leaves and fruit for Tertian due to postpartum infections (Jovel *et al.*, 1996)

Mestizo populations use it for treatment of diabetes, by taking a glass of macerated root combined with honey, twice daily for 60 days. The root infusion is taken for hepatitis; the leaf infusion is used as a diuretic, for asthma, malaria, inflammation and as a disinfectant, the unripe fruit is used to treat scabies (Mejía and Rengifo, 2000). In Bolivia, the indigenous community of Tacana, the root decoction is used to treat fever (Bourdy *et al.*, 2000). In Kenya, the infusion of the whole plant is used for worms and stomach pain (Geissler *et al.*, 2002).

In the state of Paraíba, people use the leaves infusion as a sedative and against inflammations of bladder, spleen and kidney; in the case of inflammations a tea is taken until symptoms disappear and as sedative it is drunk at night (Agra *et al.*, 2007). In Indonesia use the root decoction as a remedy for postpartum, muscle aches and hepatitis (Roosita *et al.*, 2008). In India, leaf paste is used as an external application for wounds (Sudhakar *et al.*, 2009). In Maruda community, state of Para, root is taken as a tea for hepatitis symptoms, anemia, urine infection, stomachache, prostate and kidney stones (Coelho-Ferreira, 2009). *P. alkekengi* can be effective on immunity system, cancer, thyroid hormones, liver enzymes, and sexual and reproductive hormones (Ge *et al.*, 2009)

The leaves are also used for asthma, dermatitis, diuretic, earache, fever, gonorrhea, hemorrhage, hepatitis, infections, inflammation, liver disorders, malaria, postpartum infection, rheumatism, skin diseases, to prevent abortion, worms (schistosomiasis). (Lawal *et al.*, 2010). The berries, the calyces or the whole plants of *P. peruviana are* an integral part of folk medicine traditions in many countries. In Peruvian and Columbian medicine fruit is used empirically to treat cancer, hepatitis, asthma, malaria, dermatitis, rheumatism to reduce blood

glucose; to decrease albumin; to control cataract, pterygium and amebiasis (Puente *et al.*, 2011)

In many countries across Europe and South Asia folk medicines recommend the use of *P. alkekengi* 8 diuretic for renal and urinary tract ailments, and in the treatment of gout and rheumatism (Sharma *et al.*, 2015). Kusumaningtyas *et al.*, (2015) recorded that the aerial parts extracts of *P. angulate* have been used as traditional medicine for the treatment of diseases such as malaria, asthma, hepatitis, dermatitis, rheumatism, liver disorders, fever, bronchitis and others.

Namjoyan *et al.* (2015) stated that *P. alkekengi* is used in traditional medicine include anti-inflammatory, antibacterial, antiseptic, analgesic, laxative, diuretic, antimitotic, hypoglycemic, antispasmodic, liver corrective and sedative, as well as relief of malaria and syphilis symptoms. Leaves and dried seeds are used as curing agents for skin diseases, jaundice, ulcer, fever, glaucoma, abdominal upsets; as antiseptics, diuretics and antibiotics (Anjalam *et al.*, 2016).

In traditional, Chinese medicine *P. alkekengi* fruit, calyces, roots and whole plants have been used (internally or externally) for a variety of conditions, such as sore throat, cough, eczema, hepatitis, urinary problems, and tumors (Shu *et al.*, 2016). In Mexican and Ecuadorian folk medicine the fruit of *P. ixocarpa* is used as eyewash, tonic, diuretic and laxative, as treatment for gastrointestinal and respiratory problems, and as an application in inflammations, enlargement of the spleen, ascites and bladder ulceration. Crushed leaves are applied over snakebites (Khan *et al.*, 2016).

Most of *Physalis* species have a long history of ethnomedical use in the treatment of various ailments, including malaria, asthma, hepatitis, liver and kidney

problems, dermatitis, and many others, and as immune modulatory, antitumor, antibacterial or antipyretic agents (Zhang and Tong, 2016).

The discovery and development of the antibiotic, penicillin during the 1900s gave a certain hope to medical science, but this antibiotic soon became ineffective against most of the susceptible bacteria. The antibiotic resistance in bacteria is generally a natural phenomenon for adaptation to antimicrobial agents. Once bacteria become resistant to some antibiotic, they pass on this characteristic to their progeny through horizontal or vertical transfer. The indiscriminate and irrational use of antibiotics these days has led to the evolution of new resistant strains of bacteria that are somewhat more lethal compared to the parent strain. Cases The indiscriminate and irrational use of antibiotics these days has led to the evolution of new resistant strains of bacteria that are somewhat more lethal compared to the parent strain. Cases of widespread occurrence of resistant bacteria are now very common which leads to many health-related problems (Iwu *et al.*, 1999).

Infectious disease caused by bacteria, viruses, fungi and parasites are still a major threat to public health, despite the tremendous progress in human medicine. The past three decades have seen a dramatic increase in microbial resistance to antimicrobial agents. Such situation stimulates the development of new anti-microbial agents in order to treat the infectious disease in an effective manner. So this matter continued to an era to identify the potential antimicrobial agent from the natural resources. The edible plants that used for traditional medicine contain a wide range of substance that can be used to treat abundant of infectious disease with reduced side effects (Subramanion *et al.*, 2010).

Plants protect from disease and damage and contribute to the plant's colour, aroma and flavour. In general, the plant chemicals that protect plant cells from

environmental hazards such as pollution, stress, drought, UV exposure and pathogenic attack are called as phytochemicals (Gibson *et al.*, 1998; Mathai, 2000). Hasler and Blumberg (1999) stated that the phytochemicals are biologically active, naturally occurring chemical compounds found in plants, which provide health benefits for humans further than those attributed to macronutrients and micro nutrients.

The withanolides are a group of naturally occurring steroids built on an ergostane skeleton, in which C-22 and C-26 are appropriately oxidized in order to form a lactone ring. These compounds are specific for the Solanaceae family, and, in particular, for the genera *Withania, Acnistus, Dunalia, Physalis, Datura, Lycium* and *Jaborosa*. So far, notable activities were reported for withanolides, including anticancer, anticonvulsive, immunosuppressive, and antioxidant properties (Glotter, 1991). A flavonol glycoside, myricetin 3-O-neohesperidoside was isolated from cytotoxic MeOH extract of leaves *P. angulata* (Ismail and Alam, 2001).

Carotenoids from fruit of *P. angulata* were determined by HPLC-PDA-MS/MS and 22 compounds have been identified. all-trans-carotene was major carotenoid, contributing 62.2% to total carotenoid, followed by 9-cis-carotene and alltrans-cryptoxanthin, contributing around 2.9 and 2.7%, respectively (De Rosso and Mercadante, 2007). *P. alkekengi* is effective on immunity system, cancer, thyroid hormones, liver enzymes, and sexual and reproductive hormones. Phytochemical

investigations indicate that this plant contains alkaloids, glucocorticoids, physalis, lycopene, ethanolic compounds, and vitamin C. (Helvaci *et al.*, 2010).

Physalins are the steroidal lactone constituents from *Physalis* and other closely related genera, belonging to the family Solanaceae. The physalins are biogenetically related to the withanolides (Chen *et al.*, 2011). Jin *et al.*, (2012) reported the discovery of three antiproliferative withanolides with an unusual carbon framework.

namely, physangulidines A, B and C, isolates from *P. angulata* L. using bioassay-directed isolation technique.

P. angulata extracts are found to be rich in polyphenols (gallic acid, ellagic acid, caffeic acid, rutin, mangiferin), and to have antioxidant, antipyretic, antimicrobial, anti-inflammatory and analgesic effects (Meira *et al.*, 2015). Like the other *Physalis species*, *P. alkekengi* accumulates various specialized metabolites and nutrients (alkaloids, vitamins, flavonoids, phenolic acids, saponins, tannins, withanolides, carotenoids, glucocorticoids, etc.), responsible for its activities and use (Bahmani *et al.*, 2016).

The total phenol and flavonoid fractions from the calyces of *P. pubescensare* reported to have antioxidant, anti-proliferation and induced-apoptotic activity and to be potential candidates for the development of antihepatoma ingredients, in contrast to those extracted from *P. pubescens* fruit (Wang *et al.*, 2016). Rivera *et al.* (2018) studied the presence of many other biologically active constituents in phytochemical analysis, e.g. flavonoids, carotenoids, alkaloids (phygrine), diterpene glycosides, as well as that of physalins and other withanolides in *P. angulate.*

Li *et al.* (2018) stated that the majority of pharmacological functions are associated with the presence of physalins, flavonoids and phenylpropanoids, in a synergy with other chemical constituents. Physalins in particular are responsible for anti-inflammatory, antimicrobial, anti-diabetic, anti-cancer (the anti-tumor effects of physalins being the hot topic in the pharmacological aspects of the plant) and immune suppressive activities; flavonoids for anti-diabetic, anti-inflammatory and anti-cancer activities; phenylpropanoids for anti-diabetic, antimicrobial and anti-cancer effect.

P. peruvianais is considered as one of the promising members of the superfruit family. Typically, the term superfruit has been introduced with the marketing strategy to promote the health benefits of exotic fruits with less popularity worldwide, which have numerous phytochemicals (such as phenolic acids, flavonoids, proanthocyanidins, coumarins, hydrolysable tannins, carotenoids, and anthocyanins) together with the corresponding antioxidant activities (Chang *et al.*, 2019).

Pietro *et al.* (2000) examined natural antimicrobial agents form plant extracts through bioassay-guide fractionation, by in vitro determination of minimum inhibitory concentration (MIC) using the micro dilution method with Alamar blue oxidation-reduction dye, demonstrated that crude CHCl3 extracts from aerial parts (500 g/ml) of *P. angulata* caused total growth inhibition of *Mycobacterium tuberculosis* H37 Rv cells. Shim *et al.* (2002) isolated and identified oleanolic acid from *P. angulata*, showing antibacterial activity against oral pathogens. The ethanolic extract of the flowers of *P. angulata*, exhibit noticeable antibacterial activity against *Streptococcus* mutans causing dental caries at all concentration tested.

The bactericidal test has showed that the methanol extract of *P. angulata* conferred fast killing effect against *S. mutans* in 2 min at 50 mg/ml concentration (Hwang *et al.*, 2004). Silva *et al.* (2005) conducted a comparative study on physalin B and enriched physalin fractions (mixture of B, D, F and G physalins) at three different concentrations using the agar diffusion technique against pathogenic gram positive and gram negative microorganisms. The demonstration is at 200 g/ml concentration which is pure physalin B exhibited at 85% of the inhibitory zone observed with the pool of physalins, at same concentration.

Lopes *et al.* (2006) analyzed the antimicrobial activity of different extracts of the fruit and root of *P. angulata*, using the agar diffusion method against *Staphylococcus aureus* ATCC 6538, of which the ethanol extract of the fruit showed better bacterial activity when compared to ampicillin, which was prepared in water at concentrations ranging from 0.25 to 4090 g/ml. The extract showed an inhibition of 11.17 mm, matching the range of linearity of curve of the antibiotic, which ranged from 9.60 to 20.30 mm. 12

Osho *et al.* (2010) examined antimicrobial properties of aerial part and the roots extract oil against *Bacillus subtilis* (MIC50 = 4.0 mg/ml) and *Klebsiella pneumoniae* (MIC50 = 4.0 mg/ml); moreover it has antifungial activity with *C. albicans* (MIC50 = 4.0 mg/ml), *C. stellatoidea* (MIC50 = 3.75 mg/ml) and *C. torulopsis* (MIC50 = 4.0 mg/ml), which are resistant to many antibiotics.

The ethanolic extract of the fruit of *P. angulata* has shown antibacterial activity against *Staphylococcus aureus* at all concentration used (100 mg g^{-1} , 125 mg g^{-1} and 150 mg g^{-1}) with inhibition zones between 34.5 mm and 50.5 mm as compared to standard antibiotic (chloramphenicol) which gave a mean zone inhibition diameter of 79.8 mm at 100 mg g^{-1} concentration (Donkor *et al.*, 2012).

Durga *et al.* (2020) evaluated the phytochemical substances and bioactive compound of the aqueous methanol extract of unripe fruit of *Physalis minima* Linn. The crude extract was separated by soxhlet using methanol as solvent followed by the phytochemical screening to identify the presence of secondary metabolites. Volatile components present in the methanol extract were separated and identified using GC/MS. The Gas chromatography - mass spectrometry chromatogram result showed the presence of total of 18 bioactive compounds in crude extract of *Physalis minima* Linn which are exhibiting different biological functions such as antioxidant, anti-

inflammation, anticancer, antidiuretics etc. Antibacterial activity of *Physalis minima* Linn extracts were also studied using disc diffusion method. The activities of methanol extracts were tested and had showed good inhibition zone against *Staphylococcus aureus*.

Patel *et al.*, (2011) found out the antibacterial potential of mature berries of *P. minima* using streak plate, well diffusion, determination of minimum inhibitory concentration and bioautographic methods against a battery of Gram positive and Gram negative bacterial strains. Results of the study showed that methanol and chloroform extracts of *P. minima* exhibited potent inhibitory activity against all the bacterial strains tested. Minimum inhibitory concentration found out was 100 µg in both the extracts.

Osho *et al.*, (2010) investigated of *Argemone Mexicana L.* as an agent against *Bacillus subtilis, Klebsiella pneumoniae, Staphylococcus aureus, Pseudomonas aeruginosa, Candida albicans, Candida stellatoidea and Candida torulopsis,* using well diffusion and minimum inhibitory concentrations methods. The sensitivity of *Bacillus Subtilis, Klebsiella pneumoniae, Pseudomonas aeruginosa and Staphylococcus aureus* to the essential oils of both the aerial and root parts were determined. *Pseudomonas aeruginosa* was resistant to the essential oil from both the aerial and root part of the plant. *C. torulopsis, C. stellatoidea and C. albicans* were susceptible to the essential oils from the aerial and root part of the plant. The minimum inhibitory concentrations ranging between 3.75mg/ml and 4.0mg/ml were recorded for *Bacillus subtilis, Klebsiella pneumoniae* by the aerial and the root extracts, but *P. aeruginosa* and *S. aureus* were not susceptible to the aerial and root extracts.

Litopenaeus vannamei shrimp is one of the leading export commodities in fisheries from Indonesia, however White Feces Diseases (WFD) cause deaths up to 30% and decrease their growth and production. WFD is caused by the accumulation of several *Vibrio* bacteria in shrimp water and intestines. One of the local herbs that has the potential as antiproliferation, antidiabetic, anticytotoxic commonly used in the treatment of humans diseases is *Physalis angulata* known as ciplukan. The aim of this study was to obtain ciplukan extract which has an antibacterial activity of *Vibrio* sp., and to analyze the anti-bacterial effect of ciplukan extract on *Vibrio* bacteria from vannamei infected with WFD (Saraswathi and Wijaya 2018).

Donkor *et al.*, (2012) investigated the inhibitory activity of zinc oxide-ointment formulation as well as the unformulated crude extract of fruits of *Physalis angulata* against clinical wound isolates of *Staphylococcus aureus* and *Pseudomonas aeruginosa*. The zinc oxide-ointment formulation and the unformulated *P. angulata* crude extract were found to be ineffective against *P. aeruginosa* at all concentrations used, but potent against *S. aureus* at varying degrees. The zinc oxide-ointment (100 mg g-1 , 125 mg g-1 and 150 mg g-1) and *P. angulata* crude extract/zinc oxide-ointment (100 mg g-1 , 125 mg g1 and 150 mg g-1) formulations were only slightly active against *S. aureus* at the highest concentration of 150 mg g-1 . The unformulated *P. angulata* crude extract alone exhibited the highest inhibitory activity against *S. aureus* at all concentrations used with zones of inhibition between 34.5 mm and 50.5 mm, followed by a formulation of the extract with only oleaginous base (ointment), with zones of inhibition between 12.8 mm and 20.3 mm.

Goztok and Zengin (2015) evaluated the antimicrobial activity was according to the microdilution method by using *Bacillus megaterium* DMS 32, *Pseudomonas aeruginosa* DMS 50071, *Escherichia coli* ATCC 25922, *Klebsiella pneumoniae* FMC

5, *Proteus vulgaris* FMC 1, *Enterobacter aeregenes* CCM 2531, *Candida albicans* FMC 17, *Candida globrata* ATCC 66032, *Candida tropicalis* ATCC 13803, *Trichophyton* sp. and *Epidermaphyton* sp., the fruit juice extract of *P. peruviana* inhibited the growth of microorganisms used in the test at different ration. MIC values of fruit juice was determined as 3.125- 25 µl.

Ethanol extract of *Physalis peruviana* and water extract of *Hyphaene thebaica* were tested for their inhibitory effects on eight bacterial strains (*Escherichia coli, Proteus mirabilis, Salmonella typhimurium, Salmonella entrica , Shigella dysenteriae, Bacillus cereus, Bacillus licheniformis and Staphylococcus aureus*) by using well diffusion technique. The study demonstrated that *Staph. aureus* was the most sensitive gram-positive strain and *E. coli* was the most sensitive gram-negative strain to ethanol extract of *Physalis peruvianae* and water extract of *Hyphaene thebaica*. The inhibition zone diameters near to the synthetic antibiotic ciprofloxacin (Abd-ELmageed *et al.*, 2019).

Kumar *et al.* (2017) discussed the establishment of green route for the rapid synthesis of silver nanoparticles (AgNPs) using aqueous extract of *Physalis angulata* (AEP) leaves which act as a reducing as well as a capping agent. The change in color from watery to dark brown confirmed the synthesis of AgNPs. A characteristic surface plasmon resonance (SPR) band at 436 nm advocated the presence of AgNPs. The synthesis process was optimized using one factor at a time approach where 2.0 mmol L^{-1} $AgNO_3$ concentration, 4.5% (v/v) of AEP inoculum dose and 10 min of sunlight exposure were found to be the optimum conditions. The synthesized AgNPs was characterized by several characterizing techniques such as Transmission Electron Microscopy (TEM), Selected Area Electron Diffraction (SAED), Scanning Electron Microscopy (SEM), Energy Dispersive X-Ray (EDX), X-

Ray Diffraction (XRD), Fourier Transform Infrared Spectroscopy (FTIR) and Atomic Force Microscopy (AFM) analysis. The synthesized AgNPs showed strong antibacterial activity against both Gram-negative and Gram-positive bacteria as well as antioxidant activity.

Gold nanoparticles have come to prominence among noble metals due to their characteristics. Bio-transformed nanoparticles have a low toxicity and are biocompatible and ecologically benign. *Physalis minima* has been used in traditional medicine for its diuretic, laxative, and anti-inflammatory qualities since ancient time. In a UV–visible spectrophotometer, the resulting Au NPs formed a broad surface peak at 546 nm. According to the FTIR data, the peaks represent flavonoids, phenols and polyphenols meaning that flavonoids, phenols, and polyphenols were utilised as the reducing agent to create gold nanoparticles (Sekhar *et al.*, 2022).

Gold nanoparticles (Au NPs) show wound healing properties. In addition, ethnobotanical information from Siaya County in Kenya shows the leaves of *Physalis peruviana* L. to be effective in wound management. A combination of Au NPs and leaf extracts of *Physalis peruvian* [18] ـh a one pot biogenic synthesis leads to a new effective wound management substance. The synthesis was done at room temperature 25° C and at 85°C. The UV-visible spectroscopy results show efficient sharper plasmon bands with a blue shift indicating a decrease in λ max compared to red shift which show an increase in λ max. The surface plasmon resonance is a sharper at wavelength of about 540 nm. The gold nanoparticles synthesized from *Physalis peruviana* showed antimicrobial activities against gram-positive bacteria and, gram-negative bacteria as well as gram-positive fungus. The inhibition zones for Au NPs of different concentrations vary significantly between concentrations. The highest antibacterial activity is at 100 mM of Au NPs against *Escherichia coli*,

Bacillus subtilis, and *Staphylococcus aureus*. The inhibition zones for Au NPs at concentration of 100 mM and *Physalis peruviana* extract vary significantly in all the microbial cells, except for *Pseudomonas aeruginosa* (Odongo *et al.*, 2022).

MATERIALS AND METHODS

MATERIALS AND METHODS

Collection and identification of plant material source of plant material:

The fresh twigs of *Physalis angulata* belongs to the family Solanaceae are collected from Pudur Pandya Puram in Thoothukudi district in Tamil Nadu, during December 2022 to January 2023. After collection the stem and the leaves were separated and the stem were cut into small pieces. The fresh leaf and stem of the collected plant were washed three times with sterile water and dried at room temperature. Then grinded to powder after chopping into small pieces.

Preparation of extracts:

2.5 grams powdered sample was sequentially extracted with 50 ml of benzene, acetone, ethanol, chloroform and aqueous solution soaked with 24 hours and filtered through the cheese cloth. The filtrate stored in airtight bottles. The prepared extracts were used for phytochemical screening and anti-bacterial activity.

Raw *Physalis angulata* stem and leaves flour:

Collected fresh leaves of *Physalis angulata* stem and leaves are spread on a tray and shade dried at room temperature for a period of 15-20 days for proper drying. The dried leaves are homogenized to fine powder in a mechanical pulverizer and stored in airtight bottles.

Phytochemical qualitative analysis:

The phytochemical tests were done for analyzing different chemical groups present in the extracts. These were done to find out the presence of bioactive chemical constituents such as alkaloid, flavonoids, tannins, phenol, terpenoids, glycoside, cardiac glycosides, anthroquinone, steroids and saponins. Detection of active phytochemical constituents was carried out for all the extracts using the standard procedures (Patel *et.al.*, 2008).

Test for alkaloids:

Mayer's Test:

3 ml of extracts was added to 1% HCL and then steamed. Few drops of Mayer's reagent were added to the mixture, Turbidity indicates the presence of alkaloids.

Test for flavonoids:

Lead acetate Test:

To 1 ml of extract, 1ml of 10% lead acetate was added. Formation of yellow precipitate showed the presence of flavonoids.

Test for Tannins:

Ferric chloride Test:

To 1ml of extract, 1 ml of distilled water was taken and stirred, few drops of Ferric chloride solution were added to the mixture bluish green colour precipitate showed the presence of tannins.

Detection of Phenols:

$FeCl_3$ Test:

About 2ml of plant extract was taken and warmed at 45-50°c. Then 2ml of 0.3% $FeCl_3$ was added. Formation of green or blue colour indicated the presence of phenols

Test for Terpenoids:

Salkowski Test:

About 2ml of chloroform was added to 1ml of the extract. Then 3ml of concentrated H_2SO_4 carefully added to form a layer. A reddish brown coloration of the interface indicated the presence of terpenoids.

Test for Glycosides:

2ml of extract was dissolved in chloroform and 2ml of acetic acid was added to the mixture. The solutions were cooled and then add few drops of sulphuric acid. A colour change from blue to green indicated the presence of glycosides.

Test for Cardiac glycosides:

Keller- Killiani Test:

1ml of extract was dissolved in 5ml of water 2ml of glacial acetic acid containing one drops of ferric chloride solution was added. This was under layer with 1ml of concentrated sulphuric acid. A brown ring at the interface indicated the presence of a deoxysugar characteristics of cardenolides. A violet ring may appear below the brown ring, while in the acetic acid layer a greenish ring may from just above the brown ring and gradually spread this layer.

Test for Anthraquinone:

1ml of the extract was boiled with 10 ml of sulphuric acid and filtered while hot. The filtered was shaken with added to 5m of chloroform. The chloroform layer was pipette into another test tube followed by addition of 1ml dilute ammonia. The resulting solution was observed for colour changes to violet indicated presence of anthraquinone.

Test for steroids:

Sal kowski Test:

To 2 ml of extract, was dissolved in chloroform, 2ml of concentrated sulphuric acid was added to the mixture. Red colour formation indicated the presence of steroids.

Test for saponins:

2 ml of extract dilute with 5 ml of distilled water and warmed. The formation of stable form indicated the presence of saponins.

ANTIBACTERIAL ACTIVITY:

Bacterial strains used:

The test organism were obtained from the Department of Botany, St. Mary's College (Autonomous), Thoothukudi. The two gram positive bacteria viz , *Bacillus subtilis* G +ve, *Staphylococcus aureus* G+ve were used in the present study. *Bacillus subtilis* is responsible for causing food borne gastroententis. *E.coli., Staphylococcus aureus* cause disease like mastitis, abortion and upper respiratory complications.

Disc diffusion assay (Bauer *et al.*, 1966)

Antibacterial activity was evaluated by agar disc diffusion method. Test solutions were prepared with known weight of different solvent extracts dissolved in 5% dimethyl sulphoxide (DMSO). What man No:1 sterile filter paper discs (5mm) were impregnated with 2o µl of these extracts and allowed to dry at room temperature. The spread plates were prepared by proper concentration of inocula. Each sample loaded disc was placed in the seeded agar plate. After 24-48 hours of 37° c incubation, the diameter of the inhibition zone was measured. For positive control, streptomycin disc (100µg / ml) was used, where as negative control, respective solvents were loaded on the sterile disc.

FT-IR (Fourier transforms infra –red spectroscopy) spectroscopic analysis (Vijaybasker and Shiyamala, 2s012)

Ten milligram of *P. angulata* leaf powder was mixed with 100 mg of dry potassium bromide (FT-IR grade) and then compressed into a pellet using hydraulic

press (5000-10000 psi). The pellet was immediately put into the sample holder and FT-IR (Systronics 166) spectra were needed in the range of 400 to 4000 cm^{-1}

SYNTHESIS OF SLIVER NANOPARTICLES: (Hamlata *et al.*, 2020)

Procedure:

For the green synthesis of sliver nanoparticles, we used the aqueous leaf extract of *P. angulata*. For this 9 ml of leaf extract was added 1 ml of 1Mm aqueous sliver nitrate solution, followed by heating at 80° C for 1 hour constant stirring. The formation of the AgNPs was preliminarily detected by the change in color from yellow to dark brown. The final synthesized sliver nanoparticles were denoted as AgNPs , which were freeze dried and then stored at 4° until further use. It showed that aqueous sliver ions could be reduced by aqueous extract of plant parts to generate extremely stable sliver nanoparticles in water.

plate 1 : Physalis angulata R

RESULTS AND DISCUSSION

RESULT AND DISCUSSION

In the present investigation, the phytochemicals, antibacterial activity are evaluated and chemical components are identified by FTIR. Sliver nanoparticles are synthesized from the leaves of *Physalis angulata* stem and leaves.

Systematic position

In the Bentham and Hooker system of classification, the systematic position of *Physalis angulata* R.Br stem and leaves is as follows,

Kingdom: phanerogams

Class: Dicotyledons

Subclass: Gamopetalae

Series ; Bicarpellate

Order: Polymoniales

Family: Solanaceae

Genus: *Physalis*

Species: *angulata*

Description:

Physalis angulata is an erect, many branched, spreading and quadrangular herb that lives several years. The plant can grow up to 6 to 7 feet tall. The plant and its all parts like root, seeds, leaves and fruits have been used for the medicinal purpose. *P. angulata* are simple, hairy, shortly stalked. It has small green / yellowish – white flowers which from narrow, long spikes, it grows up to 60cm long. The bracts around the flowers become sharp, pointed tips making the heads spiny.

Ecology

Physalis angulata grows best in moist, fertile soil, is tolerant of partial shade and occours widely as a weed of crops and pastures, and in waste are it can be found up to 3000 m altitude. Light forced does not kill it. At high temperature the plant does not develop well.

Origin and geo graphic distribution:

Physalis angulata is native to tropical America, and is now distributed pan tropically as a weed. In tropical Africa it occurs in most countries.

Phytochemical screening:

The bio-activity of natural products is due to phytochemical having therapeutic, prophylactic, nutrional and antibacterial properties. Phytochemical screening of plant material is thus vital in the knowledge of their therapeutic properties. They have been found to inadvertently confer anti- microbial protections to humans due to compounds synthesized in the secondary metabolism as well as being immunodulatory (Selvaraj *et.al* ., 2014).

The phytochemical analysis of different extracts of selected medicinal plant showed the presence of secondary metabolites such as alkaloids, flavonoids, tannins, phenols, terpenoids, glycosides, cardiac glycosides, anthroquinone, steroids, saponins (Table 1 and 2).

Each individual species has their own specific mechanisms for the synthesis and metabolism of phytochemicals. In some countries the species is used as medicine. The plant is used to treat several diseases such as asthma, malaria, boil, liver problem. All the plant parts of the herb are used for its medical value (Susanthi et al., 2015)

Table 1: Preliminary phytochemical analysis of *physalis angulata* leaf
(+ : present; -: absent)

Phytochemical analysis of *Physalis angulata leaves*						
		Extracts				
S.NO	Phytochemical	Ethanol	Acetone	Chloroform	Methonal	Aqueous
1.	Alkaloids	-	+	-	+	+
2.	Flavonoids	+	+	-	-	-
3.	Tannins	+	+	+	+	+
4.	Phenols	+	+	-	+	-
5.	Terpenoids	+	+	+	+	+
6.	Glycosides	+	+	+	+	-
7.	Cardiac glycosides	+	-	+	-	+
8.	Anthroquinone	+	+	+	-	+
9.	Steroids	+	+	+	+	-
10.	Saponins	+	+	+	-	-

Table 2: Preliminary phytochemical analysis of *physalis angulate stem*
(+ : present; -: absent)

Phytochemical analysis of *Physalis angulata stem*						
S.NO	Phytochemical	Extracts				
		Ethanol	Acetone	Chloroform	Methonol	Aqueous
1.	Alkaloids	+	+	+	+	+
2.	Flavonoids	+	+	+	+	-
3.	Tannins	+	+	+	+	+
4.	Phenols	+	-	-	-	+
5.	Terpenoids	+	+	-	+	-
6.	Glycosides	+	+	-	+	+
7.	Cardiac glycosides	-	-	-	+	+
8.	Anthroquinone	-	-	-	+	-
9.	Steroids	+	+	+	+	+
10.	Saponins	+	+	-	+	+

Antibacterial Activity

Life threatening diseases and high rate of mortality occur in animal and human population due to bacterial infection. Many bacteria both Gram positive and Gram negative contaminate food, water, air, soil, etc., and cause biological / microbial pollution. *Bacillus subtilis* is responsible for causing food borne gastroenteritis. *Eschercia coli*, cause diseases like mastitis albortion and upper respiratory complication. (Zhang e*t.al* .2005).

The acetone, ethanol, chloroform, bezene, aqueous extracts are of two selected plants are tested against pathogenic microbes *E.coli*, *Staphylococcus aureus* and *Bacillus subtilis*. The result of antibacterial activity of different plant extracts are shown in (Table 4 & 5). The antibacterial activity of these plants extract range from 5 mm to 17mm against *B.subtilis* (Table 4). The antibacterial properties exhibited by the extracts may be associated with the presence of the secondary metabolites through different mechanism.

The chloroform extract of *P. angulata* showed higher antibacterial activity against the tested organisms (Plate :4) These activities may be due to the phytochemical such as tannin, saponin and glycosides which could serve as the lead to the isolation of haemotherapatic agent. This in indication that the extract possesses substances that can inhibit the growth of some microorganisms.

Meanwhile *P. angulata* showed low antibacterial activity against *S.aureus* and *E.coli* range from 20 mm and 26 mm to 14 and 11 mm respectively. This low activity might be due to negligible amount of active principles present in the plants (Plate :3).

Table: 3 Antibacterial activity of different solvents of extracts of P *hysalis angulata* leaves

Bacterial cultures	Streptomycin	Chloroform	Methanol	Ethanol	Acetone	Aqueous extract
Eschercia coli	9mm	13mm	14mm	19mm	16mm	12mm
Bacillus ceresus	22mm	13mm	20mm	18mm	22mm	14mm
Staphylo cocus aureus	28mm	13mm	10mm	10mm	2mm	21mm

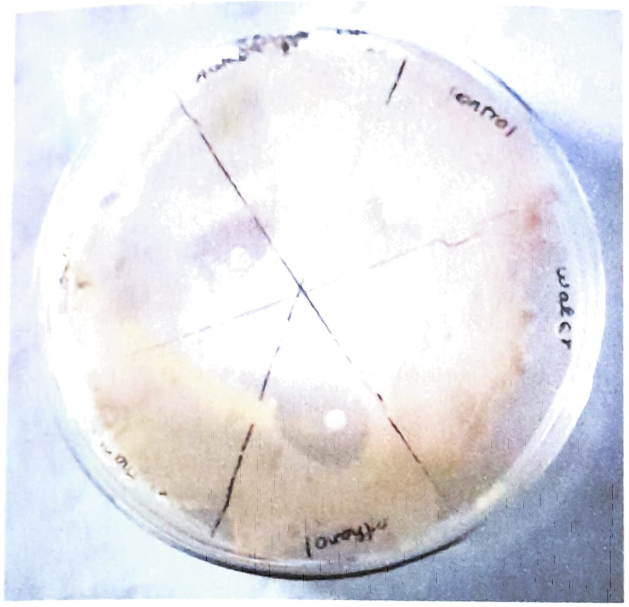
Control: Sterptomycin-

(2.5 mg/ ml)

Leaf extract

(2.5mg/ ml)

Antibacterial activity of Physalis angulata leaves against human pathogens



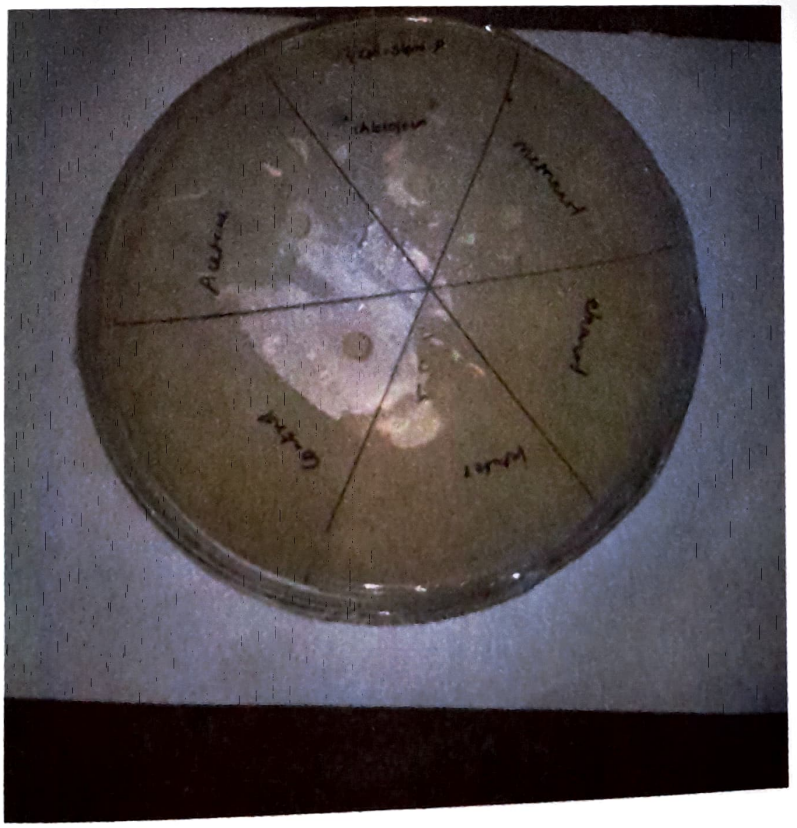


Table : 4 Antibacterial activity of different solvents of extracts of *physalis angula* stem

Bacterial Cultures	Acetone	Chloroform	Methonol	Aqueous	Ethanol	Streptomycin
Staphylo coccus	19mm	22mm	12mm	18mm	15mm	28mm
Escheria coli	16mm	13mm	14mm	12mm	19mm	9mm
Bacillus subtilis	20mm	12mm	10mm	13mm	12mm	26mm

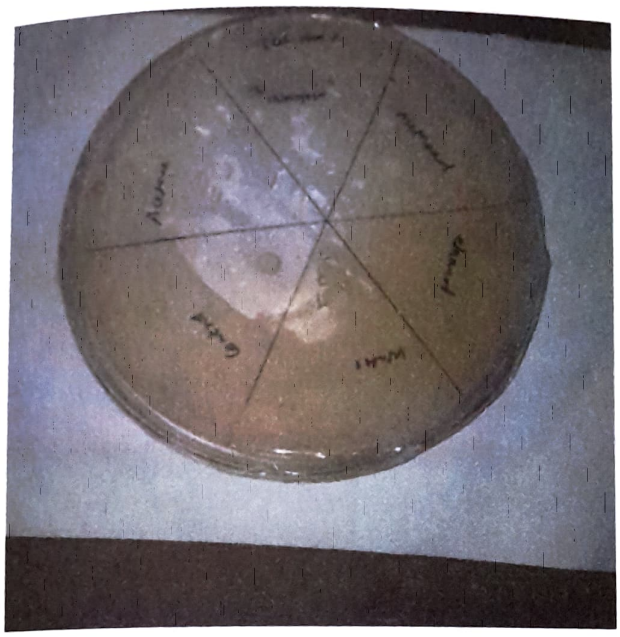
Control: Sterptomycin-

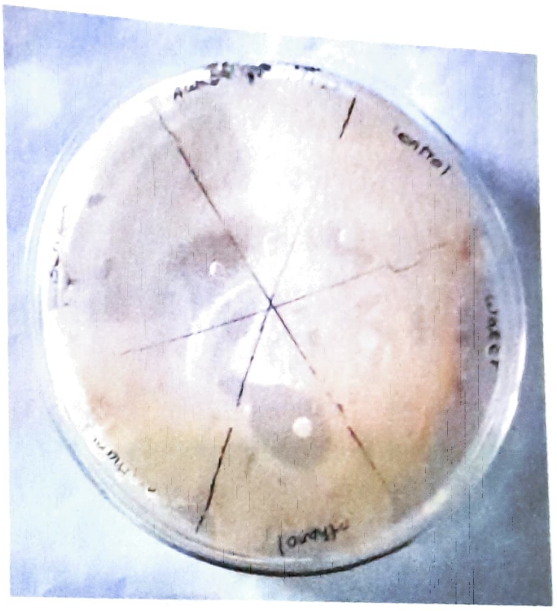
(2.5 mg/ ml) Leaf

extract -

(2.5mg/ ml)

Antibacterial activity of Physalis angulata leaves against human pathogens





The methanolic extracts of *Physalis angulata* showed high antibacterial activity against *B.subtilis* 20 mm followed by acetone, chloroform extract 12 mm, methanol extract 20 mm, aqueous 13 mm ethanol extract 18 mm.

Secondary phytochemical acted as an antibacterial agent by disrupting constituent of peptidoglycan in bacterial cells, leading to the disorganization of the cell wall and its death. It acted as an antibacterial agent by forming a complex compound with extracellular protein which impaired the integrity of bacterial cell membranes (Juliantina, 2008).

Polyphenol can link with and disable some bacterial enzymes essential for bacterial cell wall synthesis, action for simple phenols are attributed to the interaction with sulfhydryl groups that are present in microbial enzymes which leads to the enzyme inhibitions or through the interactions with a nonspecific chain of amino acids (Omar and Aldulaimi, 2017)

FTIR ANALYSIS

FTIR is an easy, simple, fast, suitable, non-invasive and cost-effective method to identify the role of biomolecules in the reduction of silver nitrate to silver zero oxidation state. Fourier Transform Infrared Spectrophotometer (FTIR) is perhaps the most powerful tool for identifying the types of chemical bonds (functional groups) present in compounds. The wavelength of light absorbed is characteristic of the chemical bond as can be seen in the annotated spectrum. By interpreting the infrared absorption spectrum, the chemical bonds in a molecule can be determined. The FTIR spectrum of leaf and stem , the data on the peak values and the probable functional groups present in the leaf and stem of *Physalis angulata* presented in Tables 3 to 4 and Figure 1 and 2 . The FTIR spectroscopic studies revealed different characteristic peak values with various functional compounds in the extracts. The

Table: 5 FT-IR Spectroscopy Analysis *of physalis angulata* leaf

S.NO	PEAK	BOND	FUNTIONAL GROUP
1	467.71	WEAK, STRONG	C-Br
2	517.85	STRONG, AROMATIC	N-H
3	615.25	STRONG, PRIMARY AMINES	N-H
4	675.04	STRONG , PRIMARY AMINES	C-H
5	780.15	SRTONG, HYDRO COMPOUND	S-O
6	831.26	SRTONG, SULPHINIC ACID GROUP	N-O
7	896.84	WEAK NITRATE GROUP	C-O
8	1032.81	VERY STRONG, RING STRETCH	C-O
9	1162.03	VERY STRONG, ESTER GROUP	C-O-C
10	1244.97	STRONG, ARALKYL ASYMMETRIC	C-O
11	1320.18	STRONG, HYDROXYL GROUP	C-O
12	1373.22	STRONG, HYDROXYL GROUP	N=O
13	1465.8	STRONG AMINES	N=O

14	1514.98	AROMATIC, ASYMMETRIC	C=N
15	1636.49	WEAK, STRONG,AROMATIC	C=O
16	1739.67	STRONG , ESTER	C=O
17	1861.18	STRONG, STRETCH	C=O
18	2916.17	MEDIUM, ASYMMETRIC	H-C-H
19	3408.95	MEDIUM, WEAK	O-H

Figure 1: FT-IR Spectrum of *of physalis angulata* leaf

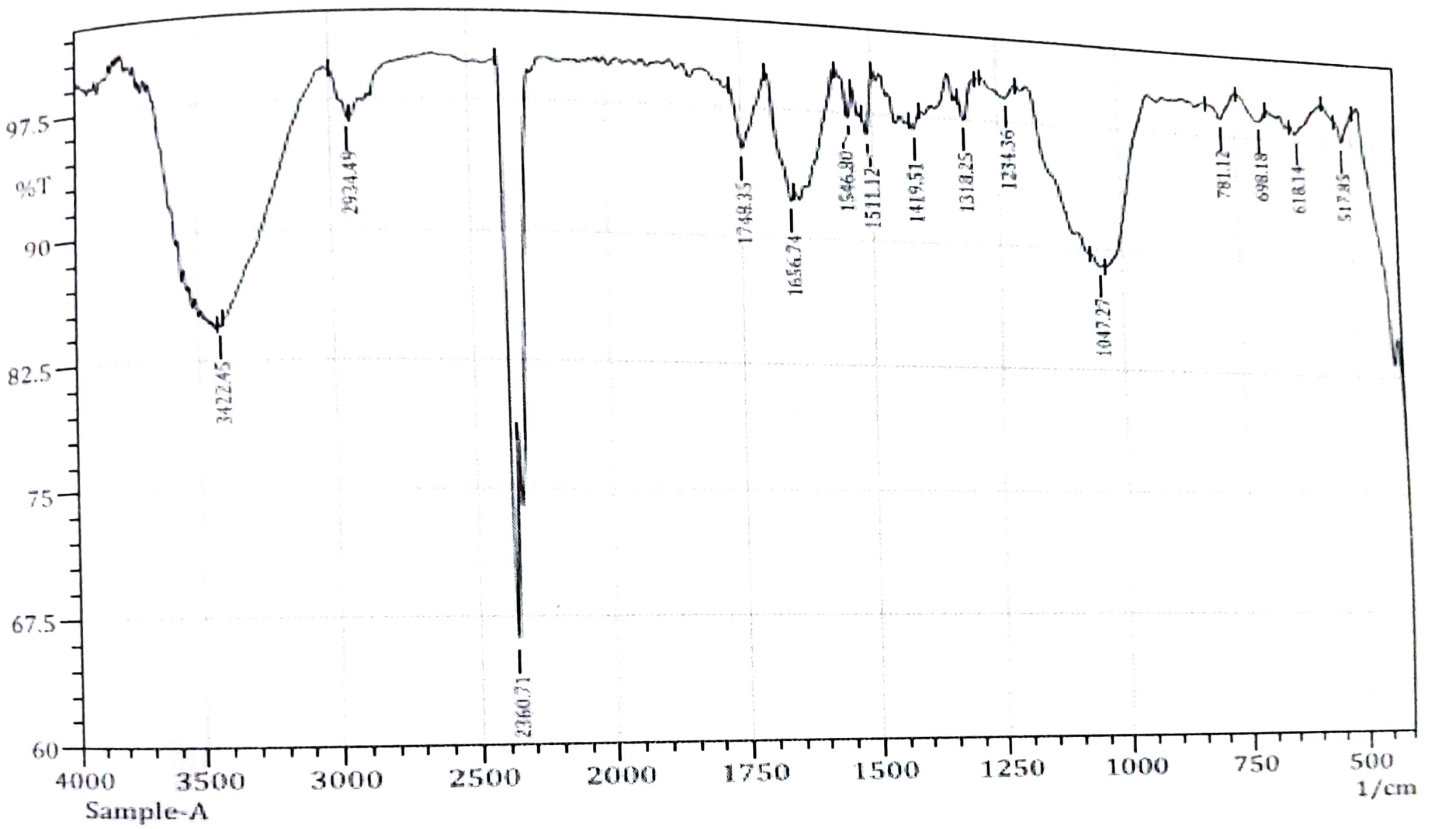
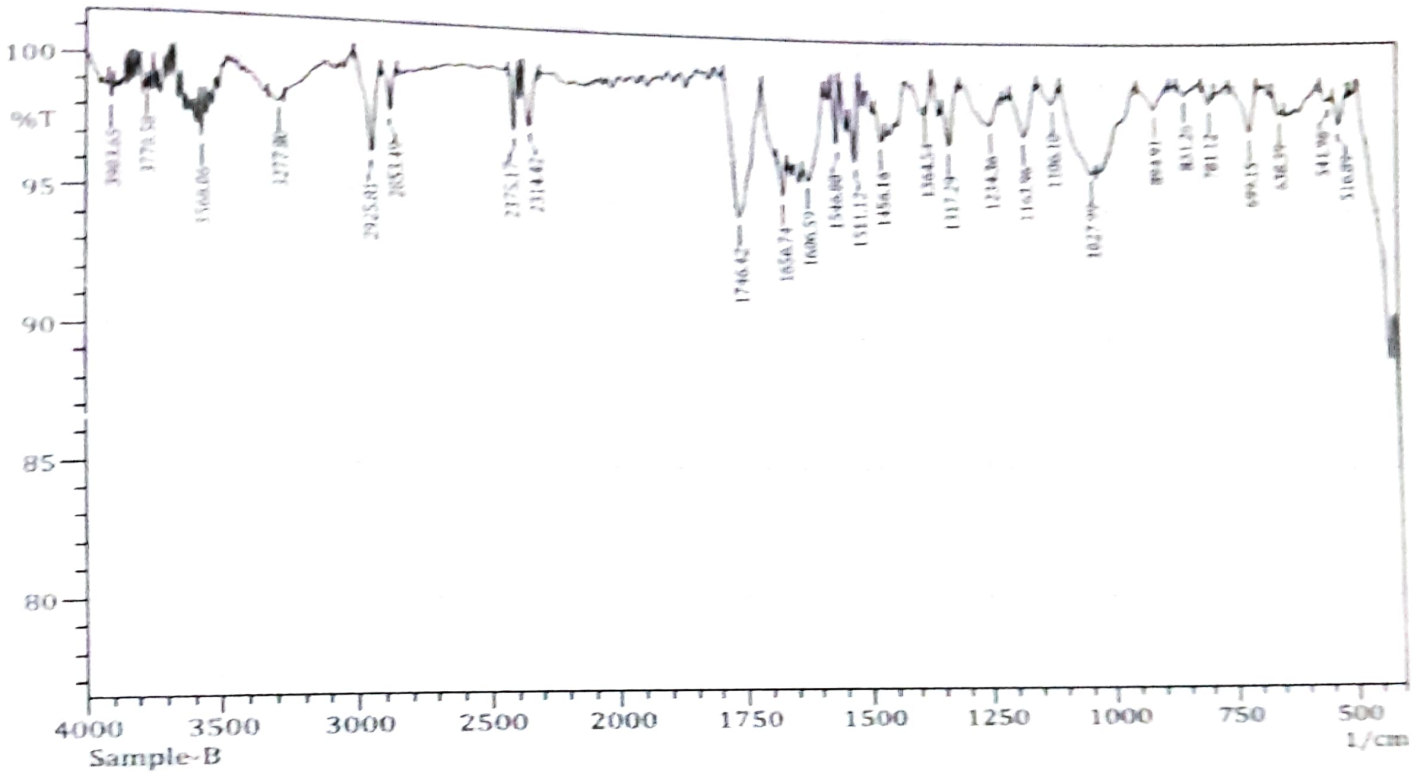


Table: 6 FT-IR Spectroscopy Analysis *of physalis angulate stem*

S.NO	PEAK	BOND	FUNTIONAL GROUP
1	514.96	STORNG, AROMATIC	C-Br
2	619.11	STRONG, PRIMARY AMINES	N-H
3	671.18	STRONG, VINYL AMIDES	C-H
4	781.12	STRONG, HYDRO COMPONDS	C-H
5	832.23	STRONG, SULPHINIC ACID GROUPS	S-O
6	894.91	WEAK, NITRATE GROUP	N-O
7	1019.31	STRONG, EHTER	C-O
8	1108.99	STRONG, ESTERS GROUPS	C-O
9	1160.1	WEAK, VERY STRONG	C-O
10	1238.21	STRONG, ACETATES	C-O-C
11	1320.18	STORNG ARYL TERTIARY AMINE	C-N
12	1387.69	STRONG , NITRO GROUP	N=O
13	1638.42	MEDIUM, PRIMARY AMIDES	N-H
14	2850.59	MEDIUM, SYMMETRIC STRETCH	H-C-H
15	2918.1	MEDIUM, URETHANES	N-H
16	3418.59	MEDIUM	O-H

Figure 2: FT-IR Spectrum of *of physalis angulata* stem



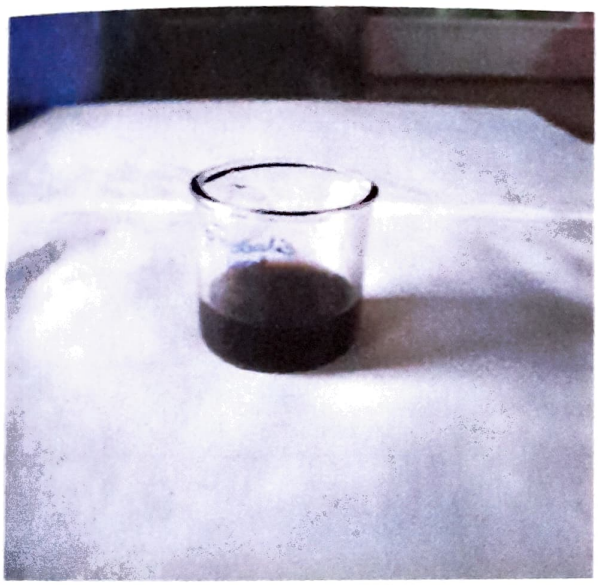
FTIR analysis of methanol and , chloroform leaf extracts of *Physalis angulata* confirmed the presence of amide, alcohols, phenols, alkanes, carboxylic acids, aldehydes, ketones, alkenes, primary amines, aromatics, esters, ethers, alkyl halides and aliphatic amines compounds, which showed major peaks.

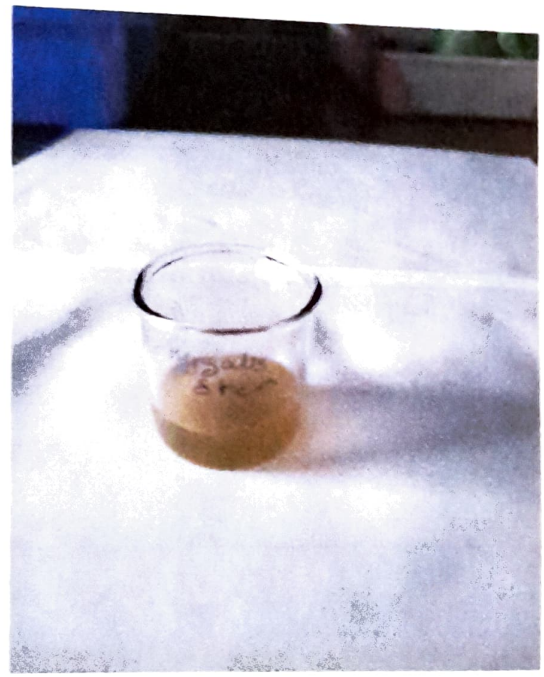
BIO SYNTHESIS OF NANO PARTICLES

Biosynthesis of metal nanoparticles utilising an environmentally safe and green synthesis process is presently in high demand to reduce unwanted effects in therapeutic and medical applications (Kalimuthu *et al.*, 2020). The silver nanoparticles that had been synthesised in this study was examined using UV–visible spectroscopy. After adding aqueous plant extract to Ag+ ions solution, biotransformation of silver ions into bunches of atoms (AgNPs) was detected. The resulted brown colour is due to Surface Plasmon Resonance (SPR). Colour shifts in phytofabricated AgNPs revealed the presence of excited electrons . At wavelengths spanning from 200 to 800 nm, UV spectra were obtained. , the resulting Ag NPs formed a broad surface peak at 546 nm. According to the FTIR data, the peaks represent flavonoids, phenols ands polyphenols meaning that flavonoids, phenols, and polyphenols were utilised as the reducing agent to create silver nanoparticles. Ag-NPs have recently been used in imaging and medicinal applications to investigate their unique properties. Engineered NPs may now be labelled with biological, physiological, and biomedical carriers and used in nano-based systems to deliver potent and precise drug delivery in cancer cell imaging zones (Jat *et al.*, 2020).

Plate 2 : Synthesis of silver nano particals using leaves of *P. angulate* leaf

Initial





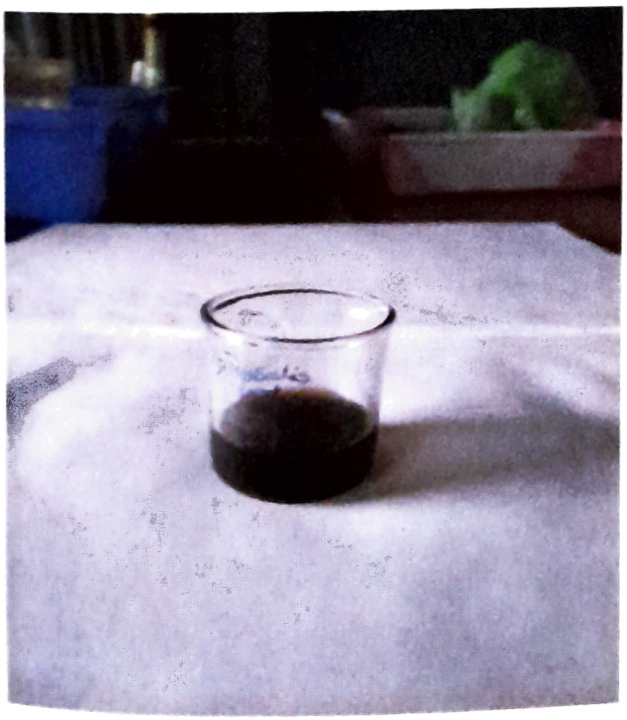


Table : 7 Synthesis of sliver nanoparticles (*P.a.l* and P.*a.s*)

nm	OD (*P.a.l*)	OD (P.*a.s*)
400	0.307	0.599
420	0.067	0.048
440	0.107	0.519
460	0.223	0.562
480	0.372	0.491
500	0.377	0.482
520	0.312	0.446
540	0.314	0.433
560	0.416	0.461
580	0.447	0.407
600	0.631	0.436

SUMMARY AND CONCLUSION

SUMMARY AND CONCLUSION

The project entitled "Evaluation of phytochemical, antibacterial activity and synthesis of sliver nanoparticles of *Physalis angulata* R.Br deals with the evaluation of phytochemical profile, antibacterial activity, synthesis of sliver nanoparticles from the leaves of *Physalis angulata* of a family Solanaceae.

The present work is focused on the following aspects:

- The phytochemical evaluation of dried leaf and stem powder *of Physalis angulata*
- Identification the chemical compounds of *Physalis angulata* stem and leaves.
- Antibacterial activity of leaf powder of *Physalis angulata* by disc diffusion method.
- Synthesis of sliver nanoparticles of *Physalis angulata* stem and leaves.

The phytochemical analysis shows the presence of alkaloids, flavonoids, tannins, terpenoids, phenols, glycosides, cardiac glycosides, anthroquinone, steroids and saponins. The antibacterial activity results indicated that the plant extracts significantly affected the cell membrane of Gram-positive and Gram-negative bacteria.

Considering current environmental problems and pollution associated with chemical synthesis, green synthesis offers alternative development prospects and potential applications. In conclusion, plant extracts are of great value as natural antimicrobials and can use safely as food preservatives . This study laid sufficient background for the further research on extracts from stem and

leaves of *Physalis angulata* for identification, subsequent purification and isolation of compounds having antibacterial activity and green chemistry. This method utilized less energy, minimized the toxic chemicals, simplified the procedure and exploited the natural materials being able to regenerate. So that further investigation will be carried out to characterize the nanoparticles to evaluate their efficiency of antibacterial activity.

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BIODEGRADATION OF TRICLOSAN BY A GREEN MICROALGAE CHLORELLA VULGARIS AND SCENEDESMUS DIMORPHUS

A dissertation submitted to

ST.MARY'S COLLEGE (Autonomous), Thoothukudi

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MASTER OF SCIENCE IN BOTANY

By

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April - 2023

CERTIFICATE

This is to certify that this dissertation entitled, **"Biodegradation of triclosan by a green microalgae *Chlorella vulgaris* and *Scenedesmus dimorphus*"** submitted by **J. Anitha Nancy Mary Reg. No. 21APBO01 to St. Mary's College (Autonomous),** Thoothukudi in partial fulfilled for the award of the degree of "Master of Science in Botany' is done by her under my supervision. It is further certified that this dissertation of any part of this has not been submitted elsewhere for other degree.

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DECLARATION

I do here by declare that this dissertation entitled, **"Biodegradation of triclosan by a green microalgae *Chlorella vulgaris* and *Scenedesmus dimorphus*"** submitted by me in partial fulfillment for the award of the degree of "Master of Science in Botany', in the result of my original and independent work carried out under the guidance of Dr. G. Flora M.Sc., M.Phil., Ph.D., Assistant Professor, Department of Botany, St. Mary's college (Autonomous), Thoothukudi and it has not been submitted elsewhere for the award of any other degree.

Station: Thoothukudi

Date: 12.01.2023

(J.Anitha Nancy Mary)

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Contents

INTRODUCTION

The environmental pollution is growing on a massive and unprecedented scale, mainly due to the burning of fossil fuels and the release of materials and chemicals to the environment, especially contaminants of emerging concern, for which there is no established treatment strategy (Sousa *et al.,* 2022). Triclosan is a synthetic broad spectrum antimicrobial agent which inhibits the activity of bacteria, viruses, and fungi (APUA, 2011). It was first registered as a pesticide with the EPA in 1969, but since 1990's it is being widely used in manufacturing household products. FAD approved the use of triclosan in Colgate toothpaste in 1997, but the serious effects of the chemical were discovered in the later years (FDA, 2010). The chemical is detectable in waterways, aquatic organisms ranging from algae, fishes, dolphins as well as in human urine, blood and breast milk. Laboratory studies have shown that triclosan is an endocrine disruptor capable of interfering with the hormones critical for normal development and reproduction. It enters into the aquatic systems through various pathways and poses a great threat to algae and other aquatic biota. It adsorbs to the soil and sediments and may bioaccumulate posing a concern for aquatic and terrestrial organisms (Kumar *et al.*, 2015).

Triclosan is considered as an important contaminant and is widely used in personal care products as an antimicrobial agent. The effects of culture media and light on biodegradation of triclosan and the changing morphology of microalgae were systematically studied (Tastan *et al.*, 2017). Triclosan is also incorporated into fabrics and plastics, including children's toys, toothbrush handles, cutting boards, pizza-cutter and mop handles, as well as surgical drapes and hospital over-the-bed table tops. Searches of patent databases further reveal a multitude of suggested or actual uses of

animals when its concentration reaches a certain value. Triclosan has a high adsorption potential and is easy to precipitate in sludge and sediment due to its high octanol-water partition coefficient (Quan *et al.*, 2019). Emerging contaminants (ECs) or "chemicals of emerging concern" are attracting considerable interest due to increased awareness of their risks to human health and aquatic biota. The most prevalent ECs include, but not limited to pharmaceuticals and personal care products (PPCPs), endocrine disrupting compounds (EDCs), perfluorinated compounds (PFCs), surfactants, gasoline additives, disinfection by-products, algal and cyanobacterial toxins, organometallic compounds, brominated and organophosphate flame retardants, plasticizers and nanoparticles (Maryjoseph and Ketheesan, 2020).

Microalgae are an abundant and economic source of biomass. In fact, this biomass has been used successfully for the removal of triclosan. However, most of the studies focused on freshwater, and there is no information on the behaviour of this biomass in more complex solutions such as seawater, in which triclosan also causes environmental problems (Santaeufemia *et al.*, 2019). Microalgae are single-celled eukaryotic microorganisms that represent a promising biodiesel feed stock that does not require land but produces greater qualities of lipids compared to conventional feed stock crops. (Yang *et al.*, 2023). Microalgal biotechnology only really began to develop in the middle of the last century. Now-a-days, there are numerous commercial applications of microalgae. For example, (i) microalgae can be used to enhance the nutritional value of food and animal feed owing to their chemical composition, (ii) they can be crucial role in aquaculture and (iii) they can be incorporated into cosmetics. Wastewater constitutes a great opportunity for microalgae as it can be considered as a medium for growing them at a low-cost and new potential market. Microalgae are known to be pollutant scavengers for a broad category of chemicals issued from the domestic, industrial and

agricultural sectors. Microalgae provide oxygen for bacteria while bacteria provide carbon dioxide for microalgae this leads to a significant decrease in the oxygen needs of the wastewater treatment process (Delrue *et al.*, 2016). A common strategy employed for mass culture of microalgae is to select a strain that can grow rapidly and synthesize large amounts of a desirable/target product and to develop a specific process to maximize the yield of the target product. *Scenedesmus sp* and *Chlorella sp.* have been evaluated for lipids production for the pharmaceutical and biofuels industries. *Chlorella sp.* are being commercially exploited as a protein source for food and feed. Many microalgae seem to possess a common default photosynthetic carbon partitioning mechanism which may result in 30-50% proteins, 20-40% carbohydrate and 8-15% lipid on a per total organic matter basis under favorable culture conditions, regardless of species or strains. As primary producers of unique qualities, algae provide valuable links in food webs. The mode of action comprises all physiological and behavioral signs that characterize a type of adverse biological response. In contrast, the mechanism of action is a detailed understanding of biochemical interactions that result in toxicity. Triclosan is an antimicrobial agent, effective against various types of gram-positive and gram-negative bacteria, some fungi and viruses. The question about the mode of action of triclosan was answered first in 1998. Triclosan blocks the lipid synthesis. In the recent rise of omics techniques like genomics, trasncriptomics, proteomics and metabolomics has opened new perspectives in the field of risk assessment of environmental stressors (Larras *et al.*, 2020).

This use of indirect photolysis plays a dominant role in the removal of organic chemicals and the rates are much faster than those of direct photolysis (Hom-Diaz *et al.*, 2022). To know the importance of microalgae in removing toxic chemicals from the aquatic environment, there is a need to understand the deep mechanism involved in the

bioremediation process. To explore and understand the depth information about the occurrence, toxicity and bioremediation efficiency of microalgae was studied through this research. The employment of microalgae in bioremediation techniques has emerged as a potential alternative and thus microalgae has been reported for remediation of a wide variety of toxic chemicals from the wastewater such as pharmaceutics, pesticides, heavy metals, personal care products and oils from contaminated sites. Microalgae are employed for treating wastewaters consisting of various industrial pollutants including triclosan (Bhatt *et al.*, 2022). Hence this present study was undertaken to understand the tolerance of microalgae on triclosan pollution and also study its toxicity. Through this research the society will be saved and living organisms are relieved from this suffocated environment.

SCOPE AND OBJECTIVES

Triclosan is an endocrine disrupting chemicals used as an antimicrobial agent in pharma and personal care products. It is a pollutant commonly occupied in aquatic environment. We can't able to overcome the triclosan polluted environment without remediation strategies. Bioremediation one of the boon to revive the contaminated areas without any unwanted effects. If the remediation is not done, the triclosan concentration in environment will be increased and creates a irreversible impacts. Hence the bioaccumulation of triclosan using *Chlorella vulgaris* and *Scenedesmus dimorphus* was studied and the results revealed that both the microalgae were have the potential to remediate the triclosan polluted environment. This research definitely becomes a boon to our society. Furthermore research seeks to understand potential effects of triclosan on organisms and environmental health.

Objectives

- To evaluate the toxicity of triclosan.
- To examine if triclosan biodegradation can be increased by optimizing the culture conditions.
- To evaluate the triclosan biodegradation capacity of *S. dimorphus* and *C. vulgaris.*
- To estimate the pigments, carbohydrate, proteins, H_2O_2 content of triclosan treated microalgae.

LITERATURE REVIEW

Pollution creates a great impact on living organisms; remediation will be helpful to reduce these impacts. Though many kind of remediation approaches are there, but many scientists majorly done researches in bioremediation of these pollutions.

Wang *et al.* (2018) revealed the antimicrobial ingredient triclosan, which is often found in personal care items, has contaminated a number of aquatic environments. The treatment of triclosan in wastewater has been shown to be greatly aided by biodegradation. The metabolic process is, however, poorly understood. The removal and biodegradation of triclosan from an aqueous culture medium was accomplished in this study using three common freshwater microalgae: *Chlorella pyrenoidosa* (*C. pyrenoidosa*), *Desmodesmus sp.*, and *Scenedesmus obliquus* (*S. obliquus*). When the three microalgae treated 400 g/L with triclosan for a day, a high clearance rate of up to 99.7% was seen. Triclosan elimination was credited to *Desmodesmus species* and *S. obliquus* for biotransformation and *C. pyrenoidosa* for cellular absorption. Simultaneously, triclosan metabolites resulted from hydroxylation, reductive dechlorination, or ether bond cleavage and their conjugates produced through glucosylation and/or methylation were detected in the biodegradation samples. Metabolic pathway of triclosan by algae were firstly proposed in this work, shedding light on the environmental fate of triclosan.

Pancha *et al.* (2014) studied the *Scenedemus sp.* CCNM 1077's morphological and biochemical changes as a result of nitrogen limitation and successive nitrogen starvation. The findings showed that consecutive nitrogen deprivation and nitrogen limitation circumstances severely reduced the organism's crude protein content and photosynthetic activity, but had little effect on dry cell weight or biomass production up to a nitrate

concentration of about 30.87mg/l *Scenedesmus sp.* CCNM 1077 was found to be significantly affected by nitrate stress in terms of cell shape. The growth medium had the maximum lipid (27.93%) and carbohydrate content (45.74%) after the total elimination of nitrate, which made it a promising feed stock for bio-diesel and bio-ethanol production. This method is distinctive for figure out freshwater microalgae respond to situations of sequential nitrate removal as well as nitrate limitation in terms of morphology and biochemistry.

Chokshi *et al.* (2017) described microalgae are one of the most promising sustainable sources for the generation of bio-fuels since they collect a significant amount of lipids and carbohydrates in nutrient-poor environments. They used a short-term nitrogen starvation of 1,2 and 3 days in a green microalgae called *Acutodesmus dimorphus* in the current work to produce biomass with greater lipid and carbohydrate contents. The production of bio-fuel oxidative stress-tolerant microalgae appears to be very effective, according to a few recent findings. Reactive oxygen species (ROS), cellular enzymatic anti-oxidants superoxide dismutase (SOD), catalaze (CAT), ascorbate peroxidase (APX), and non-enzymatic scavengers proline and polyphenols were investigated in order to study the function of oxidative stress caused by nitrogen deprivation.

Bano *et al.* (2021) studied the occurrence of emerging contaminants like pharmaceutical drugs and personal care products in aquatic systems is identified as a potential risk to human health. High amounts of several of the regularly used medications are reportedly present in many effluents because traditional wastewater treatment systems are unable to sufficiently remove them. In recent years, microalgae-based systems have been researched as an environmentally secure substitute for chemical oxidation techniques for the removal of these developing pollutants. Therefore, the ability of a method including the micro-algal consortium to tolerate high levels of pharmaceutical in the environment

while also concurrently eliminating the synthetic hormone estradiol, the anti-inflammatory drug diclofenac and the antibacterial agent triclosan was evaluated. Estradiol, diclofenac and triclosan had an effective concentration with 50% mortality for the consortium of 16, 8, and 8 mg/L for each pollutant. Then, these three medications were independently spiked at effective concentrations with 50% mortality in algal growth media and the micro-algal growth in their presence and the drug elimination were observed in a shake-flask arrangement. According to the study, the microalgal consortium significantly removed estradiol (91.73% 0.0175), diclofenac (74.68% 0.0092), and triclosan (78.47% 0.015) while they were in their growing phase. Additionally, it was discovered that microalgae's primary removal mechanism rather than adsorption was the breakdown of the medicines. According to measures of the biomass and chlorophyll content, neither estradiol nor diclofenac had any immediate detrimental effects on the microalgal growth. However the triclosan was harmful to the microalgal growth because the consortium did not endure more than 5 days after dosing. The removal of developing pollutants showed promising results, and by utilizing the naturally occurring microalgal consortia, it is possible to create a treatment system to remove various medicines from wastewater.

Vo *et al.* (2020) examined the effects of selective sole carbon source-induced micropollutants (MPs) cometabolism of *Chlorella sp.* using the following methods: (i) production of extracellular polymeric substances (EPS), superoxide dismutase, and peroxidase enzymes; (ii) MPs removal efficiency and cometabolism rate; (iii) identification of MPs' potential degradation products; and (iv) validation of degradation pathways using the Eawag Comparatively to the presence of MPs alone, the addition of the sole carbon sources boosted the concentrations of EPS and enzymes by a factor of 2 to 100. This proved that MPs co-metabolism had taken place. Tetracycline,

sulfamethoxazole, and bisphenol a all had clearance efficiencies that ranged from 16 to 99%, 32 to 92%, and 58 to 99%, respectively. The MPs concentrations collected in microalgae cells also decreased 400-fold by raising EPS and enzyme activity. There were various MP breakdown products as a result of co-metabolism. Insightful knowledge of co-metabolism for MPs remediation in wastewater was gained from this work. Based on the findings, appropriate microalgae carbon sources can be chosen for real-world applications to remove MPs from wastewater while simultaneously recovering biomass for a variety of industries and making money.

Tastan *et al.* (2017) reported Triclosan is a common antibacterial ingredient found in personal care products and is regarded as an important pollutant. This study reveals the acute toxicity of triclosan and 2,4-dichlorophenol as well as the biodegradation of triclosan by two freshwater microalgae. Systematically investigated were the effects of culture media and light on the biodegradation of triclosan and the alteration of microalgal shape. At 10 days, *Geitlerinema sp.* and *Chlorella sp.* decomposed 3.99 mg/L of triclosan by 82.10% and 92.83%, respectively. Even at higher concentrations (50 mg/L), triclosan had no harmful effects on *Chlorella sp.* after 72 hours of exposure, according to the microalgal growth inhibition experiment. *Geitlerinema sp.* and *Chlorella sp.* produced 2,4-dichlorophenol as a triclosan breakdown product, according to HPLC analyses. This study proved to be beneficial to understand biodegradation and acute toxicity of triclosan by microalgae in order to provide aquatic environmental protection.

Wu *et al.* (2009) found triclosan and triclocarban are antibacterial agents widely used in personal care products, but limited information is available on their environmental behavior in soils. This study investigated their adsorption and degradation in soils with

and without biosolids. Competitive sorption was observed when coexisted, but the cosolute effect was concentration dependent.

According to Dhillon *et al.* (2015) the Triclosan (TCS) is a multi-purpose antimicrobial agent used in everyday household personal care and consumer products, which has been detected in sewage treatment plant effluents, surface, ground and drinking water. There is increasing concern about its potential negative effects on human and animal health, but conventional water and wastewater treatment processes are unable to completely remove it.

Wn e*t al.* (2007) observed gas chromatography-ion trap mass spectrometry was used to examine the presence of triclosan in samples of Hong Kong's trash, rivers and seawater. For the quantitative study, 13C12-triclosan served as an internal standard. Use of a C18 solid-phase extraction cartridge was used to prepare and clean up water samples. The recoveries of triclosan in three distinct amounts of contaminated coastal water ranged from 83 to 110%. The relative standard deviations and relative error were less than 11.0 and 12.3%, respectively (n = 3), and the technique detection limit was 0.25 mg/L for triclosan in 1 L of water. Water samples were gathered from rivers, coastal water bodies, and wastewater treatment facilities. The approach was successfully used to test the samples at mg/L levels.

Yan *et al.* (2023) described triclosan (TCS) has been rigorously prohibited from being added to consumer items, but its ongoing use in hospitals and other healthcare facilities and the various residues it leaves behind still represent a risk to aquatic life and aquatic ecosystems. In this work, they looked at how *Chlorella vulgaris* responded to various TCS concentrations in terms of growth, biochemical changes, and physiological reactions. Analysis was also done on the potential harmful pathways brought on by

excessive reactive oxygen species (ROS) generation and PSII disruption. According to the findings, TCS significantly impacted *C. vulgaris* physiology, cellular ultrastructure, and proliferation in a dose-effect dependent manner. TCS prevented the growth of *C. vulgaris*, which resulted in the expansion of the mitochondria, the disarray of the thylakoids, the rupturing of the cell wall, the loss of organelles, and the lysis of the cytoplasm. Malondialdehyde (MDA) levels were raised, ROS accumulated, and antioxidant enzyme activities were up regulated as a result of the severe oxidative damage caused by TCS. Furthermore, in TCS-induced algal cells, chloroplasts, mitochondria, and cell membranes were the primary locations of ROS accumulation, with mitochondria being the place where ROS accumulated the greatest. In addition, TCS caused damage to the reaction center (RC inactivation), donor side (OEC damage), and accepted side (electron transport from Q_A to Q_B) of PSII in *C. vulgaris*, leading to inhibition of photosynthetic activity. These results could provide novel insights into the mechanisms of TCS-induced ROS accumulation and photosynthetic inhibition in *C. vulgaris*, which would contribute to a deep understanding of TCS toxicity on algae.

Dai *et al.* (2021) revealed that the widespread use of triclosan as an additive in personal care and medical antibacterial goods, as well as the rising levels of triclosan discharged into aquatic ecosystems, aquatic ecological systems may be at risk. In this work, they looked at how *Chlorella vulgaris's* growth, chlorophyll fluorescence, and antioxidant enzyme activity were affected by exposure to various triclosan doses. The findings demonstrated that *Chlorella vulgaris* growth can be stimulated by low concentrations of triclosan (0.75 mg/L), however Chlorella vulgaris growth was significantly inhibited by 1.05 mg/L triclosan. *Chlorella vulgaris* may be better able to tolerate and make use of bright light if there is a low concentration of triclosan (0.75 mg/L). When exposed to 1.05 mg/L triclosan, we saw a considerable rise in the malondialdehyde concentration of

Chlorella vulgaris. *Chlorella vulgaris* exposed to triclosan had higher intracellular superoxide dismutase and catalase (CAT) activities than the control groups, and the rise in these activities was positively associated with the quantity of triclosan. The findings also demonstrated that when *Chlorella vulgaris* is exposed to 1.05 mg/L triclosan, increased H_2O_2 may, in turn, harm the CAT structure and ultimately render CAT activity inactive. This study offered a theoretical framework for assessing triclosan's ecological danger in the aquatic environment.

Chen *et al.* (2018) found that antimicrobial triclosan is a common ingredient in personal care items like toothpaste, soap, deodorant, plastic, and cosmetics. Triclosan has been widely used, and its discharge into wastewater, surface water, and soils, and has attracted a lot of attention lately. According to reports, triclosan has been found in a number of environmental components. Studies on its toxicology have revealed that it could have an impact on the environment, particularly on aquatic habitats. Since then, biodegradation of triclosan has been documented in a variety of systems, including axenic cultures of microorganisms, full-scale WWTPs, activated sludge, sludge treatment systems, sludge-amended soils, and sediments. In this study, an extensive literature was undertaken, to present the current knowledge of the biodegradation behavior of triclosan and highlights the removal and transformation processes to help understand and predict the environmental fate of triclosan. Experiments at from lab-scale to full-scale field studies are shown and discussed.

Santaeufemia *et al.* (2019) observed the developing contaminant triclosan is significant. It has spread because municipal wastewater treatment systems cannot completely remove it, which causes significant environmental issues. Triclosan removal may be environmentally favorable when using biomass from microorganisms as a sorbent of contaminants. In this study, *Phaeodactylum tricornutum*, a marine microalgae, was used

to characterize the removal of triclosan from biomass (dead and living) in cultures exposed to light and in a complex solution (seawater). The effects of pH and reuse were assessed and discussed, along with maximum removal capacity, isotherms, kinetics, and FTIR characterization. Additionally assessed was triclosan's photo-degradation. Both biomasses displayed comparable efficacy; in just 3 hours, a pollutant concentration of 1 mg L-1 was completely removed with a biomass concentration of 0.4 g L-1. The biosorption procedure was governed by a pseudo-second order model. The entire process (photo-degradation plus biosorption) was appropriately modelled using pseudo-third order and Elovich kinetics, taking into account that photo-degradation is a first-order process. The pH dropped, which led to an increase in bio-sorption. The Temkin isotherm fit the experimental data the best. After five cycles, both biomasses demonstrated good reuse, with only a 7% loss in efficiency. *P. tricornutum* biomass is an appealing eco-material for triclosan elimination because it is less expensive and easier to handle than other sorbents.

Kurad *et al.* (2021) revealed the industrial revolution in the production of pharmaceuticals and personal care products (PPCPs) has significantly improved public health. However, this advancement has also resulted in water pollution as a result of the unintentional disposal of these synthetic chemicals, resulting in unsanitary conditions. Conventional wastewater treatment systems can remove the majority of contaminants, but they are ineffective at removing PPCPs. Plant-based remediation is a simple, yet highly effective and environmentaly friendly method that can supplement existing wastewater treatment. Phytoremediation of emerging contaminants is still in its early stages, and many key concepts, such as uptake and detoxification mechanisms, remain unexplored when compared to microbial processes. The latest studies on the biochemistry and application of phytoremediation for the removal of PPCPs from

wastewater, focusing on the mechanisms of uptake and detoxification through the enzymatic biotransformation of PPCPs and the latest field applications using innovative engineered systems. Future research recommendations are addressed, including the need for topics to be investigated in PPCPs interactions with plant tissues, their metabolic transformation in plants, the development of new predictive uptake models, and futuristic advancements involving cutting-edge genetic engineering methodologies for the realization of advanced phytoremediation technologies.

Chan *et al.* (2021) reported Industrial effluents that contained persistent organic pollutants (POPs) have a number of negative effects on the ecosystem, human health, and the environment. Scientists have been working on improving and developing POPs removal from wastewater treatment for the past many decades. However, the traditional approaches of POP removal from wastewater are expensive and risk causing secondary pollutants, such as soil and water body pollution. The use of environmentally friendly processes including biosorption, bioaccumulation, and biodegradation has recently attracted attention and hinted at the potential of green technology globally. One of the latest wastewater treatment methods has been discovered to be microalgae-bacteria consortiums. The current biological wastewater treatment system could be effectively improved by the synergistic interactions between bacteria and microalgae. The mechanics of the microalgae-bacteria symbiosis system and a comparison of contemporary sophisticated wastewater treatment systems and conventional systems will be critically examined by them.

Chu *et al.* (2022) observed recently, the biotransformation of sulfamethoxazole by microalgae has attracted increasing interest. In particular, cytochrome P450 has been suggested to be the main enzymatic contributor to this biodegradation. However, the molecular evidence of Cytochrome P450 enzymes being involved in SMX

biodegradation remains relatively unclear, hindering its applicability. Herein, the biodegradation of SMX by *Chlorella sorokiniana* (*C. sorokiniana*) was investigated and comprehensively elucidated the reaction mechanism underlying CYP450-mediated SMX metabolism. *C. sorokiniana* was able to efficiently remove over 80% of SMX mainly through biodegradation, in which CYP450 enzymes responded substantially to metabolize SMX in cells. Additionally, screening of transformation products (TPs) revealed that N^4-hydroxylation-SMX (TP270) was the main TP in the SMX biodegradation pathway of microalgae. Molecular dynamics simulation suggested that the aniline of SMX was the most prone to undergo metabolism, while density functional theory (DFT) indicated that SMX was metabolized by CYP450 enzymes through H-abstraction-OH-rebound reaction. Collectively, this work reveals key details of the hydroxylamine group of SMX, elucidates the SMX biodegradation pathway involving CYP450 in microalgae in detail, and accelerates the development of using microalgae-mediated CYP450 to eliminate antibiotics.

Zhu *et al.* (2018) observed the toxicity of single microplastics on organisms has been widely reported; however, their combined toxicity on phytoplankton with other contaminants has received little attention. We investigated the toxicity of triclosan (TCS) on the microalgae *Skeletonema costatum* using four different microplastics: polyethylene (PE, 74 m), polystyrene (PS, 74 m), polyvinyl chloride (PVC, 74 m), and PVC800 (1 m). Growth inhibition as well as oxidative stress, such as super-oxide dismutase (SOD) and malondialdehyde (MDA), were studied. We discovered that TCS had a clear inhibitory effect on microalgae growth at the test concentrations, and that single microplastics had a significant inhibitory effect in the order PVC800 > PVC > PS > PE. However, the combined toxicity of PVC and PVC800 with TCS was lower than that of PE and PS. One possible explanation for the lower toxicity of TCS was its higher

adsorption capacity on PVC and PVC800. Because of the small particle size, the combined toxicity of PVC800 remained the most significant (PE PVC PS PVC800). The joint toxicity systems were all antagonistic, according to the independent action model. Furthermore, the reduction of SOD was greater than that of MDA, indicating that physical damage was more severe than intracellular damage. Microplastic aggregation and physical damage to algae were clearly visible in SEM images. This study showed that the presence of organic pollutants can influence the effects of microplastics.

Chandra *et al.* (2021) studied industrial wastewaters characterized by the presence of toxic pollutants are recognized as a serious threat to human health. Given that the concentrations of these pollutants are in the range of mg to μg, they are re-referred to as pollutants of emerging concern (PECs). The major sources of these PECs are pharmaceuticals, cosmetics, and plastics. There is a growing concern to detect and develop strategies for the remediation of PECs. The conventional sewage treatment plants cannot efficiently remove these compounds and pose challenges toward safe disposal. Various methodologies viz. advanced oxidation techniques, photocatalysis, membrane separation, and adsorption have been extensively studied for the removal/degradation of PECs. However, these are characterized with challenges viz. high cost, low efficiency, and high chemical input. In this regard, microalgae have gained recognition for the removal of emerging contaminants.

Singh *et al.* (2021) studied water pollution is a major concern in the world, with direct discharge of wastewater causing eutrophication and degrades their physico-chemical characteristics. Conventional treatment approaches are expensive and inefficient, but exploring microalgae has been found to be an efficient and ecofriendly technique for purification of aquatic environs. Microalgae can effectively remove N (90-98.4%), P (66%–98%), Pb (75%–100%), Zn (15.6-99.7%), Cr (52.54%–96%), Hg (77%–97%), Cu

(45%–98%) and Cd (2-93.06%) from contaminated aquatic systems. Additionally, microalgae play a pivotal role in degrading the complex pesticides (Endosulfan, lindane, isoproturon and glyphosate) and emerging contaminants (triclosan, bisphenol A, 17-ethinylestradiol, tramadol and diclofenac). Microalgae also produce biomass, making phycoremediation more frugal and sustainable.

Subashchandrabose *et al.* (2013) described organic chemical substances are present in both soil and aquatic environments, and their biological responses include accumulation and degradation. Cyanobacteria and microalgae are highly adaptive and can grow autotrophically, heterotrophically or mixotrophically. Laboratory culturing of strict phototrophic algae has limited their potential as bioremediation agents, but mixotrophic algae can contribute to sequestration of carbon, which is otherwise emitted as carbon dioxide to the atmosphere under heterotrophic conditions. Molecular methods and metabolic and genomic information are needed to identify and select mixotrophic species with capabilities to degrade organic pollutants, and to monitor the efficiency of remediation efforts under the field conditions.

Hu *et al.* (2021) study revealed that the, landfill leachate was pre-treated with NaClO, and then diluted to 5%, 10% and 15% for microalgae growth of *Chlorella vulgaris* and *Scenedesmus dimorphus* in the mono- and co-culture modes to investigate the nutrient removal and growth characteristics of microalgae. The results revealed that land fill leachate with the 10% dilution rate was conducive for microalgae growth and exhibited robust biomass growth and the highest nutrient removal efficiency. The co-culture biomass in 10% landfill leachate achieved 0.266 g/L within 10 days and demonstrated the improved nutrient utilisation efficiency of microalgae. In addition, the chemical oxygen demand, ammonia nitrogen, total nitrate and total phosphorus removal efficiencies accordingly reached 81.0%, 80.1%, 72.1% and 86.0% in 10% landfill

leachate. Meanwhile, both the enzyme activity and fluorescence parameters proved that the cell activity of co-culture was higher than that of mono-culture.

Mustafa *et al.* (2021) explained in their study that the environmental pollution is increasing due to anthropogenic activities and different types of toxic contaminants enter the environment from different sources. Bioremediation is one of the promising techniques for remediation of these pollutants. Microalgae have the capacity to remove different types of contaminants through different methods such as biosorption, bioaccumulation and biodegradation.

Delrue *et al.* (2016) described that the microalgae have been shown to be a source of multiple bio-based products ranging from high value molecules to commodities, and can also be used for the de-pollution of wastewaters of different origins. This study focuses on harnessing the bioremediation capacity of microalgae to treat wastewaters in order to develop the microalgae industry and find other alternatives to the classic wastewater treatment processes. The research discussed, the both strategies of selecting the best microalgae strain to treat a specific wastewater or pollutant and using a natural or an artificial consortium to perform the treatment. The process options for treating wastewaters using microalgae are discussed up to the final valorization of the biomass, and challenges need to be addressed in order to further develop the potential.

Leng *et al.* (2020) found that microalgae-based technology has been explored as a potential alternative for the treatment of wastewater containing antibiotics by adsorption, accumulation, biodegradation, photo degradation, and hydrolysis. They discussed the toxicities of antibiotics on microalgae, the mechanisms of antibiotic removal, and the integration of microalgae with other technologies such as ultraviolet irradiation (photocatalysis), advanced oxidation, and complementary microorganism degradation.

Nguyen *et al.* (2021) revealed micropollutants have become a serious environmental problem, with many efforts to remove them using physical, chemical and biological methods. They explained the potential of microalgae-based systems for wastewater treatment to obtain high-quality effluents, recover algal biomass for fertilizers, protein-rich feed, biofuel, and other practical uses. They also discussed the inhibition of micropollutant on microalgae growth, and other treatment methods,

Li *et al.* (2022) provided a critical summary of algae-based technologies and their important role in antibiotic wastewater treatment. It covers mechanisms such as bioadsorption, bioaccumulation, and biodegradation, integration of algae with other microorganisms, hybrid algae-based treatment and constructed wetlands, and factors affecting algal antibiotic degradation. It also highlights the use of algae as a precursor for the production of biochar, modification of biochar with other materials to improve its antibiotic removal capacity, and recent novel approaches for enhancing antibiotic removal.

1. Media preparation

Chu's medium No. 10 was used to grow the microalgae. The media composition is as follows.

Chemicals	g/L
Calcium nitrate	0.232
Dipotassium hydrogen phosphate	0.01
Magnesium sulphate	0.025
Sodium carbonate	0.02
Sodium silicate	0.044
Ferric ammonium citrate	3.5 mg
Citric acid	3.5 mg
Trace metal- 1 Ml	
Boric acid	2.4
Manganese chloride	1.4
Zinc chloride	0.4
Calcium chloride	0.02
Copper chloride	0.1
Distilled water	1000 mL

The pH of the medium should be 7.1. The prepared media was autoclaved and stored in glass containers for future use.

2. Culture condition

The freshwater microalgae *Chlorella vulgaris* NRMC-F 0128 and *Scenedesmus dimorphus* NRMC-F 0174 were purchased from National Repository for Microalgae and Cyanobacteria (NRMCF, Trichy, India). Initially 2 mL of inoculum was transferred into 10 mL sterile Chu's medium No. 10 and kept inside the incubation room at 25±2°C, under a 16:8 light: dark cycle provided by cool white fluorescent lights. After 3 days of incubation, the algal cells attained sufficient growth; then transfer the whole broth culture 50 mL of fresh broth medium and incubate. After 5 days of incubation, transfer to 100 mL of fresh broth medium and incubate till well growth of algae. Use this as a mother culture and sub-cultured at regular intervals.

3. Experimental setup

The experiment was carried out in triplicate manner in 250 mL Erlenmeyer flasks containing 100 mL of Chu's Medium No. 10 which inoculated with 10% of the actively growing culture of *C. vulgaris* and *S. dimorphus* cells. The microalgae were cultivated at 25^0C±2 under 15 µmol/m^2/s of light intensity (cool white fluorescent lights were used as the light source) and 16:8 h of light: dark period. When the culture attained optical density 0.2 in the absorbance wavelength 750 nm, then it was treated with different concentration of triclosan. A control and solvent control flasks were also maintained. Flasks were manually shaken thrice a day to avoid the adherence of the cells to the surface of the flasks. Totally 15 days incubation period was given to the culture. Morphological changes in the cells were observed under light microscope (Pancha *et al.*, 2013).

Growth analysis

Microalgal growth was monitored at regular intervals (0 day, 3rd day, 6th day, 9th day, 12th day and 15th day) by measuring optical density at 750 nm using UV-vis spectrophotometer.

Pigment estimation

For the analysis of pigments content 2ml culture was centrifuge at 10000 rpm for 5 minutes, the supernatant was discarded and 2 ml of absolute methanol was added to the pellet. The content was mixed properly and incubated at 45^0C for 24 h in the dark. The absorbance of the supernatant were read at 470, 652.4, and 665.2 nm and corrected for the turbidity by subtracting the absorbance at 750nm. The pigments contents were calculated using the equation (Pancha *et al.*, 2013).

$$Chlorophyll\ a\ (mgL^{-1}) = 16.72\ A_{665.2} - 9.16\ A_{652.4}$$

$$chlorophyll\ b\ (mgL^{-1}) = 34.09\ A_{652.4} - 15.28\ A_{665.2}$$

$$Carotenoid\ (mgL^{-1}) = \frac{1000A_{470} - 1.63\ chlorophyll\ a - 104.9\ chlorophyll\ b}{221}$$

Measurement of ROS

For the measurement of H_2O_2 content, microalgal cells were harvested by centrifugation and the cell pellet was homogenized in 0.1% w/v TCA solution. The homogenate was centrifuged for 10 minutes at 10,000 rpm. An aliquot of 0.5 ml of the supernatant was mixed with 0.5 ml of the supernatant was mixed with 0.5 ml of 10 mM potassium idodide. The absorbance of the solution was read at 390 nm. The H_2O_2 concentration in the sample was determined form a calibration curve prepared using the known concentrations of H_2O_2 (Pancha *et al.*, 2013)

Determination of Carbohydrates

1.5 ml of culture was centrifuged at 1000 rpm for 5 minutes, discard the supernatant and 0.2ml of distilled water was added to the pellets. Then 0.4 mf of 40% (w/v) KOH was added and heated at 90°C for the 1hr. after incubation cool down at room temperature and 1.2 ml of cold absolute alcohol was added to the treated sample. The sample was kept in refrigerated at -20°C overnight. Centrifuge and discard the supernatant. Added 1.5ml of distilled water to the pellet and take took 0.2 ml of sample from this was mixed with 0.4 ml of pre-chilled 75% H_2SO_4 in test tube and then vortexed. 0.8 ml of anthrone reagent (2 gL-1 75% H_2SO_4, freshly prepared) was added to the sample and boiled at 100°C for 15 minutes and cooled at room temperature. Read absorbance at 620 nm. For blank, 0.2 ml sample mixed with 2.2 ml 75% H_2SO_4 without anthrone reagent. D-glucose was used as standards (Chen and Vidyanathan, 2013).

Determination of protein

2ml of sample was centrifuged, 1 ml 0.5 N NaOH was added to the pellet and extracted at 80°C for 10 minutes with occasional stirring. After cooling and centrifugation, the supernatant was transferred to a new tube. The alkali extraction was repeated 3 times. The final repeat was heated at 100°C for 10 minutes for complete extaction of residual proteins. All the three extractions were pooled and mixed well before analysis. An aliquot (0.05 ml) of copper sulphate (0.21% $CuSo_4$-$5H_2O$ in 30% NaOH) was added to 0.1 ml of the samples and colour formation monitored at 310 nm. Bovine serum albumin (BSA) was used as the standard for calibration (Chen and Vidyanathan, 2013).

RESULT AND DISCUSSION

Triclosan is one of the emerging contaminants of aquatic ecosystem. It creates unpleasant impacts on both flora and fauna; it causes many disorders to human beings at higher concentration. Therefore the current study was investigated the degrading ability of microalgae on triclosan pollution. *Chlorella vulgaris* and *Scenedesmus dimorphus* was obtained from NRMC-F Trichy and cultivated in our laboratory. The selected microalgae were treated with triclosan at various concentrations (20 mg L^{-1}, 60 mg L^{-1}, 100 mg L^{-1}) and incubated for 15 days. The data were obtained at regular intervals. The selected microalgae were shown different tolerability. *S. dimorphus* was tolerable up to 20 mg L^{-1} of triclosan whereas *C. vulgaris* was tolerable up to 60 mg L^{-1} concentration of triclosan. This tolerability was indicated by the growth rate. The growth rate of triclosan degradation by *C. vulgaris* and *S.dimorphus* was shown in Fig. 1 & 2. For the comparative study of triclosan degradation resulted that high level of degradation was done by *C. vulgaris* than *S. dimorphus*. Similar results were obtained by Lee and Chu, 2013. In their study *Burkholderia xenovorans* and *Rhodococcus ruber* were unable to degrade triclosan while propane-grown *Mycobacterium vaccae* can completely degrade 5 mg/L triclosan. *R. jostii* grown on biphenyl, propane & LB medium was able to degrade 5 mg/L triclosan incompletely (Lee and Chu, 2013).

The triclosan (20 mg L^{-1}) treated *C. vulgaris* showed high amount of chlorophyll a (4.93 µg ml^{-1}), Chlorophyll b (10.76 µg ml^{-1}) and carotenoids (2.09 µg ml^{-1}) at 15th day culture than control, solvent control and all other concentrations shown in Table. 1. The triclosan (20 mg L^{-1}) treated *S. dimorphus* showed high amount of chlorophyll a (5.93 µg ml^{-1}), Chlorophyll b (9.76 µg ml^{-1}) and carotenoids (3.09 µg ml^{-1}) at 15th day culture than control, solvent control and all other concentrations shown in Table. 2. The data obtain

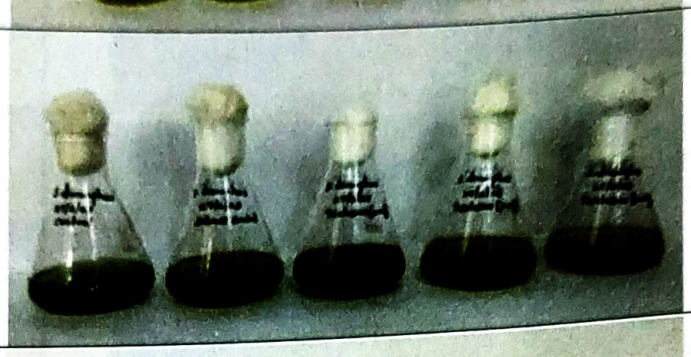
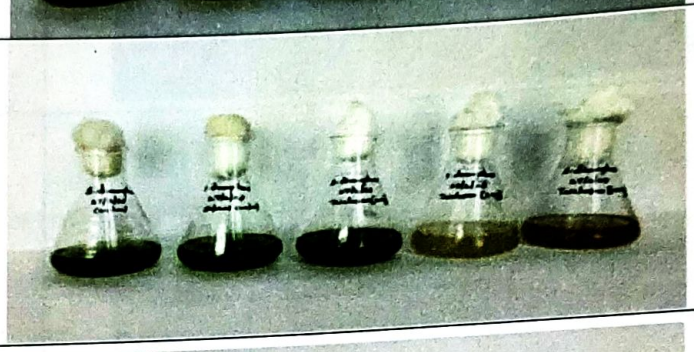
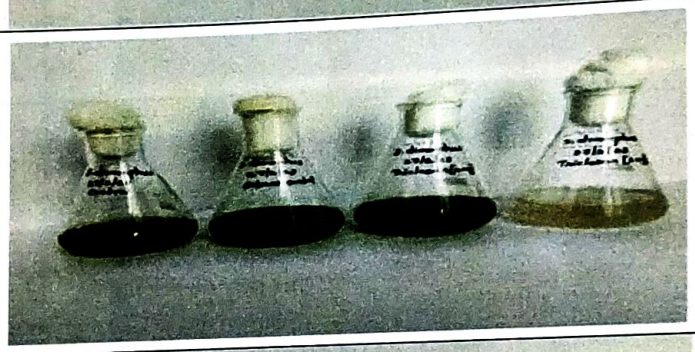
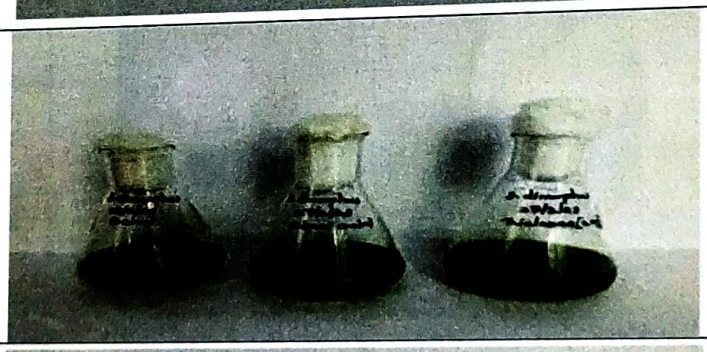
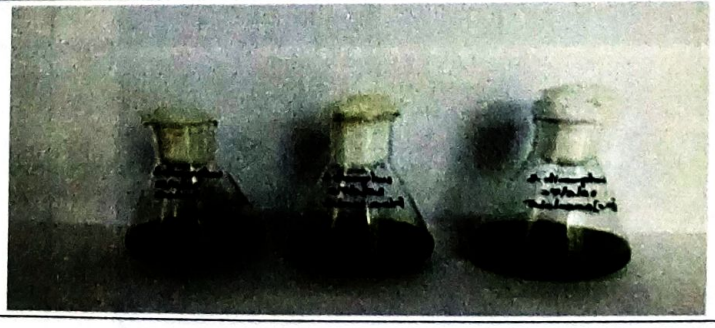
0 day	
3rd day	
6th day	
9th day	
12th day	
15th day	

Plate 1. Study of triclosan degradation on *Scenedesmus dimorphus* NRMC-F 0174

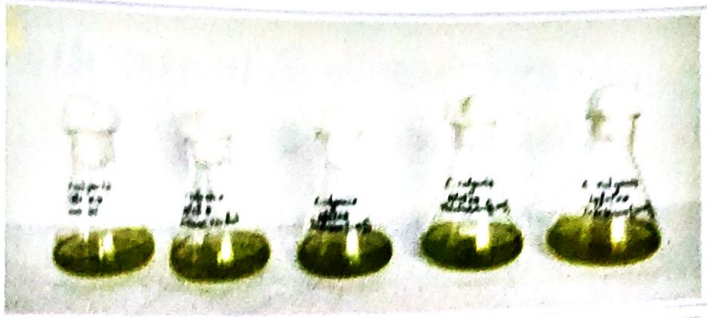
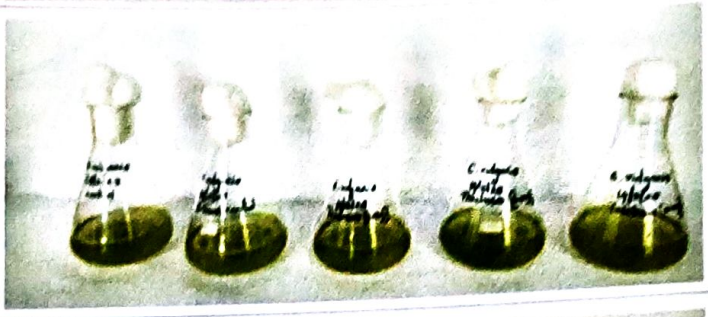
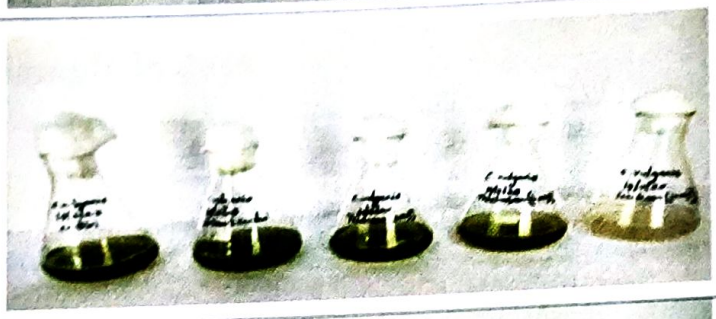
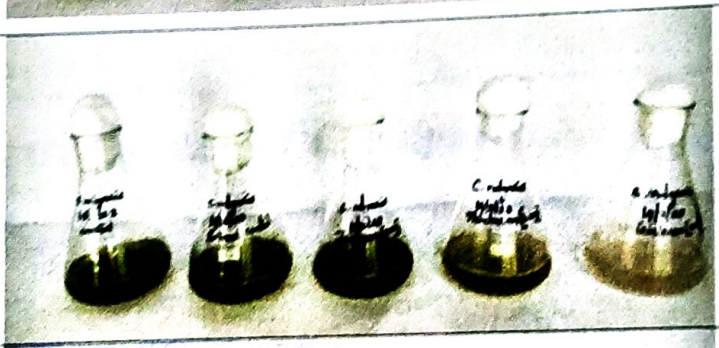
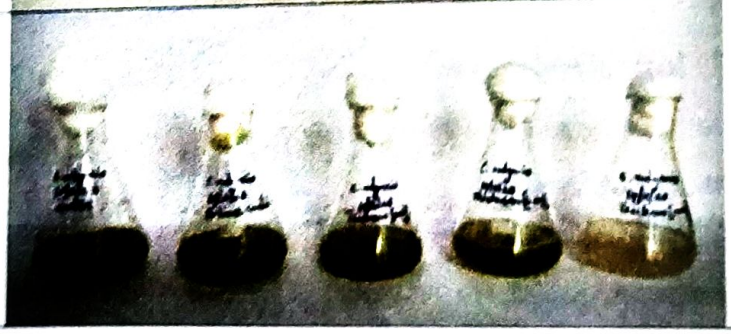
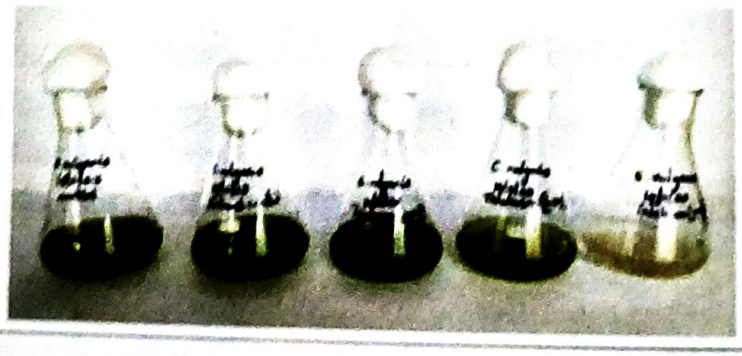
0 Day	
3rd Day	
6th Day	
9th day	
12th Day	
15th Day	

Plate 1. Study of triclosan degradation on *Chlorella vulgaris* NRMC-F 0128

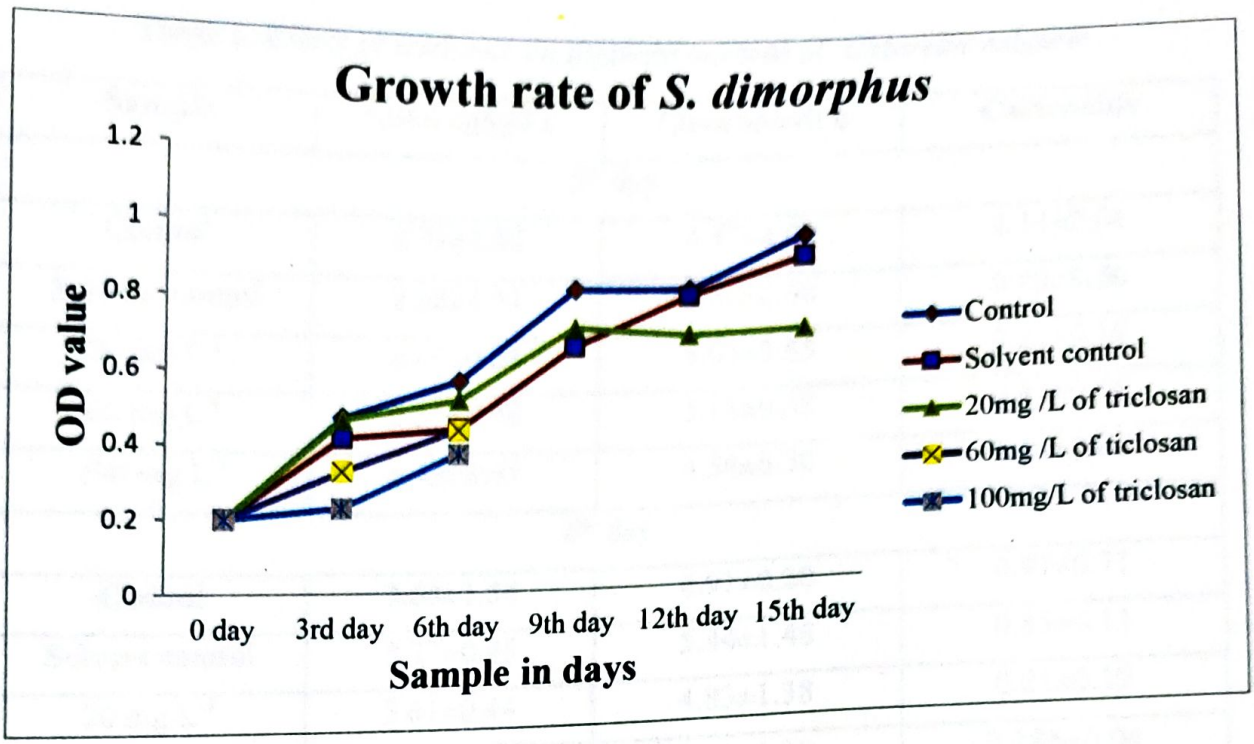


Fig. 1 Determination of growth rate of *S. dimorphus* after treatment with triclosan

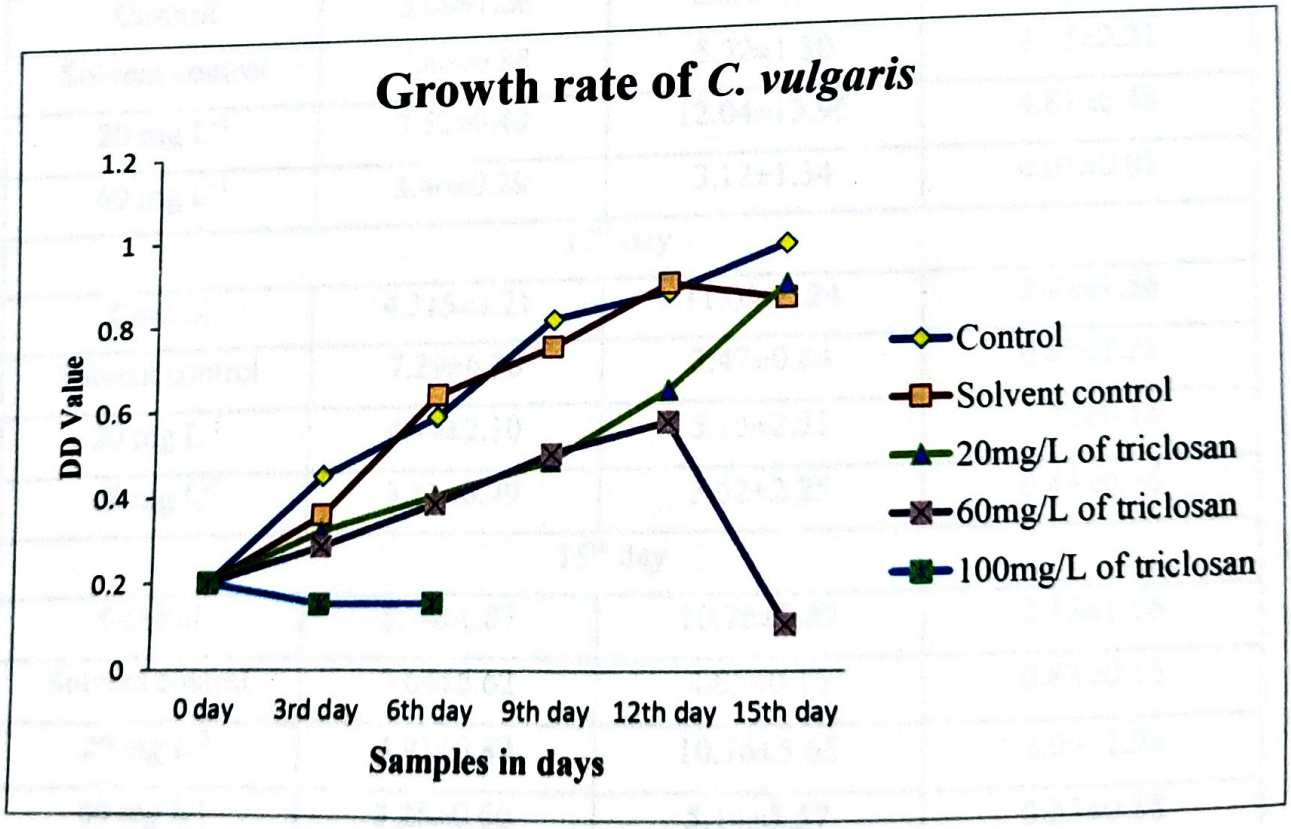


Fig. 2 Determination of growth rate of *C. vulgaris* after treatment with triclosan

Table 1. Effect of triclosan on pigment content of *Chlorella vulgaris*

Sample	Chlorophyll a	Chlorophyll b	Cartenoids
3rd day			
Control	2.71±3.42	4.67±4.07	2.14±2.04
Solvent control	2.68±3.33	2.58±2.58	0.69±0.66
20 mg L^{-1}	6.49±0.88	5.05±3.65	0.82±0.18
60 mg L^{-1}	2.57±0.86	3.18±0.58	0.87±0.54
100 mg L^{-1}	6.43±8.43	4.54±6.30	2.16±2.94
6th day			
Control	3.64±1.54	6.91±0.20	0.97±0.71
Solvent control	3.27±0.45	5.44±1.48	0.85±0.11
20 mg L^{-1}	3.61±0.44	4.83±1.38	0.21±0.19
60 mg L^{-1}	3.13±2.51	10.1±1.19	2.185±0.04
100 mg L^{-1}	0.92±0.77	1.65±1.19	0.18±0.23
9th day			
Control	3.08±1.26	2..31±1.51	1.42±1.96
Solvent control	3.66±0.88	5.32±1.30	2.18±2.31
20 mg L^{-1}	3.52±0.40	12.04±13.96	4.81±6.48
60 mg L^{-1}	3.46±0.29	3.12±1.34	0.01±0.01
12th day			
Control	4.315±3.21	11.09±0.24	2.48±1.56
Solvent control	7.29±6.38	3.47±0.84	0.87±1.21
20 mg L^{-1}	4.74±2.10	5.15±2.31	1.72±0.16
60 mg L^{-1}	3.11±0.79	3.62±2.25	0.43±0.36
15th day			
Control	5.74±1.87	10.76±0.82	2.12±1.06
Solvent control	0.64±6.62	4.85±0.19	0.83±0.15
20 mg L^{-1}	4.93±0.89	10.76±5.65	· 2.09±1.95
60 mg L^{-1}	3.25±0.60	5.14±3.57	0.33±0.38

Table 2. Effect of triclosan on pigment content of *Scenedesmus dimorphus*

Sample	Chlorophyll a	Chlorophyll b	Cartenoids
3rd day			
Control	1.52±1.79	3.18±3.71	0.53±0.50
Solvent control	2.29±1.53	1.79±1.99	2.03±2.99
2mg L^{-1}	0.93±0.38	2.14±1.78	0.28±0.19
6mg L^{-1}	1.34±1.70	1.59±2.27	0.10±0.08
10mg L^{-1}	1.2±1.34	2.37±2.75	0.26±0.21
6th day			
Control	0.90±0.41	3.39±1.68	0.62±0.57
Solvent control	1.51±0.90	6.51±3.42	1.91±1.17
20 mg L^{-1}	4.04±5.10	3.56±4.13	1.31±1.39
60 mg L^{-1}	2.02±1.53	3.6±2.91	0.93±0.85
100 mg L^{-1}	1.18±1.57	2.04±2.59	1±1.25
9th day			
Control	3.085±1.26	2.31±1.51	1.42±1.95
Solvent control	3.66±0.88	5.33±1.31	2.17±2.31
20 mg L^{-1}	3.53±0.40	12.04±13.96	4.82±6.48
12th day			
Control	4.32±3.22	11.09±0.25	2.48±1.55
Solvent control	7.29±6.38	3.47±0.84	0.87±1.21
20 mg L^{-1}	4.75±2.10	5.15±2.32	1.72±0.16
15th day			
Control	5.745±1.87	10.76±0.82	2.12±1.06
Solvent control	6.45±6.63	4.85±0.19	0.83±0.15
20 mg L^{-1}	5.93±0.89	9.76±5.65	3.09±1.95

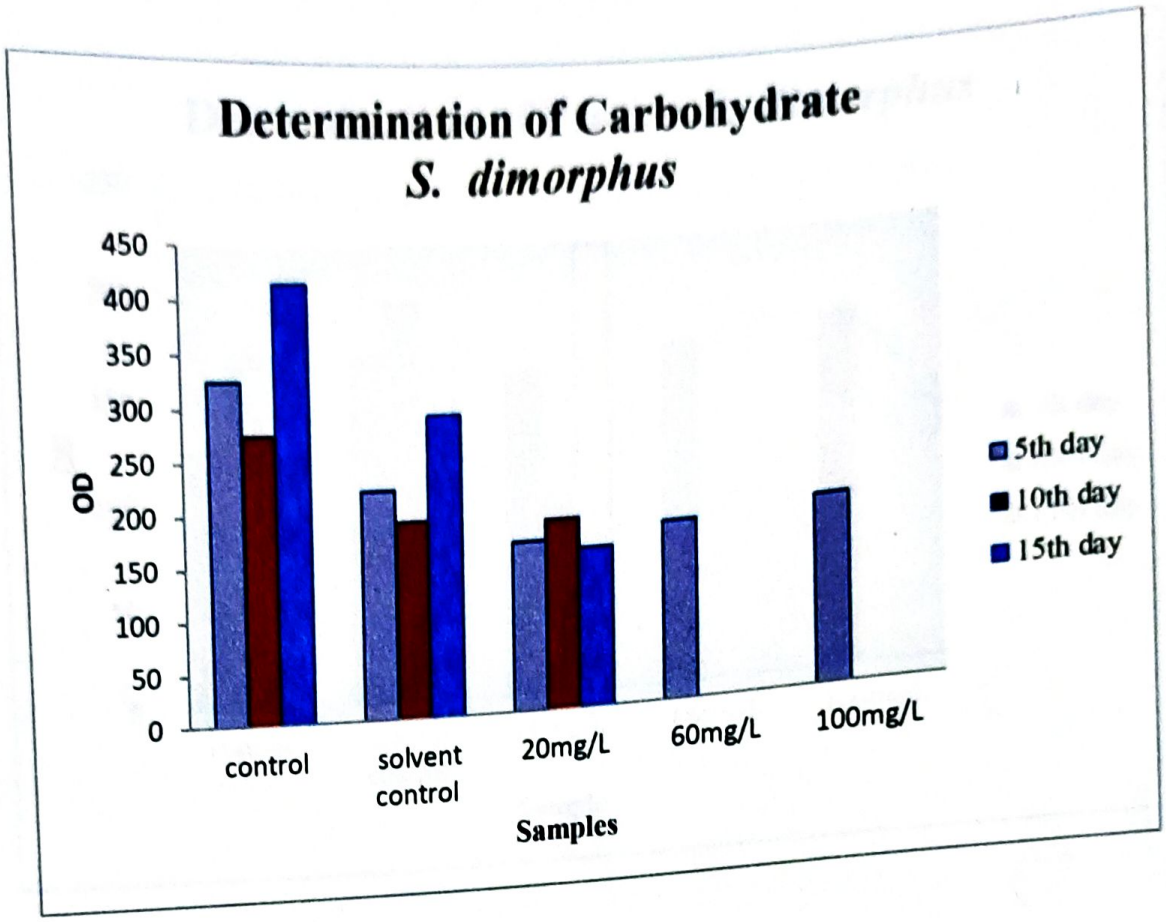


Fig. 3 Determination of carbohydrate content of *S. dimorphus* after treatment with triclosan

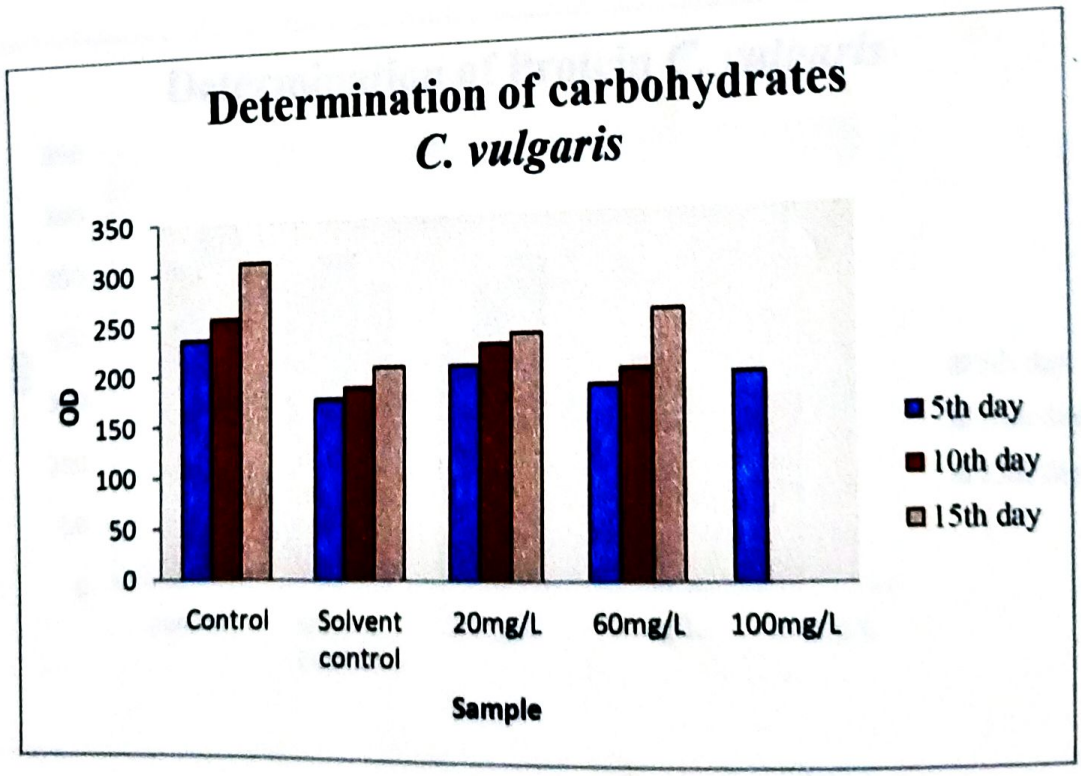


Fig. 4 Determination of carbohydrate content of *C. vulgaris* after treatment with triclosan

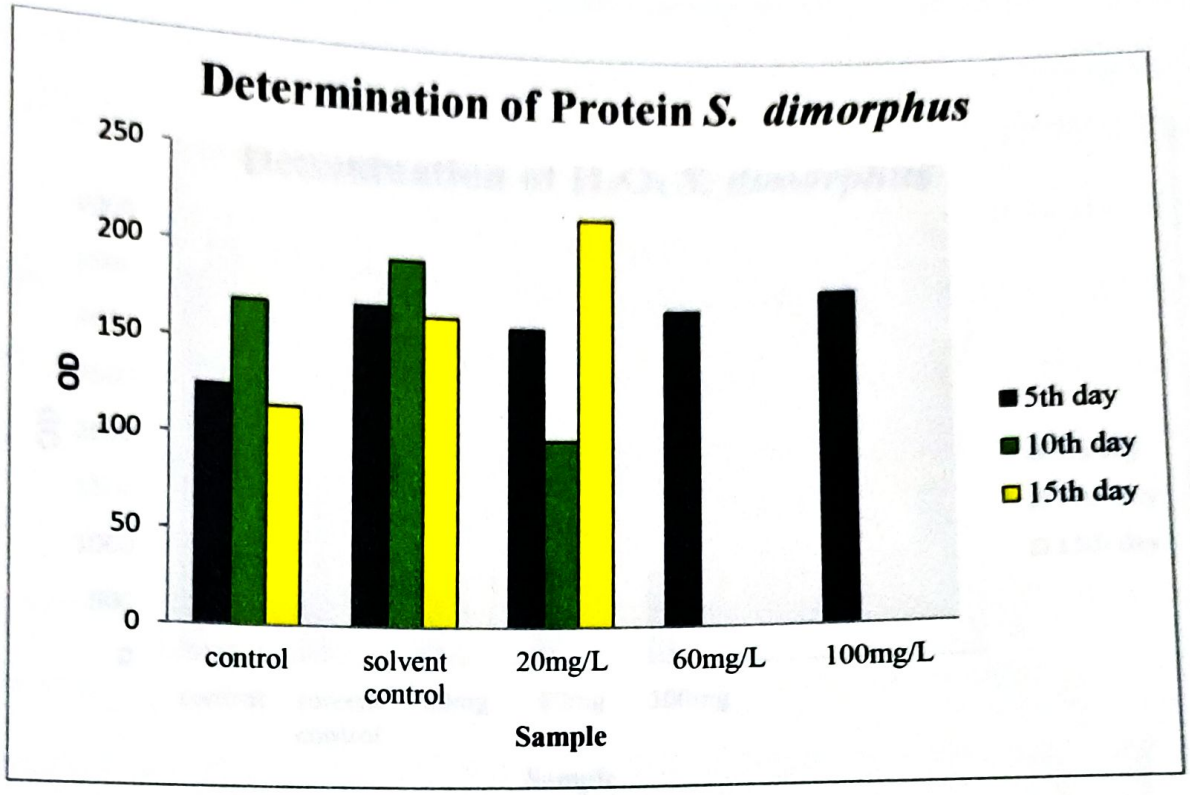


Fig. 5 Determination of protein content of *S. dimorphus* after treatment with triclosan

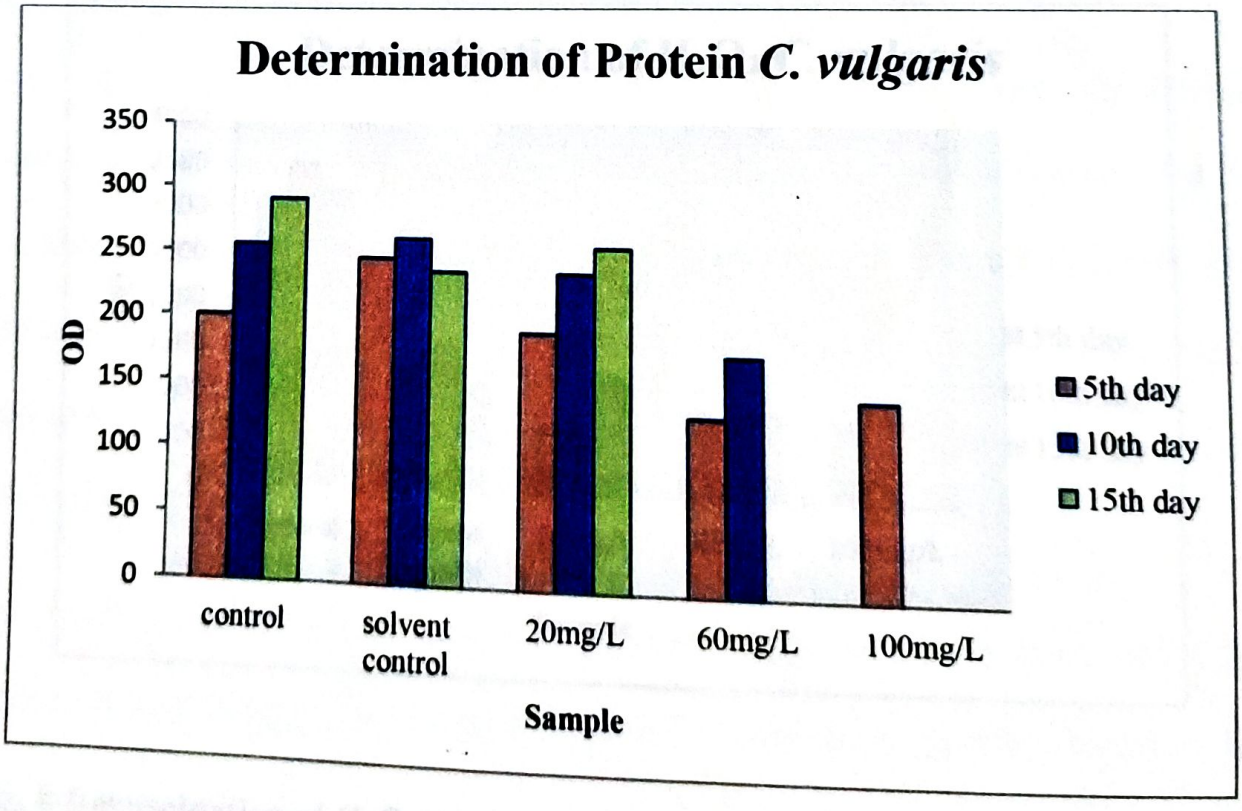


Fig. 6 Determination of protein content of *C. vulgaris* after treatment with triclosan

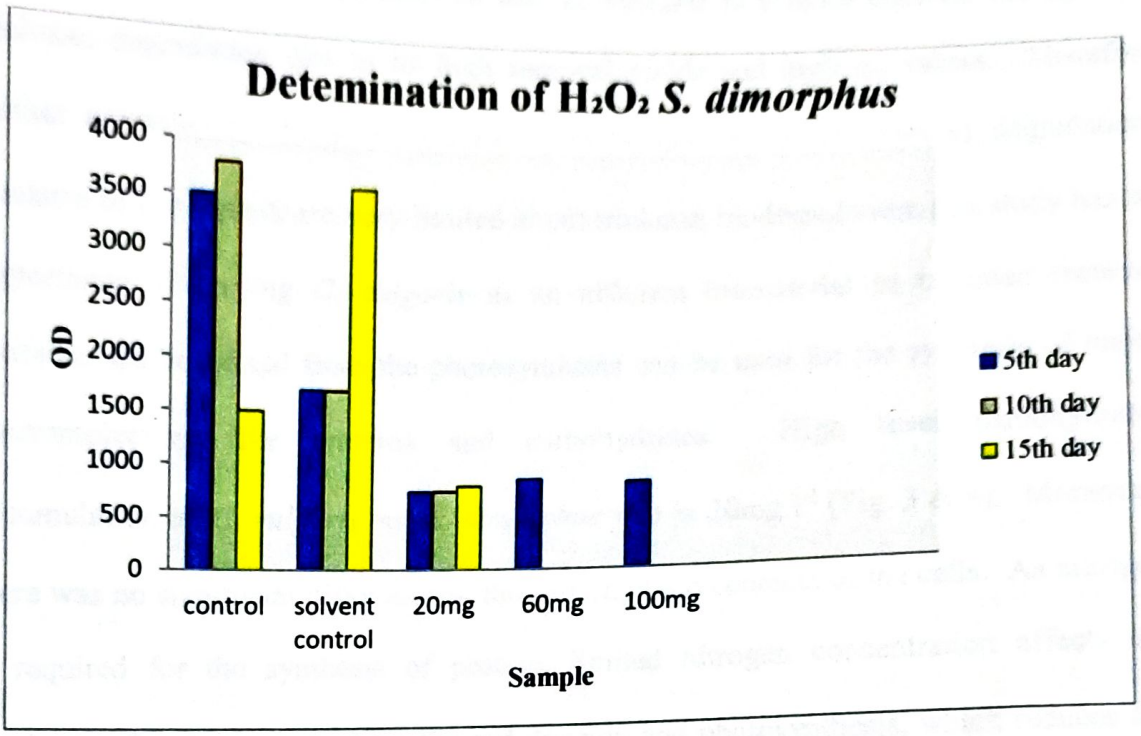


Fig. 7 Determination of H_2O_2 content of *S. dimorphus* after treatment with triclosan

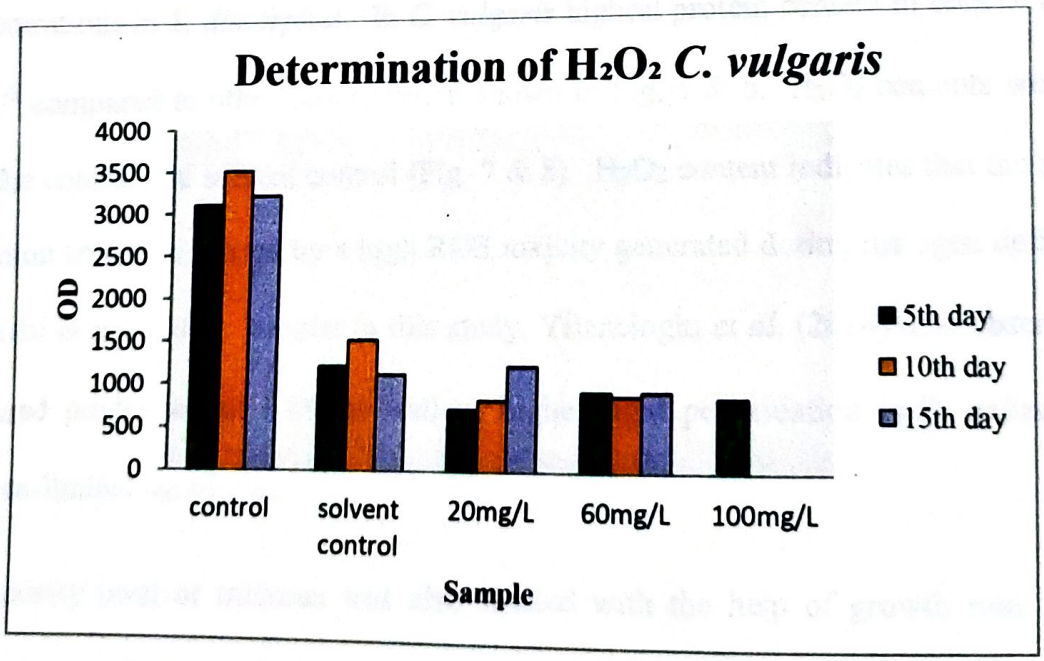


Fig. 8 Determination of H_2O_2 content of *C. vulgaris* after treatment with triclosan

from these series demonstrated that *C. vulgaris* and *S. dimorphus* degraded triclosan with a higher efficiency. Result showed that *C. vulgaris* is a more efficient bio-agent in triclosan degradation due to its high removal yields and high q_m values. Therefore further experiments should be needed to understand the mechanism of degradation. Because of the reports are very limited about triclosan biodegradation, this study has an importance of serving *C. vulgaris* as an efficient biomaterial in triclosan removal process. Carbon fixed from the photosynthesis can be used for the synthesis of major macromolecules like proteins and carbohydrates. High level carbohydrates accumulation by *C. vulgaris* and *S. dimorphus* was in 20mg l^{-1} (Fig. 3 & 4). Moreover, there was no significant difference in the carbohydrate contents of the cells. As nitrogen is required for the synthesis of protein, limited nitrogen concentration affects the synthesis of protein required for the cell division and photosynthesis, which reduces the cell growth rates and thus biomass production. In the present study highest protein content was observed in solvent control and 20 mg L^{-1} compared to the other concentrations in *S. dimorphus*. In *C. vulgaris* highest protein content in control and 20 mg L^{-1} compared to other concentration shown in Fig. 5 & 6. H_2O_2 contents were less than the control and solvent control (Fig. 7 & 8). H_2O_2 content indicates that the growth inhibition might be caused by a high ROS toxicity generated during nitrogen deficiency (Chokshi *et al.*, 2017). Similar to this study, Yilancioglu *et al.* (2014) also observed an increased production of ROS as well as higher lipid peroxidation in *D. salina* under nitrogen-limited conditions.

The toxicity level of triclosan was also studied with the help of growth rate. In the concentration of 60mg L^{-1} to 100 mg L^{-1} showed no prominent growth whereas it shows growth inhibition effects on *S. dimorpous* during the 6th day cultivation compared with the 20mg L^{-1}, solvent control and control (Plate 1 & 2). Triclosan biodegradation by

using an effective biomaterial and finding the cost effective method is still a challenging problem. The maximum triclosan biodegradation yield was achieved by *C. vulgaris* as 60 mg/L and *S. dimorphus* as 20 mg/L triclosan concentration but at 100 mg L^{-1} concentration both microalgae shown no growth but shown a bleached alga due to high level of toxicity. The results showed that *C. vulgaris* can be used as a potential bioaccumulator for triclosan biodegradation process up to high triclosan levels (60 mg L^{-1}) comparable to the *S. dimorphus*.

Therefore, understanding of metabolic pathway of pollutants by algae is useful for risk assessments. In this work, freshwater microalgae were applied for the investigation of biodegradation pathway of triclosan in aquatic environment. When triclosan was treated with the algal species, high removal rate was observed. It was detected that cellular uptake was the predominant mechanism for the depletion of triclosan by *C. vulgaris*, while biotransformation accounted for the elimination of triclosan by the other species (Wang *et al.*, 2013).

SUMMARY AND CONCLUSION

Triclosan biodegradation in aqueous solutions by using efficient, eco-friendly and economical methods is still a challenging problem. The aim of this study was to investigate if microalgae could be used as effective biomaterials for triclosan degradation from fresh water. Results showed that the highest biodegradation yield of triclosan was obtained at 20 mg L^{-1} and 60 mg L^{-1} by *S. dimorphus* and *C. vulgaris* respectively but that is more than the concentration of triclosan present in aquatic environments. Both the microalgae have certain potentiality which helps to overcome triclosan pollution, although *C. vulgaris* was better biomaterial when compared with *S. dimorphus*. The removal of pollutants from environment is an important issue because of their potential harmful effect on environment and human beings. The pollutants usually go though complex chemical and biological conversion in environments, which might induce the formation of more toxic and persistent by-products. *S. diimorphus* and *C. vulgaris* can accumulate or degrade the triclosan and convert into less toxic compounds. The major finding of this present study was triclosan pollution will definitely be reduced with the help of *C. vulgaris* and *S. dimorphus* in nearby future. Furthermore research should be needed to understand the level of degradation at large-scale level and also understand the degradation mechanism of triclosan.

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A COMPARATIVE PHARMACOGNOSTICAL AND PHYTOCHEMICAL EVALUATION OF TWO CISSUS QUADRANGULARIS L.

A dissertation submitted to

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affiliated to **MANONMANIAM SUNDARANAR UNIVERSITY**

in partial fulfilment of the requirements for the Degree of

Master of Science in Botany.

By

M. DHARSHINI
Reg No: 21APBO02

DEPARTMENT OF BOTANY

ST. MARY'S COLLEGE (Autonomous)

THOOTHUKUDI -628001

2022-2023

CERTIFICATE

This is to certified that is this short term project work entitled dissertation entitled **A COMPARATIVE PHARMACOGNOSTICAL AND PHYTOCHEMICAL EVALUATION OF TWO CISSUS QUADRANGULARIS L.** submitted to **ST. Mary's College (Autonomous)** affiliated to **MANONMANIAM SUNDARANAR UNIVERSITY** in partial fulfilment of the requirements for the Degree of Master of Science in Botany and is a record of work done in the department of Botany **ST. Mary's College (Autonomous)** THOOTHUKUDI during the year 2022-2023.

GUIDE

Dr. Tmty. S. Beulah Jerlin
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DECLARATION

I do here by declare that this dissertation entitled **A COMPARATIVE PHARMACOGNOSTICAL AND PHYTOCHEMICAL EVALUATION OF TWO CISSUS QUADRANGULARIS L.** Submitted by me in partial fulfilment for the award of the degree of **'Master of Science in Botany',** in the result of my original and independent work carried out under the guidance of **Dr. Mrs. S. Beulah Jerlin M.Sc, M.Phil., Ph.D.** Assistant Professor. Department of Botany, St.Mary's College (Autonomous) THOOTHUKUDI and it has not been submitted elsewhere for the award of any other degree.

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M. DHARSHINI

CONTENT

INTRODUCTION

The human plant intimate relation dates back to the origin of human on this planet. With the development of social sense in primitive men, their dependence on the plant resources increased, not only for food, but also for fodder, fuel, drug and shelter. In India there are about 550 tribal communities covered under 227 ethnic groups residing in about 5000 villages in different forest and vegetation types. Botanically derived medicinals have played a major role in human societies throughout history and prehistory (Singh *et al.*, 2010).

Mother nature has gifted divine medicinal plants to humans from her green remedies from plant kingdom to fight death from disease and cure themselves from ailments. It is up to us to explore, seek, search and reap the benefit of this treasure. India is endowed with a rich wealth of medicinal plants. These plants are the major collaborator in the formulation of Indian material medica (Joshi, N. and Joshi, K. 2021) .

The importance of plants is known to us well. The plant kingdom is a treasure house of potential drugs and in the recent years there has been an increasing awareness about the importance of medicinal plants. Drugs from the plants are easily available, less expensive, safe, and efficient and rarely have side effects. The plants which have been selected for medicinal use over thousands of years constitute the most obvious choice of examining the current search for therapeutically effective new drugs such as anticancer drugs , antimicrobial drugs , antihepatotoxic compounds (Yadav, R. N. S. and Agarwala, M. 2011).

India is a varietal emporium of medicinal plants and is one of the richest countries in the world in regard to genetic resources of medicinal plants. It exhibits a wide range in topography and climate, which has a bearing on its vegetation and floristic composition. Moreover, the agro-climatic conditions are conducive for introducing and domesticating new exotic plant varieties (Parekh, J. and Chanda, S. 2007).

The tribal tracts are the storehouses of information and knowledge on the multiple uses of plants. However, such traditional knowledge is rapidly disappearing. There is an urgent need to document this knowledge, as otherwise it will be lost forever. The knowledge of the use of natural plant products amongst our people is truly phenomenal (Mohan, V. R. 2008).

As per a report by World Health Organization (WHO), over 80% of the people of developing countries are relaying on the traditional medicines that are extracted from the plants for their primary health needs. Use of these traditional medicines for the preparation of modern medical preparations is indispensable and thus 'Phytomedicines' are a link between the traditional and modern medicine (Balamurugan, V *et al.,* 2019).

Knowledge of the relatedness of species, commonly characterized as a phylogeny, clarifies potential sources of chemical and medicinal compounds along with genetic material for agricultural and horticultural programmes (Timmons, S. A *et al.,* 2007).

In biology, morphology is the branch that deals with the form of living organisms. For plants, plant morphology or phytomorphology is the study of the physical form and external structure of plants, whereas plant anatomy is the study of the internal plant structure, mostly at the cellular/microscopic level (Carrillo-Lopez, A. and Yahia, E. M. 2019).

The comparative study of plant structure, morphology and anatomy, has always been the backbone of plant systematics, which endeavours to elucidate plant diversity, phylogeny and evolution. The second half of the 20th century has been a fascinating period in which systematics and structural studies greatly profited from new techniques and methods. (Endress, P. K *et al*., 2000).

Nature has been a source of medicinal agents since the beginning of human civilization. Medicinal plants or their derived material, have been widely employed in all

cultures, throughout the history, for the prevention and treatment of diseases. Recently there has been a tremendous increase in the use of plant-based health products in developing as well as developed countries resulting in an exponential growth of herbal products globally. Plants continue to serve as possible sources for new drugs and chemicals derived from various parts of plants. (Ashwathy, G *et al.,* 2020).

Plant materials are a rich source of biologically active metabolites. The active secondary metabolites produced by some of these plants have potential bioactive compounds of interest in the pharmaceutical industry. Plant-derived substances have recently become of great interest due to their applications as drugs, as model compounds for drug synthesis or as intermediates for synthetic drugs (Morsy, N. 2014).

Herbal medicines are a valuable and readily available resources for primary health care and complementary health care system, undoubtedly the plant kingdom still holds many species of plants containing substances of medicinal value that have yet to be discovered, though large number of plants are constantly being screened for their antimicrobial effect, these plant may prove to be a rich source of compounds with possible antimicrobial activities, but more pharmacological investigations are necessary (Pandey, A *et al.*,2011).

Naturally occurring substances are of plants, animals and mineral origin. They are organic substances and could be obtained in both primary and secondary metabolic process; they also provide a source of medicine since the earliest time. The plant kingdom has proven to be the most useful in the treatment of diseases and they provide an important source of all the world's pharmaceuticals. The most important of these bioactive constituents of plants are steroids, terpenoids, carotenoids, flavanoids, alkaloids, tannins and glycosides. Plants in all facet of life have served a valuable starting material for drug development (Ajayi, I. A *et al.,* 2011).

Phytochemicals (from the Greek word phyto, meaning plant) are biologically active, naturally occurring chemical compounds found in plants, which provide health benefits for humans further than those attributed to macronutrients and micronutrients (Hasler and Blumberg, 1999). They protect plants from disease and damage and contribute to the plant's colour, aroma and flavor. In general, the plant chemicals that protect plant cells from environmental hazards such as pollution, stress, drought, UV exposure and pathogenic attack are called as phytochemicals (Mathai, 2000).

Recently, it is clearly known that they have roles in the protection of human health, when their dietary intake is significant. More than 4,000 phytochemicals have been cataloged and are classified by protective function, physical characteristics and chemical characteristics (Meagher and Thomson, 1999).

Phytochemicals are certain non-nutritive plant chemicals which have some disease preventive properties. They are not required by the human body for life sustainence, but they offer protection against pathogens. There are different ways in which a phytochemical can work. It can act as an antioxidant and protect cells against free radical damage, eg. polyphenols, carotenoids etc. It can stimulate certain enzymes, thereby reduce risk for breast cancer, eg. terpenes. It may act as an anti-bacterial and hormonal-stimulant component. It may even act as binders which may prevent the adhesion of pathogens to the human cell walls. Phytochemicals are already a part of our diet through vegetables and fruits (Mathew, B. B *et al.*, 2012).

Plant materials remain an important resource to combat serious diseases in the world. The traditional medicinal methods, especially the use of medicinal plants, still play a vital role to cover the basic health needs in the developing countries. The medicinal value of these plants lies in some chemical active substances that produce a definite physiological

action on the human body. The most important of these bioactive constituents of plants are alkaloids, tannin, flavonoid and phenolic compounds (Thirumurugan, K. *et al.,* 2010).

Natural drugs have been a part of the evolution of human, healthcare for thousands of years. Nowadays nearly 88% of the global populations turn to plant derived medicines as their first line of defence for maintaining health and compacting diseases. One hundred and nineteen secondary plant metabolites derived from plants are used globally as drugs, 15% of all angiosperms have been investigated chemically and of that 74% of pharmacologically active plant derived components were discovered (Raja. *et al.,* 2009).

The test used in phytochemical screening should be simple, standard and one should be aware of false positive result and hence the need for carrying out confirmatory tests. The chemical constituents that are of medicinal importance are mainly the secondary metabolites, and the examination of the chemical constituents of the plant can only reveal those compounds that have accumulated to some extent at a specific organ of a given plant. The presence or absence of such compounds depends largely on the extent of accumulation, the amount of plant material used and the analytical method employed (Harborne, 1973)

The people consume several plant or plant derived formulations to cure helmintic infections. Plants are endowed with various phytochemical molecules such as vitamins, terpenoids, phenolic acids, lignins, stilbenes, tannins, flavonoids, quinones, coumarins, alkaloids, amines, betalains and other metabolites which are rich in antioxidant activity. Studies have shown that many of these antioxidant compounds posses antitumor, anti-inflammatory, anti-atherosclerotic, anti-mutagenic, anticarcinogenic, anti-bacterial, anti-viral and anti-parasitic activities (Rajesh, K. *et al.,* 2014).

Polyphenols are a large groups of natural compounds widely distributed in variety of plants. They are known to have antioxidant properties with potential health benefits (Subhashini, R. *et al.,* 2010).

Medicinal plants are the plants or their parts used for the health care. They probably constitute a single larger functional group of the plants globally. According to an estimate, 120 or so plant based drugs prescribed for use through the world come from just 95 plant species (Lewington, 1990).

Medicinal plants have bioactive compounds which are used for curing of various human diseases and also play an important role in healing. Phytochemicals have two categories i.e., primary and secondary constituents. Primary constituents have chlorophyll, proteins, sugar and amino acids. Secondary constituents contain terpenoids and alkaloids. Medicinal plants have antifungal, antibacterial and anti-inflammation activities (Wadood, *et al.*, 2013).

The evaluation of all the drugs is based on phytochemical and pharmacological approaches which leads to the drug discovery referred as natural product screening (Foye et al., 2008). Any part of the plant may contain active components such as bark, leaves, flowers, roots, fruits and seeds (Gordon and David. 2001).

Phytochemical studies of *Cissus quadrangularis* have shown the presence of various versatile constituents such as flavanoids, triterpenoids, Vitamin C, stilbene derivatives and many others, e.g. resveratrol, piceatannol, pallidolperthenocissin and phytosterols. Out of which ascorbic acid, triterpene, β-sitosterol, ketosteroid, two asymmetrical tetracyclic triterpenoids and calcium were identified as major constituents of this plant. It contains high amount of Carotene A, anabolic steroidal substances and Calcium (Mishra, G, *et al.*, 2010).

Cissus quadrangularis, for example, is used by common folk in India to hasten the fracture healing process. In Cameroon, the whole plant is used in oral re-hydration, while in Africa and Asia the leaf, stem, and root extracts are utilized in the management of various ailments. Phytochemical analyses of *Cissus quadrangularis* reveal a high content of ascorbic

acid, carotene, phytosterol substances and calcium, and there have also been reports of the presence of β-sitosterol, δ-amyrin and δ-amyrone . All these components have potentially different metabolic and physiologic effects. Although researchers have investigated several uses of *Cissus quadrangularis,* its potential application against metabolic syndrome has not yet been reported (Oben, J, *et al.,* 2006).

Cissus quadrangularis is a traditional medicine usually said to come from Ayurveda but appears to have a wide range of locations which have used it medicinally due to its growing in numerous locations. Traditionally it was mostly used in treatment of female disorders (menopause, libido, and menstrual disorders) and treating bone disorders (increasing bone mass or accelerating fracture healing rates) which gives it the traditional name of the 'Bone Setter' (Hadjod), some other traditional usages are in regards to its supposed antiulcer properties, Antihemorrhoid properties, pain relieving properties and wound healing properties.

Phytochemical studies on methanol extract revealed the presence of triterpenes including α- and β- amyrins, β sitosterol, ketosteroids, phenols, tannins, carotene and vitamin C. Seven alicyclic lipids constituents have also been reported from *Cissus quadrangularis.* unsymmetric tetracyclic triterpenoids such as d-amyrin, onocer-7-ene-3a, 21b-diol, damyrone and 3,3',4,4'-tetra hydroxy biphenyl, 3,3',4,4'- tetrahydroxybiphenyl have been isolated from plant and were quantitatively determined by HPTLC and HPLC methods in samples collected from five different geographic zones of India (Siddiqua, A. and Mittapally, S. 2017).

Cissus quadrangularis is a perennial herb with medicinal properties distributed throughout the tropical world. It is one of the most frequently used medicinal plants in India. It is believed that the plant is native to India, Sri Lanka, Malaysia, Java and West Africa. This plant is studied for its phytochemical constitution, pharmacological activities and toxicological evaluation. It is used for bone healing Reddy *et al.*, (2017), Shirwaikar *et al*.,

(2003) and Singh *et al.,* (2013). Ayurveda prescribe this plant for several medicinal ailments. *Cissus quadrangularis* synonym Cissus succulent popularly known as horjora in Hindi and pirandai in Tamil belongs to the family Vitaceae. The plant is widely seen in tropical forest regions of Asia and Africa Chopra *et al.,* (1975), Oben *et al.*, (2006) and Mehta *et al.,* (2001).

The current study focuses on morphological, anatomical, and biochemical characteristics to distinguish the morphovariants of *Cissus quadrangularis* L. These investigations are done to distinguish between species, subspecies, and morphotypes within a species. Plant morphological characteristics are used by plant biologists to identify, categorise, and describe various plant species as well as to determine how they differ or are similar to one another. The identification of plant species can also be done using anatomical traits. The use of biochemical traits is also crucial for identifying different plant species.

SCOPE AND OBJECTIVES

Plants have long been a vital source of organic compounds for preserving human health, especially in the last ten years with more in-depth research on natural remedies. Several disorders have been treated with *Cissus quadrangularis,* a popular traditional medicine. It is used to cure conditions including anorexia, diabetes, peptic ulcer, haemorrhoids, gout, syphilis, venereal illnesses, leucorrhea, worm infestation, and excessive cholesterol. In India's north-eastern states, it is also utilised as a supplement for muscle building; the stem is a vegetable. It is used in the Siddha School of medicine to treat gonorrhoea, piles, asthma, and cough in addition to being an anti-aging herb. (Siddiqua, A., and Mittapally, S. 2017). Hence the present investigation was undertaken with the following objectives.

- To identify the morphological characters.
- To evaluate and compare the anatomical characters of *Cissus quadrangularis* Variant I and Variant III.
- To perform qualitative analysis of phytochemical in aqueous extracts of two variants of *Cissus quadrangularis* which can further lead provide a beneficial information towards the quality of the drug.

REVIEW OF LITERATURE

Cissus quadrangularis L. is a succulent plant of family Vitaceae usually found in tropical and subtropical xeric wood. It is a beefy desert plant like liana generally utilized as typical nourishment in India. It finds application in medicine. Experts have made efforts to test the plant's suitability using rational analysis. Some of the pharmacological use of the plant are linked to cell reinforcement, free radical search, hostile to microbials, bone regeneration, ulceration, pain relief, mitigation and diuretics. Hence, we document the available pharmacological data on *Cissus quadrangularis* L. in the literature for further use (.Jaganath *et al*., 2020).

Cissus quadrangularis is a succulent vine belongs to Vitaceae family is widely distributed throughout tropical and subtropical regions of the world and used frequently to various disorders. The plant has been reported to contain flavonoids, triterpenoids, phytosterols, glycosides and rich source of calcium. This study aims to bring a systematic review of *C. quadrangularis* in various pharmacological mechanisms. Evidence from the previous studies suggested the efficacy of *C. quadrangularis* with antimicrobial, anti-diabetic, anti-inflammatory, anti-obesity, anti-oxidant, bone turnover, cardiovascular and hepatoprotective activities. In conclusion, *Cissus quadrangularis* appears worthy of pharmacological investigations for new drug formulations (Sadiya *et al*., 2020).

The comparative study of plant structure, morphology and anatomy, has always been the backbone of plant systematics, which endeavours to elucidate plant diversity, phylogeny and evolution. The second half of the 20th century has been a fascinating period in which systematics and structural studies greatly profited from new techniques and methods (Peter K *et al.*, 2012).

Morpho-Anatomical and Physiological Responses Can Predict the Ideal Period for the Transplantation of Hydroponic Seedlings of *Hymenaea courbaril*, a Neotropical Fruit Tree. (Daniele *et.al*., 2020).

The smaller flow of nutrients and water from the culture medium influenced the morphogenesis of the parenchyma tissues. The palisade parenchyma thickness declined, mainly in the plants grown with 100 and 200 μM of Cd. The reduced thickness of this tissue can be related to the smaller translocation of water, which interferes with cell expansion (Silva-Cunha *et al*., 2021).

However, high-throughput methods for assessing internal anatomy remain elusive, precluding the widespread inclusion of internal anatomy in many modern -omics-level studies (Yadav *et al.,* 2021).

Morpho-Anatomical traits and soluble sugar concentration largely explain the responses of three deciduous tree species to progressive water stress (Jonathan *et.al*., 2021).

Morpho-Anatomical Feature and Phytochemical Assessments of *Lasia spinosa* (L) Thwaites. (Arya Lakshmi *et.al*., 2021).

Morphological, Phytochemical and FTIR spectroscopy analysis of Portulacaceae members were studied. The result of preliminary phytochemical screening indicated that leaf and stem of both species of Portulacaceae were free from steroids. Moreover, quantitative estimation of phytochemicals also exhibited that leaf and stem of both species. Secondary metabolites, which are abundant in plants and have fascinating biological activities, are an important source with a variety of structural arrangements and properties. They have a rich source of protein and have high antioxidant (Beulah and Santhiya 2022).

The qualitative and quantitative distribution of these metabolites differs from plant to plant and part to part. Alkaloids found in low concentrations relative to the phenolic compounds are offset by their high biological potency in vegetative tissues. Besides this,

alkaloids are found in higher concentration in storage tissues (roots, fruits and seeds) as compared to the green leaves (Walton and Brown, 1998).

Phytochemicals are knows as secondary plant metabolites and have biological properties such as antioxidant activity antimicrobial effect, modulation of detoxification enzymes, stimulation of immune system, decrease of platelet aggregation and modulation of hormone metabolism and anticancer property. There are more than thousand known and many unknown Phytochemicals. It is well known that plants produce these to protect themselves, but recent researches demonstrate that many Phytochemicals can also protect human against diseases (Rao 2003).

The chemicals are often referred to as "secondary metabolites" of which there are several classes including alkaloids, flavonoids, Coumarins, glycosides, polysaccharides, phenols, tannins, terpenes and terpenoids (Okwu, 2004).

Falodun *et al*., (2006) reported the occurrence of flavonoids, saponins, diterpenes and phorbolestersin in the aqueous and methanol extracts of *Euphorbia heterophylla.*

In recent years, secondary plant metabolites (phytochemicals), previously with unknown pharmacological activities, have been extensively investigated as a source of medicinal agents. Thus, it is anticipated that phytochemicals with adequate antibacterial efficacy will be used for the treatment of bacterial infections. Since time immemorial, man has used various parts of plants in the treatment and prevention of various ailments (Parekh, J and Chanda, S. 2007).

Kareru *et al*.,(2008) carried out the spectral analysis for saponins in the crude dry power of 11 plants and detected that *Albizia anthelmintica, senna singuaena, Mytenus senegalenisis, Senna didymomatrya, Terminalia brownie* and *Prunusa fricana*were likely to be bidesmosidic, oleanne type triterpenoids, while those detected in *Entada leptostachya* and *Rapanearhododendroides* might be monodesmosidic saponin

Raja *et al..,* (2009) reported the leaf extract of *Azadiracta indica* Krishna, K. L., Mruthunjaya, K., & Patel, J. A. (2009) conducted preliminary Phytochemicals, total phenolics and flavonoids content analysis in the methanol extract of *Justicia gendarussa*.

Jena *et al.* (2011) reported that presence of tannins, flavonoids, steroids, saponins and glycosides in leaves of *Pterospermum suberifolium.*

Antibiotics or antimicrobial substances like saponins, glycosides, flavonoids and alkaloids etc are found to be distributed in plants, yet these compounds were not well established due to the lack of knowledge and techniques. The phytoconstituents which are phenols, anthraquinones, alkaloids, glycosides, flavonoids and saponins are antibiotic principles of plants. From these phytoconstituents, saponins have been reported to exhibit hemolytic and foaming activity, antifungal, anti-inflammatory, fungistatic ,molluscidal (Ajayi, I. A., Ajibade, O., and Oderinde, R. A. 2011).

Plant derived substances have recently become of great interest owing to their versatile applications. The medicinal importance of plant is due to the presence of chemical constituents like alkaloids, glycosides, resins, volatile oils, gums, tannins etc. These compounds are synthesized by primary or rather secondary metabolism of living organisms (Yadav and Agarwala 2011).

Deshpande and Bhalsing (2011) made phytochemical investigation in the species, *Cassia obtusifolia, C. auriculata, Tephrosia purpurea, Helicteres isora* and *Centella asiatica.* Ethanolic extract of leaves, stem, seeds and roots of *Cassia obtusifolia* and *C. auriculata* revealed the presence of alkaloids, flavonoides, tannins and saponins in large amount. However in the species, *Helicteres isora* alkaloids only detected.

According to world health organization (WHO), any plant which contain substances that can be used for therapeutic purpose or which are precursor of chemo-pharmaceuticals semi synthetic new drugs is referred as medicinal plant. Medicinal plant

would be the best source to obtain a variety of drugs as the phytochemical are more specific. Phytochemical offer unique platform for structural diversity & biological functionality which is indispensable for drug discovery (Pandey, A., Kaushik, A., and Tiwari, S. K 2011).

Jitin A *et al.*, 2011 studied the phytochemicals present in various extract of aerial parts of *A. parviflora* and to determine the total phenolic and flavonoid content in ethanolic extract. Total phenol and flavonoid content was determined by folin-ciocalteu assay and aluminum chloride colorimetric assay respectively. Ethanolic extract showed the presence of alkaloids, sterols/ triterpenoids, flavonoids, tannins and coumarins. The phenolic and flavonoid content of ethanolic extract using gallic acid and rutin as standards was found out to be 1.09 ± 0.007mmgGAE/g and 1.163 ± 0.0208 mgRE/g respectively. The study showed significant amount of gallic acid and rutin equivalents were present in extract which may be responsible for valuable pharmacological property of extract. As phenolics and flavonoids are responsible for antioxidant activity of plant, present data implies that *A. parviflora* is a perfect candidate for in-vitro antioxidant activity and isolation of phytochemicals.

The phytochemical analysis revealed of significant amount of polyphenols and flavonoids (90% and 80%respectively). These findings suggest that *Theprosia purpurea* root extract posses prominent medicinal properties and can be exploited as natural drug to treat the disease anociated with free radical formation oxidative stress and xanthine, oxidase activity (Khobra 2011).

Mathew, B. *et al* (2012) reported the aqueous extracts of the pulp revealed the presence of carbohydrates, alkaloids, tannins, fixed oils, reducing sugars, proteins, cardiac glycosides, steroids, phytosterols, phenols and flavonoids, whereas the ethanolic pulp extracts showed only the presence of fixed oils, reducing sugars, cardiac glycosides, steroids, phytosterols, flavonoids and amino acids. On the other hand, the aqueous peel extracts showed the presence of carbohydrates, alkaloids, tannins, fixed oils, proteins, cardiac

glycosides, steroids, phenols and flavonoids and amino acids, whereas the ethanolic peel extracts revealed that they contained carbohydrates, saponins, tannins, fixed oils, cardiac glycosides, steroids, phytosterols, phenols and flavonoids.

Phytochemical analysis gave positive results for steroids, triterpinoids, reducing sugars, alkaloids, phenolic compounds, flavonoids and tannins (Vinoth, B. *et al.,* 2012).

Abbas *et al.*, (2012) studied the qualitative and quantitative phytochemical analysis of fifteen weed seed extracts. Alkaloids, saponins, glycosides, terpenoids, anthraquinine, steroids, flavonoids and tannins were detected from the weed seeds. Tannins and alkaloids were in high concentration. Tannins ranged from 7.97 to 24.17%, alkaloids 0.88 to 4.00%, saponins 0.54 to 1.29% and flavonoids 3.91 to 15.55%. Wheat weeds care medicinally important but their phytochemical potential needs to be further investigated.

Nilofer *et a.,l* (2013) studied the qualitative phytochemical analysis was done in rat tubers of six species *Dioscorea* found Meghalaya. The test conform the presence of various phytochemicals like flavanoids, saponins, steroids, cardiac, glycosides and terpenoids in two aqueous extract of methanol and ethyl acetate.

Chede (2013) observed the phytochemical screening of fruit pulp of *Citrus sinensis.* The aqueous as well as the ethanolic extracts of the pulp revealed the presence of carbohydrates, alkaloids, tannins, fixed oils and lipids, sugars, proteins, steroids and amino acids where as terpenoids are present only in the ethanolic pulp extracts.

Atiq Mehsud *et al*., (2013) studied the morphology and anatomy of seven most common weed species infesting agricultural and non-agricultural lands of rainfed area of Bannu region were investigated during 2012. The study included *Datura metel L., Euphorbia hirta L., Fagonia cretica L., Heliotropium europaeum L., Parthenium hysterophorus L., Solanum surattense Burm f. and Withania somnifera (L.) Dunal.* Due to some special morphological and anatomical features, the capacity of rapid absorption of water along with

minerals from the soil may be facilitated to compensate the rapid water loss, and thus can also be regarded as common xerophytes. Their morphological, anatomical and histological characteristics are suitable for their successful growth in rain-fed condition of the region.

Ajiboye*et al.*, (2013) studied to found out the presence of phytochemicals in the aqueous extracts of *Senecio biafrae* leaves of both qualitative and quantitative screening methods. In qualitative analysis, the phytochemical compounds such as alkaloids, saponin, tannin, phlobatannin, phenol, anthraquinoes, flavonoids, glycosides, steroids, terpenes, cardenolides, flavonoid and chalocones were determined in the sample aqueous extracts by using standard methods. Also, quantitative analysis of the important secondary metabolites such as alkaloids, phenolic compounds, flavonoids, saponins and tannins were tested in the sample extracts. Results concluded that the presence of these active compounds may be responsible for the medicinal purposes of the plan.

Mehta K and Jain B.K. (2013) examined the leaf extract of *Phyllanthus fraternus* contains tannins, saponin, terpenoid, steroid, lignins, niranthin, nirtetralin, phyltetralin and alkaloids like morphine and boldine.

Phytochemical studies carried out in various *Ranunculus* species revealed the presence of different secondary metabolites in them which indicates the existence of therapeutic uses of these species. The preliminary screening of *Ranunculus arvensis* showed the presence of glycosides, phenolics compounds, steroids, di and tri terpenes, coumarins and flavanoids (Ahlam Hachelaf *et al.*, 2013).

Cardioactive glycosides, tannins and saponins were reported for the first time in aerial parts of *R. muricatus* (Aslam, M.S *et al.*, 2013).

The medicinal plants are useful for healing as well as for curing of human diseases because of the presence of phytochemical constituents. Phytochemicals are naturally occurring in the medicinal plants, leaves, vegetables and roots that have defense mechanism

and protect from various diseases. Phytochemicals are primary and secondary compounds. Chlorophyll, proteins and common sugars are included in primary constituents and secondary compounds have terpenoid, alkaloids and phenolic compounds. Terpenoids exhibit various important pharmacological activities i.e., anti-inflammatory, anticancer, anti-malarial, inhibition of cholesterol synthesis, anti-viral and anti-bacterial activities. Terpenoids are very important in attracting useful mites and consume the herbivorous insects. Alkaloids are used as an aesthetic agents and are found in medicinal plants (Wadood, A *et al.,* 2013).

Medicinal plants contain some organic compounds which produce definite physiological action on the human body and these bioactive substances include tannins, alkaloids, carbohydrates, terpenoids, steroids and flavonoids .They are of great importance to the health of individuals and communities. Many of these indigenous medicinal plants are used as spices and food plants. Phenolics have been known to possess a capacity to scavenge free radicals. The antioxidant activity of phenolics is principally due to their redox properties, which allow them to act as reducing agents, hydrogen donors. Phenolics are especially common in leaves, flowering tissues and woody parts, such as stems (Soni, A., and Sosa, S. 2013).

Plants are a source of many drugs such as antispasmodics, emetics, antimicrobials, antipyretics, antidiarrheals, antioxidants, and antitumor agents. A large number of the plants are claimed to possess valuable properties in traditional medicine and are also used extensively by tribal people worldwide. Research has emphasized the evaluation and characterization of various plants and plant constituents against a number of diseases. Detection, estimation and extraction of the bioactive plant constituents have always been a challenging task (Morsy, N. 2014).

Lohith *et al.,* (2014) evaluated phytochemical constituents of pharmacological importance in the leaf extract of *Moullava spicata.*

In recent years, secondary metabolites (phytochemicals), previously with unknown pharmacological activities, have been extensively investigated as a source of medicinal agents. Thus, it is anticipated that phytochemicals with adequate anti-parasitic efficacy will be used for the treatment of various nematode infections. Considering the rich diversity of Indian medicinal plants including Pteridophytes, it is expected that, the screening of plant extract may be beneficial for humans and animal diseases. The aim of this study was to evaluate the phytochemicals from aqueous, ethanolic and petroleum ether extracts of four fern species (Rajesh K D *et al.*, 2014).

Shinde *et* al., (2014) studied some parameters such as morphological, microscopical, physicochemical evaluation, florescence analysis; preliminary phytochemical analysis, thin layer chromatographic study and antimicrobial potential of alcoholic extract of *P. grandiflora* were carried out. Macroscopically the leaves are fleshy leaves, watery, needle shape. Flowers are racemes form; fruits are ovoid with small black coloured seed. Chemomicroscopy revealed the presence of Rubiaceous stomata in leaf; Rosette calcium oxalate crystals and protoplast in mesophyll of leaf, cortex and pith of stem and root; pink colored cuticle of stem, collateral vascular bundle with lignified xylem, abundant of starch grains and mucilaginous cells in all aerial parts. Physicochemical evaluation used to determined standards showed a results with total ash, acid insoluble ash, water soluble ash, sulphated ash, ethanol soluble extractive, water soluble extractive, moisture content, swelling index and total crude fiber content in powder of stem. Preliminary phytochemical analysis revealed the presence of alkaloid, steroids, Triterpenoids, flavonoid, tannins, and carbohydrates. The total flavonoid content in alcoholic extract was found .The alcoholic extract of herbs showed significant inhibition of microorganism.

Silvia Netala *et al*., 2010 compared the structural features and physicochemical properties of three species of *Portulaca*. Different Parts of *Portulaca* were examined for

macroscopical, microscopical characters. Physicochemical, phytochemical and fluorescence were also analysed. The plants are succulent, prostrate herbs. Usually roots at the nodes of the stem. Leaves are opposite with paracytic stomata and characteristic Kranz tissue found in C-4 plants. Abundant calcium oxalate crystals are present in all vegetative parts of the plant. Quantitative determinations like stomatal number, stomatal index and vein islet number were performed on leaf tissue. Qualitative phytochemical screening revealed the presence of alkaloids, carbohydrates, saponins, steroids and triterpenoids.

Kabesh K *et al.*, (2015) investigate the phytochemical analysis and anti microbial activity of aqueous and methanol extracts of *Catharanthus roseus*. The enzymatic and non enzymatic (DPPH) method was employed to analyze the antioxidant property. Qualitative analysis of phytochemical screening reveals the presence of Alkaloids, Phenol, Saponins and Protein.

Medicinal plants and herbs have been proved to be of great importance to the health of the individuals and communities. In recent years, many scientific investigations of traditional herbal remedies for several diseases have been carried out and this has lead in the development of alternative drug and therapeutic strategies. Since the consumption of medicinal plants is increasing, it is interesting to use these plants as a supplement in food taking into account that these plants can present a significant amount of trace elements and other nutrients. *Cissus quadrangularis* is one such plant which is been studied for its medicinal properties like its useful in bone fractures, obesity and neuropharmacological effects (Prabhavathi ,*et al.,* 2016).

Vitaceae (the grape family) consists of about 14 genera and 900 species primarily distributed in tropical regions. The family is well known for grapes (species of *Vitis* L.) and is economically and ecologically important because many Species are major climbers in tropical and temperate Species forests. The grape family is characterized by leaf opposed

tendrils, which may be modified to form an inflorescence. The present investigation is an attempt to study taxonomical attributes on the genus of *Cissus* in Thrissur district. Taxonomic studies were conducted so as to classify the plants of the *Cissus* found in Thrissur district. *Cisssus vitiginea, Cissus quadrangularis, Cissus latifolia, Cissus repens and Cissus heynema* were studied. Observations of the taxonomical peculiarities solve the species identification problem in vitaceae. (Hari and Anto, 2016).

The chemicals that are produced by plants are called as phytochemicals. These are produced by the plant's primary and secondary metabolism. These phytochemicals are important for the plants to thrive or thwart other plants, animals, insects and microbial pests and pathogens. They also help plants and protect them from disease and damage caused by environmental hazards like pollution, UV, stress and draught. They are used as traditional medicine and as poisons from ancient days. Phytochemicals are not the essential nutrients they are rather than the essential nutrients because there is no proof for them to cause any possible health effects in humans are not still established. It is known that they have roles in the protection of human health. More than 4,000 phytochemicals have been catalogued and are classified by protective function, physical characteristics and chemical characteristics. The phytochemicals are generally classified into the following types; they include carotenoids and polyphenols which include phenolic acids, lignans. Which further have classifications like flavonoids are further classified into flavones, anthocyanins, isoflavones and flavanols (Balamurugan, V., Fatima, S., and Velurajan, S. 2019).

Ahmed.*et* al., (2019) examined the total phenolic, flavonoids content and antioxidant activities of *Citrullus colocynthis* L. and *Cannabis sativa* L. Phytoconstituents except terpenoids from *C. sativa* and *C. Colocynthis* leaves were reported while, in contrast, steroids, tannins and phenols were absent in *Citrullus colocynthis* roots. The methanol derived maximum phenolic contents from *C. sativa* and *C. Colocynthis* leaves were 36.42 and

37.69 mg gallic acid equivalent GAE/g respectively. However, total flavonoids registered from *C. Sativa* leaves and *C. Colocynthis* leaves and roots were 59.03, 50.58 and 43.32 mg quercetin equivalent QE/g respectively. Interestingly, *C. Colocynthis* leaves produced the highest flavonoids 119.63 mg QE/g using ethyl acetate extract. DPPH inhibition (%) was high in acetone 55.57, hexane 45.98 and distilled water 35.5% from *C. sativa*, *C. Colocynthis* leaves and roots respectively. Our findings suggest that studied plants contain phytochemicals, reasonable quantity of phenol and flavonoids content confer to the potential antioxidant activity responsible for insecticidal properties as safer alternatives of synthetic pesticides.

Phytomorphology is the study of the physical form and external structure of plants, whereas plant anatomy is the study of the internal plant structure, mostly at the cellular/microscopic level (Carrillo and Yahia, E.M 2019).

Muhammad A.T and Muhammad A (2020) studied the two members of Portulaca Family namely, *Portula capilosa* and *Portula caquadrifida Linn*. Dried twigs of *Portula capilosa* and *Portula caquadrifida* ground to fine powder and then analyzed using FTIR technique. Functional groups of phytochemicals were identified through FTIR spectral lines. Appropriate correlations of absorption peaks to medicinal compounds have been discussed. As a result, both herbs are found to be rich source of bioactive compounds like alkaloids, flavonoids, fatty acids, tannins, triterpenoids, amino acids and saponins.

Since ancient ages plants have served human beings as a natural source of treatments and therapies, amongst them medicinal herbs have gain attention because of its wide use and less side effects. In current scenario focus on plant research has increased throughout the world and a huge amount of evidences have been collected to show immense potential of medicinal plants used in various traditional systems. More than 15000 plants have been studied during the last 5 year period. Recently scientists are using these renewable

resources to produce a new generation of therapeutic solutions. Inspite of many synthetic compounds, the most efficient drugs available are directly or indirectly related with the plant kingdom. Many of the plant extracts have proven to posses pharmacological actions. Production and cost advantages of plant-made pharmaceuticals can allow more capital to be invested in research and development of new therapeutics, giving patients access to new drugs faster. This review highlights some of the phytochemical and pharmacological aspects of *Cissus quadrangularis* Linn. *Cissus quadrangularis*, a perennial climber widely used in traditional medicinal systems of India has been reported to posses bone fracture healing, antibacterial, antifungal, antioxidant, anthelmintic, antihemorrhoidal and analgesic activities. *Cissus quadrangularis* Linn. has been recognized as a rich source of carotenoids, triterpenoids and ascorbic acid and is proved to have potential for medical effects, including "Gastroprotective activity" in conjugation with NSAID therapy and in "Lipid metabolism and oxidative stress". Needless to say that versatile uses and various therapeutic activities has made the plant a valuable medicinal herb (Garima *et al*., 2010).

Cissus quadrangularis L. is a succulent plant of family Vitaceae commonly found in tropical and subtropical xeric wood. It is a fleshy, cactus-like liana widely used as a common food item in India. The plant is prescribed in the ancient Ayurvedic literature as a general tonic and analgesic, with specific bone fracture healing properties. The plant is believed to be useful in helminthiasis, anorexia, dyspepsia, colic, flatulence, skin diseases, leprosy, hemorrhage, epilepsy, convulsion, haemoptysis, tumors, chronic ulcers, swellings. Following various folk claims for cure of various diseases, efforts have been made by researchers to verify the efficacy of the plant through scientific biological screening. The scrutiny of literature revealed some notable pharmacological activities of the plant such as antioxidant, free radical scavenging, anti-microbial, anti -bacterial, bone healing, anti -ulcer, analgesic, anti -inflammatory and diuretic, presented in this review such that the potential use

of the plant either in pharmaceutics or as an agriculture resource can be evaluated (Unnati Shah, 2011)

Extracts and powders of *Cissus quadrangularis* have been used for many years to promote bone and tissues healing, as an analgesic, to treat infections, as an anabolic, and to promote weight loss and weight management. This review summarizes the studies in animals, humans and *in vitro* systems that have been conducted to determine the efficacy and safety of various *Cissus* preparations. Animal and *in vitro* studies provide support for the use of *Cissus* in promoting bone fracture healing and as an anti-osteoporotic. Several human studies support the use of *Cissus* extracts in weight management. No studies have been conducted demonstrating that *Cissus* exhibits anabolic and body building activities. Based on studies to date, *Cissus* extracts appear to be exceedingly safe and free of adverse effects at the doses commonly used. A wide variety of chemical constituents have been isolated and identified from *Cissus* extracts, including steroids, flavonoids, stilbenes, iridoids, triterpenes and gallic acid derivatives (Sidney and Siddarth, 2012).

Cissus quadrangularis L. is a succulent plant of family Vitaceae usually found in tropical and subtropical xeric wood. It is a beefy desert plant like liana generally utilized as typical nourishment in India. It finds application in medicine. Experts have made efforts to test the plant' s suitability using rational analysis. Some of the pharmacological use of the plant are linked to cell reinforcement, free radical search, hostile to microbials, bone regeneration, ulceration, pain relief, mitigation and diuretics (Jaganath *et al*., 2020).

Cissus quadrangularis a succulent vine belongs to Vitaceae family is widely distributed throughout tropical and subtropical regions of the world and used frequently to various disorders. The plant has been reported to contain flavonoids, triterpenoids, phytosterols, glycosides and rich source of calcium. This study aims to bring a systematic review of *Cissus quadrangularis* in various pharmacological mechanisms. Evidence

from the previous studies suggested the efficacy of *Cissus quadrangularis* with antimicrobial, anti-diabetic, anti-inflammatory, anti-obesity, anti-oxidant, bone turnover, cardiovascular and hepatoprotective activities. In conclusion, *Cissus quadrangularis* appears worthy of pharmacological investigations for new drug formulations (Sadiya *et.al.*, 2020).

Cissus quadrangularis L. is a fleshy plant found in major parts of the world, especially in Asia, Africa, and a few other warm tropical regions. It is one of the common food items in India. Ayurveda uses the whole plant for digestive aid (Pachana) and directed as palliative and roborant. *Cissus quadrangularis* also serves as a good source of triterpenoids ascorbic acid, carotenoids, flavonoids, and steroids. *Cissus quadrangularis* is also used for various treatments like fracture healing, anti-ulcer, antihelmintic, antifungal, antihemorrhoidal, analgesic, antibacterial properties, etc. It also serves in the best way to treat various infirmities such as hemorrhoids, leprosy, epilepsy, dyspepsia, skin burns, dysentery, bowel complaints, to increase appetite, etc. (Camil and Lokesh, 2020).

Cissus quadrangularis L. is a perennial herb of the Vitaceae family and is utilized comprehensively as a medicinal herb in most tropical regions by various names. This herb is documented to possess a wide-ranging ethnomedicinal uses in malaria, fever, epilepsy, gout, piles, skin diseases, colic, etc. (Piyush *et al*., 2021).

Cissus quadrangularis L. belongs to the family Vitaceae, commonly called bone setter plant. There are three morphovariants that exist for the species. They show many differences in their morphological appearances. Hence, the present study was carried out to delineate two morphovariants (variant I and III) of *Cissus quadrangularis* L.. based on their morphological, anatomical and biochemical characters.

MATERIALS AND METHODS

Class :Polypetalae

Order : Vitales

Family : Vitaceae

Genus : *Cissus*

Species : *quadrangularis L.*

Common Name : Veldt grape

Vernacular Name: Pirandai

Cissus quadrangularis L. is a succulent perennial climber belongs to the family Vitaceae. It is an edible plant native to India and Africa. *C. quadrangularis L.* has three morphovariants based on the differences in their morphological characters. Variant I is characterized as square stem and Variant III is characterized as flat stem (Plate 1 and Plate 2).

Collection and Identification of Plant Material

Fresh and healthy plant parts of *Cissus quadrangularis L.* Variant I and Variant III were collected from the St.Mary's College Garden, Thoothukudi. The collected plant sample were identified by using Flora of presidency of madras Gamble, (2004). The taxonomic identities of these plants were confirmed by using Flora of Tamil Nadu and Karnatic. Mathew (1983).

Macroscopic Characters

The morphological characters such as stem shape and phyllotaxy of two variants were studied. Variations in leaf arrangement, composition, shape, margin, apex, base and venation were observed and noted. The unbranched tendrils are produced after and opposite to the leaves at each node in a continuous pattern. Plants climb over the substrates using the structural support of firm stems and by the twining of tendrils present at every node.

Anatomical Identification

Fresh plant parts such as Node, Internode, Leaf, Petiole and Tendril of the two Variants were taken free hand sectioned. They were stained with safranin and mounted in glycerin. Semi-permanent slides were prepared and observed under compound microscope. Photographs of the sections were taken under trinocular microscope in 45x magnification using TC Capture software.

Pattern and Distribution of Stomata

Pattern and distribution of stomata were studied by stomata peel method. The peels of the epidermis were stained with safranin and mounted in glycerin. Semi-permanent slides were prepared and observed under compound microscope.

Powder microscopy (Shehla Akbar *et al*., 2014)

The cross section of Node, Internode, Leaf, Petiole and Tendril of Variant I and Variant III were studied under compound microscope.

Reagents	**Observation**	**Characters**
Strong KOH Solution	Needle shaped potassium eugenate crystals	Eugenol
Dilute Acetic Acid	Insoluble	Calcium Oxalate Crystals
Dilute Hydrochloric Acid	Soluble	Calcium Oxalate Crystals
Sulphuric Acid	Calcium Sulphate crystal formation	Calcium Oxalate Crystals
Dilute Iodine Solution	Blue	Starch
Dilute Tincture Solution	Red on standing for 30 mins	Volatile oil
Alcoholic Picric Acid	Yellow	Aluerone grains

Preparation of plant sample extract

The cleaned, healthy plant materials were collected and cut into small pieces and grind into fine paste. Paste so obtained was used for extraction. 1gm of each sample plant material were soaked in 20 ml of distilled water and kept for 24 hours. Then it was filtered through the whatman no.41 filter paper. The extraction was carried out in the room temperature.

Preliminary Phytochemical Analysis

Phytochemical analysis of the Variant I and Variant III were determined as follow.

Test for Alkaloid

Hager's test

To 1 ml of each extract, 3 drops of 2% picric acid was added. Formation of orange colour indicates the presence of Alkaloid.

Test for Cardenolides

To few ml of each extract Fehling's solution was added. Appearance of red colour fades to brownish yellow indicates the presence of Cardenolides.

Test for Glycosides

To 0.5 ml of extract was dissolved in 1 ml of water and then aqueous sodium hydroxide solution was added. Formation of yellow colour indicates the presence of glycosides.

Test for Terpenoids

To 2 ml of extract was mixed with 2 ml of acetic acid. To these 2 drops of concentrated sulphuric acid is added. Deep red colour development showed the presence of terpenoids (Yadav *et al.*, 2014)

Test for Lipid

Solubility test

Take 1 ml of extract and shake vigorously. Formation of 2 layers indicates the presence of Lipid.

Test for Resin

Turbidity test

To 1 ml of extract add acetone. Then poured in distilled water. The turbidity indicates the presence of resin.

Test for Saponin

1 ml of extract was taken in a test tube and 5 ml of distilled water was added and vigorously shaken. A persistent froth that lasted for at least 15 minutes indicated the presence of saponins.

Test for Phenols (Harbone, 1973)

To 1ml of the extract, 2ml of distilled water was added which is followed by few drops of 10% aqueous ferric chloride. Appearance of blue or green colour indicate the presence of phenols.

Test for Protein

To 2 ml of extract, 2 ml of Biuret reagent was added. Then keep it in a water bath. The colour turns into violet indicate the presence of Protein.

Test for Flavonoids

To 2 ml of extact, few drops of 20% sodium hydroxide is added. The yellow colour is formed. Then few drops of 70% dilute Hydrochloric acid is added. The disappearance of yellow colour indicates the presence of flavonoids.

Test for Carbohydrate

Benedict's test

To 0.5 ml of extract add 0.5 ml Benedict's reagent, incubate in boiling water bath. Appearance of Brick red precipitate indicates presence of sugar.

Test for Coumarin (Yadav *et al.*, 2014)

To 1ml of extract, 1 ml of 10% sodium hydroxide was added. The presence of coumarins is indicated by the formation of yellow colour.

Test for Steroid

Salkowski test

To 1 ml of extract, add 2 ml of distilled water. To these 2 drops of 5% ferric chloride solution was added followed by concentrated sulphuric acid. Formation of reddish brown ring at the junction showed the presence of steroids.

Test for Tannins

The test solution was mixed with basic lead acetate solution. Formation of a white precipitate indicates the presence of tannins.

Plate 1. *Cissus quadrangularis L.* Variant I

Plate 2. *Cissus quadrangularis L.* Variant III

RESULTS AND DISCUSSION

Macroscopical Characters:

Cissus quadrangularis belongs to the family Vitaceae, Which is a perennial plant commonly known as Veldt Grape or Devil's backbone. *C. quadrangularis* is a climber. *C. quadrangularis* L. has three morphovariants based on the differences in their morphological characters; square-stemmed - Variant I, round-stemmed - Variant II and flat stemmed- Variant III. Morphologically they are dissimilar. The character of root were similar in three variants, they showed tap root system. The internodal length is also more or less similar.

The Variant I has thick, succulent, four winged stems that constrict at the nodes. Leaves are trilobed. In Variant II, stems are rounded by the presence of wingless stem. The Variant III has flattened stem and the leaves are three lobed. Phyllotaxy of Variant I and Variant III are alternate and distichous. Leaves are flanked at the base by a pair of stipules that are attached to and as wide as the elongating shoot.

Variations in leaf arrangement, composition, shape, margin, apex, base and venation were observed and noted (Table–1). The unbranched tendrils are produced after and opposite to the leaves at each node in a continuous pattern. Plants climb over the substrates using the structural support of firm stems and by the twining of tendrils present at every node.

A well-known medicinal plant called *Cissus quadrangularis* L. has the power to mend broken bones. For the species, there are three morphophovariants. The goal of the current study was to distinguish between species using morphological, anatomical, and biochemical traits. Just the stem and leaf have noticeable morphological changes, according to the examination of morphological features. This means that even if the other factors under study were nearly identical, both variations displayed disparities in their habits. According to

earlier studies, plants with the same physical characteristics can have different biochemical characteristics (Ansarali, 2018).

Anatomical Characters

Free hand section of Node, Internode, Petiole, Leaf and Tendril were prepared and observed under microscope. The anatomical study of variant I and III were carried out and the results observed are as follows.

Transverse Section of Stem of *C. quadrangularis*: (Table-2) (Plate- 3, 4, 5, 6, 7, and 8)

The cross section of the Node and Internode of variants showed four regions, namely epidermis, cortex, vascular bundle and pith.

Epidermis:

It consisted of a single-layered epidermis, which was surrounded by the cuticle.

Cortex:

The epidermis was followed by the cortex. The cortex was divided into an outer chlorenchymatous layer and an inner parenchymatous layer. Large mucilage cells were observed in the cortex. Patches of collenchyma embedded in the chlorenchymatous layer were seen at each corner.

Vascular Bundles:

The vascular bundles were conjoint, collateral, and open. Sclerenchymatous cap was present above the phloem of each bundle. The vascular bundles at the sides were smaller than those at the corners. Medullary rays were visible between the individual vascular bundles.

Pith:

The pith was comparatively large, occupying most of the area and was parenchymatous. Large mucilage cells were also seen in the medullary region.

Some Anatomical features are varies among the variants. In variant I, the stem has a dumbbell shaped outline with slight modifications. The four corners are extended forming the wing of the stem. It has four cellular zones as in the general pattern. Six vascular bundles were located at each of the four corners of the peduncle. In variant III, The outline of stem T.S was somewhat triangular in shape. The three corners of node were occupied by six vascular bundles each. Vascular traces of Leaf were arises and prominently seen in node.

Transverse Section of Leaf of *C. quadrangularis* Variant I (Table-3) (Plate-9)

The section showed characters like thin epidermal layer of small thick-walled squarish cell. The ground tissue in the adaxial part is small circular and thick- walled. In other region of the mid rib, the ground tissue was disintegrated leaving only small lobed parenchyma cell. The vascular system of the mid rib consisted of four radiating arms of vascular strands. Mucilage cell were observed in the mid rib. Vascular bundles were centrally located. The number of vascular bundles varied from basal of apical region of the leaf. 3-5vascular bundles are seen in the mid rib region. A collenchymatous cap was present over each vascular bundle. Vascular bundles are open, collateral and endarch. Stomatal openings were observed on the adaxial surface.

Transverse Section of Leaf of *C. quadrangularis* Variant III (Table-3) (Plate-10)

Transverse section of leaf of variant III was somewhat similar to variant I. The section had an outer epidermis followed by collenchymatous zone, parenchymatous ground zone and centrally located vascular zone. The epidermal cell in the abaxial surface and adaxial protuberance region were simillar in size, square to rectangular in shape 2-6 layer of collenchyma cells were seen in the abaxial surfaces. There were compactly arranged, small, polygonal to round shaped. Mucilage cells were also observed in the midrib region. Vascular bundles are centrally located. There was only a single vascular bundle observed in the mid rib

region. A collenchymatous cap was seen over each vascular bundle. Bundles are open collateral endarch. In both cases stomata is anomocytic.

Transverse Section of Petiole of *C. quadrangularis*: (Table-4) (Plate-11 and 12)

The transverse section of Petiole shows four region. A single layer of epidermis is noted. Next to epidermis the chlorenchymatous layer of cortex is observed. In which the vascular bundles were embedded. It is followed by the parenchymatous pith. Presence of sclerenchymatous cap is observed above each vascular bundles. The variant I and variant III are more or less similar. Variant I shows nine vascular bundles present in the center region. While variant III shows 14 vascular bundles.

Transverse Section of Tendril of *C. quadrangularis*: (Table-5) (Plate-13 and 14)

Periderm, vascular tissue, and pith were the three primary zones visible in a tendril cross section. Secondary thickening may be seen in the tendril cross section. The cortex and epidermis have disintegrated as a result of secondary thickening. They developed into phellum, phellogen, and phelloderm. Phellum was a dead tissue with several air-filled gaps. Three layers made up phellogen. Chlorenchyma cells made up the five to six layers of phelloderm, along with the 7 to 12 vascular bundles. Medullary rays, secondary xylem, and secondary phloems were all developed as a result of secondary thickening. Pith was a parenchymatous tissue that was relatively big. The variant I and variant III are more or less similar.

As the morphological characters showed little differences, we conducted the anatomical studies of stem, aerial root and leaf. The anatomical studies of the stem revealed that significant difference in the orientation of vascular bundles. In Variant I there is a peripheral layer of vascular bundles, in addition the corners were occupied by the four vascular bundles each. In case of variant III the corners were occupied by three vascular bundles each. This indicates that the difference was observed among the two variants studied.

Earlier works reported that anatomical differences can be used to distinguish the medicinal plant varieties (Josiane *et al*., 2013).

The study of aerial root (tendrils) revealed that no significant difference in the orientation of different tissues except variant I with a comparatively larger pith. In the present investigation, the anatomical characters of the leaf were also analyzed. The leaf anatomy showed significant difference ie., in variant I, the midrib region carried 5 vascular bundles, however in variant III, the midrib region carries 1 vascular bundle. Hence, this can be used to distinguish two plants. Earlier workers reported that anatomical characters can be used as a key to distinguish different morphotypes (Paweena, 2017).

QUALITATIVE PHYTOCHEMICAL SCREENING

The term 'phytochemical' is reserved for those plant chemical that have a beneficial effect on human- health but are not essential from the point of view of nutrition. A medicinal herb considered to be a chemical factory as it contains a multitude of chemical compounds like alkaloids, glycosides, saponins, resins, oleoresins, oils. Antifungal activity of medicinal plants is mainly due to the presence of phyto chemicals like alkaloids, glycosides, phenols, tannins and flavonoids (Sarojini *et al*., 2011). Moreover, phytochemical screening of the drug is signified for proper identification, which further exerts importance on therapeutic activity of the medicinal plant.

Therefore the current study was to determine whether there were any presence of preliminary phytochemicals in the whole plant aqueous extract of *Cissus quadrangularis*. Presence and absence of certain important compounds in an extract is determined by colour reactions of the compounds with specific chemicals which act as dyes. This procedure is a simple preliminary pre- requisite before going for detailed phytochemical investigation. Different chemical compounds were detected in plant extract of *Cissus quadrangularis* Variant I and Variant III and were presented in Table-6. The results of preliminary

phytochemical screening indicated that the highest concentrations of flavonoids, steroids, and tannins were found. Absent were lipids, resin, and cardenolides. The presence of coumarin, alkaloids, terpenoids, phenols, and carbohydrates was moderate. There were extremely few glycosides, saponins, and proteins.

MICRO CHEMICAL TEST:

The micro chemical test of variant I and III were analysed and the results are shown in Tables 7 and 8. Crystals of calcium oxalate were found throughout the body. The stem contains eugenol and starch.

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Table: 1 Morphological characters of *Cissus quadrangularis* L.

Characters	**Variant I**	**Variant III**
Habit	Climber	Climber
Duration	Perennial	Perennial
Stem Shape	Quandrangular	Flat
Leaf Arrangement	Alternate	Alternate
Leaf Composition	Simple	Simple
Leaf Shape	Trilobed	Trilobed
Leaf Margin	Lacerate	Lacerate
Leaf Apex	Obtuse	Obtuse
Leaf Base	Lobate	Lobate
Venation	Pinnate	Pinnate

Table 2. Microscopic observation of *Cissus quadrangularis L.* Stem

Characters	Variant I	Variant III
Outline	Rectangular	Dumb bell
Epidermis	Made up of single layer surrounded by cuticle	Made up of single layer surrounded by cuticle
Cortex	• Divided into outer chlorenchyma and inner parenchymatous layer • Large mucilage layer were observed • Patches of collenchyma present at each corner	• Divided into outer chlorenchyma and inner parenchymatous layer • Large mucilage layer were observed • Patches of collenchyma present at each corner
Vascular Bundles	Collateral, Open and Endarch	Collateral, Open and Endarch
Pith	Parenchymatous	Parenchymatous
Leaf Primordia were observed in the nodal region		

Table 3. Microscopic observation of *Cissus quadrangularis L.* Leaf

Characters	Variant I	Variant III
Epidermis	• Made up of single layer • Small, thick wall, quadrangular cells.	• Made up of single layer • Small, thick wall, quadrangular cells.
Mesophyll	Made up of Chlorenchymatous cells	Made up of Chlorenchymatous cells
Vascular Bundles	• Collateral, Open and Endarch • Collenchymatous cap seen above each vascular bundles	• Collateral, Open and Endarch • Collenchymatous cap seen above each vascular bundles
Stomata	Anomocytic type of stomata were observed in the adaxial surface.	Anomocytic type of stomata were observed in the adaxial surface.

Table 4. Microscopic observation of *Cissus quadrangularis L.* Petiole

Characters	Variant I	Variant III
Epidermis	Made up of single layer	Made up of single layer
Cortex	Made up of Chlorenchymatous cells	Made up of Chlorenchymatous cells
Vascular Bundles	• Collateral, Open and Endarch • Collenchymatous cap seen above every vascular bundles	• Collateral, Open and Endarch • Collenchymatous cap seen above every vascular bundles
Pith	Parenchymatous	Parenchymatous

Table 5. Microscopic observation of *Cissus quadrangularis L.* Tendril

Characters	Variant I	Variant III
Periderm	• Phellum was a dead tissue with several air filled gaps. • Phellogen made up of 3 layers of thick walled rectangular cell • Phelloderm composed of 4 to 7 layers of chlorenchyma cells	• Phellum was a dead tissue with several air filled gaps. • Phellogen made up of 3 layers of thick walled rectangular cell • Phelloderm composed of 4 to 7 layers of chlorenchyma cells
Vascular Bundles	Collateral, Open and Endarch	Collateral, Open and Endarch
Pith	Parenchymatous	Parenchymatous

Table 6. Preliminary phytochemical screening of *Cissus quandrangularis* L.

Whole Plant Extract (Aqueous extract)

Bioactive Compound	Variant I	Variant III
Alkaloids	++	++
Cardenolides	-	-
Glycosides	+	+
Terpenoids	++	++
Lipid	-	-
Resin	-	-
Saponins	+	+
Phenols	++	++
Proteins	+	+
Flavonoids	+++	+++
Carbohydrates	++	++
Coumarin	++	++
Steroid	+++	++
Tannins	+++	+++

+++: highly present, ++: moderately present, +: Low, -: absent

Table 7. Micro chemical test for variant I

Reagents	Observation	Characters	Internode	Node	Petiole	Leaf	Tendril
Strong KOH Solution	Needle shaped potassium eugenate crystals	Eugenol	+++	++	+	-	-
Dilute Acetic Acid	Insoluble	Calcium Oxalate Crystals	+++	+++	+++	+++	+++
Dilute Hydrochloric Acid	Soluble	Calcium Oxalate Crystals	+++	+++	+++	+++	+++
Sulphuric Acid	Calcium Sulphate crystal formation	Calcium Oxalate Crystals	+++	+++	+++	+++	+++
Dilute Iodine Solution	Blue	Starch	-	-	-	-	-
Dilute Tincture Solution	Red on standing for 30 mins	Volatile oil	-	-	-	-	-
Alcoholic Picric Acid	Yellow	Aluerone grains	-	-	-	-	-

+++: highly present, ++: moderately present, +: Low, -: absent

Table 8. Micro chemical test for variant III

Reagents	Observation	Characters	Internode	Node	Petiole	Leaf	Tendril
Strong KOH Solution	Needle shaped potassium eugenate crystals	Eugenol	+	+++	++	+	-
Dilute Acetic Acid	Insoluble	Calcium Oxalate Crystals	+++	+++	+++	+++	+++
Dilute Hydrochloric Acid	Soluble	Calcium Oxalate Crystals	+++	+++	+++	+++	+++
Sulphuric Acid	Calcium Sulphate crystal formation	Calcium Oxalate Crystals	+++	+++	+++	+++	+++
Dilute Iodine Solution	Blue	Starch	++	+++	-	-	-
Dilute Tincture Solution	Red on standing for 30 mins	Volatile oil	-	-	-	-	-
Alcoholic Picric Acid	Yellow	Aluerone grains	-	-	-	-	-

+++: highly present, ++: moderately present, +: Low, -: absent

Plate 3. T.S of Variant I Node

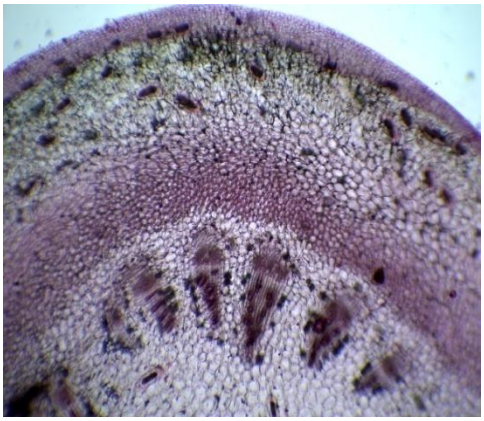

Plate 4. T.S of Variant III Node

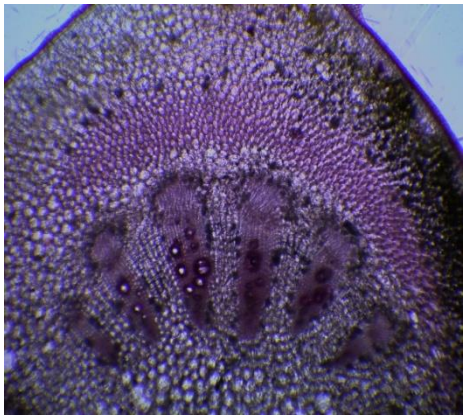

Plate 5. T.S of Variant I Leaf Primordia

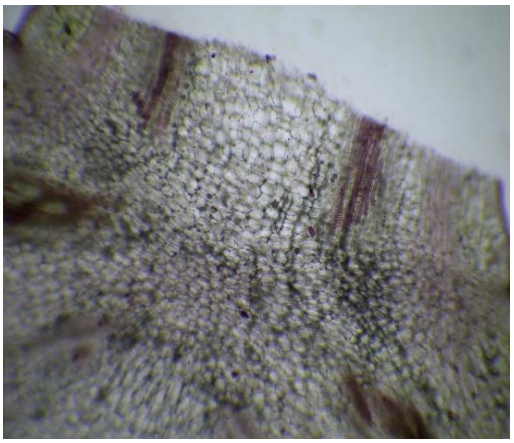

Plate 6. T.S of Variant III Leaf Primordia

Plate 7. T.S of Variant I Internode

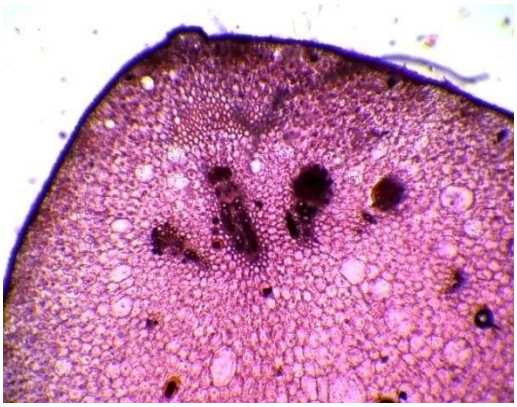

Plate 8. T.S of Variant III Internode

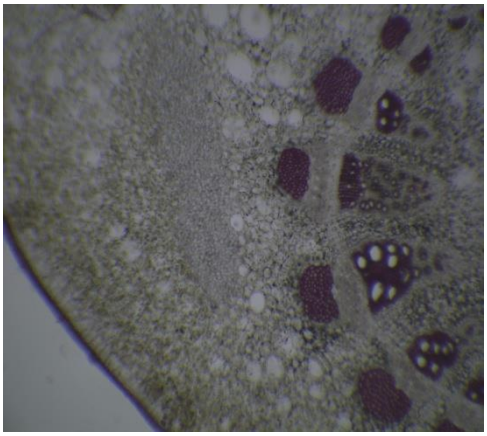

Plate 9. T.S of Variant I Leaf

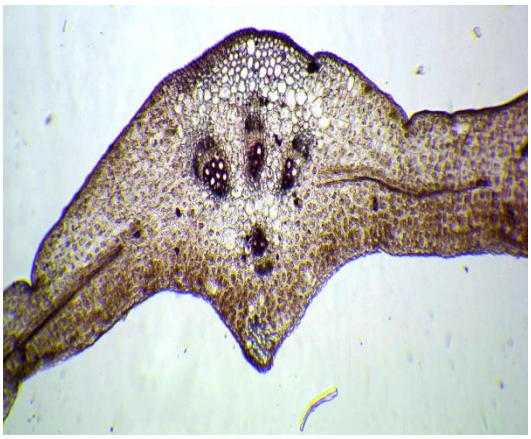

Plate 10. T.S of Variant III Leaf

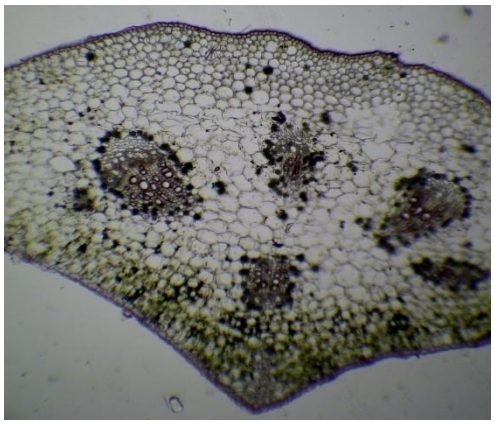

Plate 11. T.S of Variant I Petiole

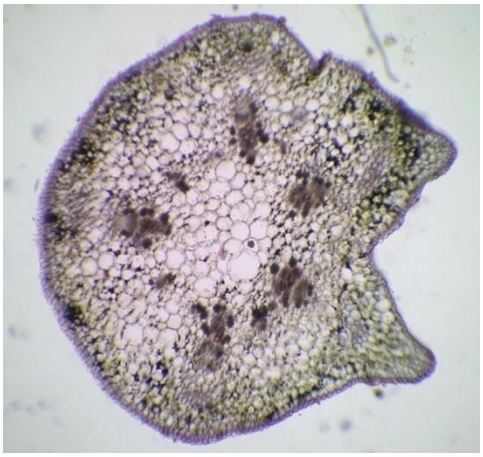

Plate 12. T.S of Variant III Petiole

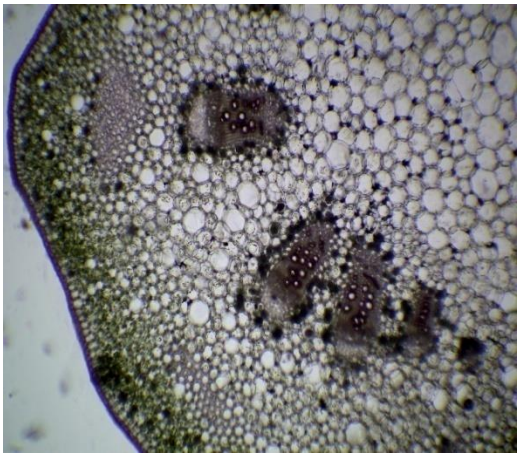

Plate 13. T.S of Variant I Tendril

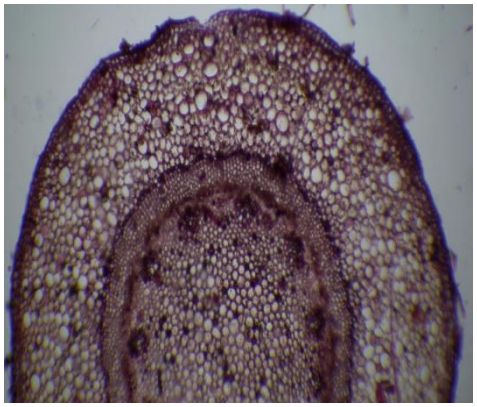

Plate 14. T.S of Variant III Tendril

SUMMARY AND CONCLUSION

Traditional recipes for treatment of physical and mental ailments exist in all major ancient civilizations of the world. Needless to mention that the root and stem extracts of the plant *Cissus quadrangularis* L. have therapeutic efficacy and are known to possess antioxidant, antimicrobial activity, and are routinely used to accelerate the process of bone fracture healing. The plant is considered as a versatile medicinal plant in both Ayurvedic and modern drug development areas for its valuable medicinal uses. It is a very rich source of some minerals, which are necessary for proper functioning of human body (Mishra, G *et al.*, 2010).

Cissus quadrangularis L. is a well- known medicinal plant that is used widely as a traditional medicinal plant. It is a succulent climber. It has the capacity to heal bone fractures. There are three morpho variants present in the species (Ashwathy G, 2020). The present study deals with the delineating the species based on their morphology, anatomy and phytochemical characters.

The Stem, Leaf, Petiole and Tendril of *Cissus quadrangularis* L. Variant I and Variant III were collected from the St.Mary's College garden, Thoothukudi for the current study.

The study of morphological characters revealed that only the stem of two variants shows significant morpological difference. Whereas the other plant parts did not show the variation among themselves. Morphological and anatomical features of plants are important in diagnosing the species. The results provide some pharmacognostic standards for the quality control of preparations from the plant in future.

The anatomical studies of the stem revealed that significant difference in the outline of the two variants. Both the variants shows anomocytic type of stomata in the adaxial

surface of leaf. The anatomical characters of Petiole and tendril were same. Thus it was concluded that the two variants were more or less similar in their characters.

Preliminary phytochemical like alkaloids, phenol, flavones, steroids, tannins, Coumarins, cardenolides, glycosides, terpenoids, lipid, resin, volatile oil, saponins, proteins and carbohydrates are quanlitatively screened.

The result of preliminary phytochemical screening indicates that *Cissus quadrangularis* L. Variant I and Variant III are a rich source of tannins, terpenoids, alkaloids, saponins, phenols, flavonoids, carbohydrate, coumarin and steroid.

The result of microchemical test indicates that it contains calcium oxalate, eugenol and starch.

SUSTAINABLE THRUST AREA

Cissus quadrangularis L. depicted an interesting fact that though the plant is a popular remedy for a variety of aliments and a range of formulations has been marketed, little effort have been made to verify its purity, quality and efficacy through scientific screening. In future study, the isolated principles from *C. quadrangularis L.* needs to be evaluated in scientific manner using specific experimental animal models and clinical trails to understand the molecular mechanism of action, in search of lead molecule from natural resource (Unnati Shah 2011).

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ASSESSMENT OF PHYSICO-CHEMICAL PROPERTIES OF DIFFERENT WATER SAMPLES OF KEELAVAIPPAR VILLAGE IN THOOTHUKUDI DISTRICT

A dissertation submitted to

ST. MARY'S COLLEGE (AUTONOMOUS), THOOTHUKUDI.

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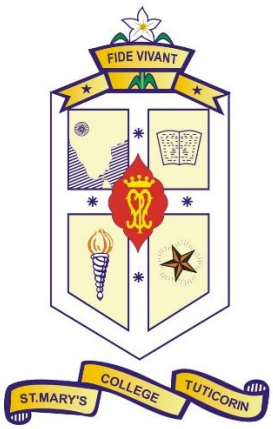
in partial fulfillment of the requirements for the Degree of

MASTER OF SCIENCE IN BOTANY

By

I. JERMINA

Reg.No. 21APBO03



DEPARTMENT OF BOTANY

ST. MARY'S COLLEGE (Autonomous)

THOOTHUKUDI -628001

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CERTIFICATE

This is to certify that this dissertation entitled, **"Assessment of Physico-Chemical Properties of Different Water Samples of Keelavaippar Village in Thoothukudi District"** submitted by **I. Jermina Reg. No. 21APBO03** to ST. MARY'S COLLEGE (Autonomous), THOOTHUKUDI in partial fulfillment for the award of the degree of **"Master of Science in Botany"** is done by her under my supervision. It is further certified that this dissertation or any part of this has not been submitted elsewhere for any other degree.

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DECLARATION

I do here by declare that this dissertation entitled, **"Assessment of Physico-Chemical Properties of Different Water Samples of Keelavaippar Village in Thoothukudi District"** submitted by me in partial fulfillment for the award of the degree of **'Master of Science in Botany'**, in the result of my original and independent work carried out under the guidance of **Dr. E. Daffodil D Almeida, M. Sc., SET, Ph. D.,** Assistant Professor, Department of Botany, St. Mary's College (Autonomous), Thoothukudi and it has not been submitted elsewhere for the award of any other degree.

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CONTENT

INTRODUCTION

INTRODUCTION

Water is the second most important need for life to exist after air. As a result, water quality has been described extensively in the scientific literature. The most popular definition of water quality is “it is the physical, chemical, and biological characteristics of water” (Spellman, 2013; Alley, 2007). Water quality is a measure of the condition of water relative to the requirements of one or more biotic species and/or to any human need or purpose (Shah, 2017; Tchobanoglous and Schroeder, 1985).

Significance of water

Water is a most abundant physical substance and transparent liquid on earth. In water molecule two atoms hydrogen and one atom oxygen are present. Water is the foundation of all form of life. Water is an essential natural resource for life of human beings, plants and animals on water planet. All processes of life are directly or indirectly connected to water therefore human beings cannot survive much longer without water, as water plays a central and critical role for every cell and organ system in the human body to function properly. Water is responsible for every activity in human body. Water is the source of life and it

is essential for all living forms and the environment health. It has the plentiful chemical substances on the Earth. It affects all form of life directly or indirectly (Ramesh et al., 2013). The presence of water determines the location and activities of human on the earth. Water is a basic medium of metabolic functions in all life on earth. Water is used in every cell of body to transport nutrients, oxygen, and wastes

to cells and organs. Water is a part of body's temperature regulating system. Water also plays an important role in the prevention of disease. Clean and freshwater must be free of contaminants to ensure wellness (Khan, 2011). The quality of water is just as important as the quantity. Water is used for drinking, bathing, washing, sanitation, irrigation, air condition, power and steam generation, fisheries, ecology, recreation etc. water is widely used in production of atomic energy, chemicals, ice, paper, and steel. Water is the major component in the body required for all body functions such as respiration, perspiration, growth, digestion, waste elimination, reproduction and a host of other important activities. Water is a basic element of social and economic infrastructure. The consumptive and non-consumptive classifications of water are important. Water used consumptively reduce the source at the point of appropriation and is not available for other uses; whereas non-consumptive water use does not reduce the source and the water is available for further reuse (Subba Rao, 1995).

Properties of water

The properties of water make it suitable for human beings to survive in differing weather conditions. Water is characterized by complex anamolus properties that differentiate it from other substances. Water is the universal solvent due to its polar nature. It dissolves a large number of different chemical substances. Its properties are as follows:

Physical properties of Water:

Water has many unique physical properties. It exists in all three physical states of matter: solid, liquid, and gas at atmospheric temperatures & pressures.

Water has a very high specific heat capacity and a high heat of vaporization. Both properties arise due to extensive hydrogen bonding between water molecules. Water's very high specific heat capacity is a good medium for spreading the earth's heat. Water has high density, which depends on the dissolved solids and temperature of the water. Water is physically unique because it is less dense as solid (ice) than as a liquid. The maximum density of liquid water occurs at 4ºC. Water has a high surface tension as compared to other liquids due to strong cohesion between molecules. Surface tension is responsible for capillary action, which allows water to move through the roots of plants. Water is the substance of which solid state can float on liquid state. Various properties of water like melting point, boiling point, viscosity, slow heating and cooling are result of intermolecular hydrogen bonding between water molecules. Water has high value of dielectric constant. Water also has an exceptionally high heat of vaporization. Water is able to dissolve most of gases like O_2, CO_2, N_2, H_2, SO_2 and NH_3. (Report of a River Pollution Survey of Scotland, 1975).

Chemical properties of Water

Water has many unique characteristics that make it ideal for life. Water is the chemical substance with chemical formula H_2O and bent shape. Water is a liquid at room temperature due to hydrogen bonding. In the water molecule both hydrogen atoms create a positive electrical charge while the oxygen atom creates a negative charge, therefore water molecules is polar in nature. Water is thermally stable but at higher temperatures dissociate into hydrogen and oxygen gases.

Water can ionize itself to a very small extent but in pure water the amounts of hydronium ions and hydroxide ions are equal (Stigter *et al.*, 2006). Hence pure water is neutral. Water is an amphoteric molecule it acts as acid as well as a base. Water oxidizes carbon to carbon monoxide behaving as an oxidizing agent while it reduces chlorine gas to hydrogen chloride acting as a reducing agent.

Biological properties of Water

Water is the universal solvent because it dissolves wide range of substances than other common solvents (Adnan and Iqbal, 2014). Water works as transporting biotic molecule, bio minerals, hormones and vitamins to different parts of animal and plant bodies. Water is significant for all the metabolic reactions essential for life to take place in solution in the cytoplasm of living cells. Water molecules are adhesive due to polar nature and therefore water sticks to other polar substances. This allows water to move upwards through the xylem of plants against gravity. Water dissolves oxygen gas from air which is necessary for aquatic life.

Major water compartments

Water compartments are a large area where water is stored. Water is stored in various global compartments. The major water compartments on earth are specified as follows:

1. **Oceans and seas:** The Ocean are a largest compartment of saline water that covers much of the Earth's surface. Oceans cover about 70% of the Earth's surface and the oceans contain roughly 97% of the Earth's liquid water. The Oceans and seas have great effect on the weather and temperature on earth. The Oceans moderate the Earth's temperature by absorbing incoming solar

radiation. The biomass in the oceans is over the 4 billion Tons.

2. **Glaciers, Ice and Snow:** Glaciers are slow moving rivers of ice. It takes a long time to from aglacier. Glacial ice often appears blue when it becomes very dense. Glacier's affect weather patterns, climate, and sea levels. Glacial ice is the largest reservoir of freshwater on Earth. Glaciers store about 75 percent of the world's total freshwater. Water in glaciers and ice caps is a small percentage of all water on the Earth.
3. **Groundwater:** Groundwaters are the hidden reserves that are connected to the surface water. Percolation is a hydrologic process in which water moves downward from surface water to groundwater. Groundwater is the subsurface water that fully saturates pores or cracks in soils. An aquifer is a geologic formation that contains sufficient saturated permeable material to yield significant quantities of water. Water added to aquifers naturally ater infiltrates into the soil. Water can be removed from aquifers bydrilling wells. Aquifers have been extremely important for livestock, irrigating crops, and as a source of municipal water (Singh, 2011).
4. **Rivers and streams:** Rivers are essential not only to humans, but to all form of life on the earth.Rivers and streams help to shape the features of the Earth. They help to drain rainwater and provide habitats for many species of plants and animals. Rivers make up only about 0.2 % of all the fresh water on Earth. Rivers and streams carry water, organisms and important gases to many areas. Rivers are providing the power for hydroelectric plants. Ultimately rivers and streams deposit that water in the ocean.

5. **Springs:** Spring is a natural situation where groundwater naturally emerges from theEarth's subsurface in a defined flow. Springs are the most obvious and interesting evidence of groundwater. Springs can discharge fresh groundwater into the beds of rivers or streams, and into the ocean. The temperature of spring water is related to the amount and rate ofgroundwater flow.

6. **Ponds and Lakes:** A pond is a small area of fresh water. It is different from a river or astream because it does not have moving water. The bottom of pond is usually covered with mud and Plants grow along the pond edge. Some ponds are formed naturally and some other ponds are man-made. Pond is areservoir of rainwater. Pond is smaller than lake and lake is deeper than a pond. Lakes are inland bodies of slowly moving water. Lakes are varied in terms of origin, occurrence, size, shape, depth and other features. Most lakes on Earth were formed by glacier activity. Lakes can be very deep or shallow. Lakes get water from precipitation, from rivers and streams and from underground water.

7. **Wetlands:** Wetland is a place where the land is covered by water, either salty or fresh.These are some of the most productive habitats on the Earth. Wetlands are variable and dynamic water bodies where water covers the soil. They are freshwater, brackish or saline, inland or coastal, seasonal or permanent, natural or man-made. Wetlands are most important ecosystems to human survival and development. Wetlands are a critical part of our environment. They protect our shores to reduce the impacts of floods, absorb pollutants and improve water quality. Names of different type's wetlands are swamp, marsh

and bog. Many animals use wetlands for all of their life- cycle. The most significant social and economic benefit that wetlands

8. **Atmosphere:** Atmosphere is the layer of gases that surrounds the Earth. The atmosphere is the smallest water reservoir of the earth. Water is located in the troposphere of the atmosphere. The water in the atmosphere presents onlya very small percentage of the total water on Earth.

In developing countries safe and sufficient drinkingwater supply is a crucial issue in rural and in many urban areas (Dahiya and Kaur, 1999). In rural areas groundwater is a reliable and finite source of water. The most common sources of water for irrigation and various purposes are surface water and groundwater. Ground water and surface water are interconnected. The surface water is present in the form of oceans, rivers, lakes, ponds and streams on the earth's surface and the groundwater present below the earth's surface in porous soils and rocks.

Water is polluted when it contains enough impurities to make it unfit for intended use. Water contaminations may be natural or human induced. Human activity affects the natural composition of groundwater. Use of contaminated water causes health hazards to people; therefore, it is important to check the activities that affect the quality and quantity of water. Groundwater is widely used as drinking purpose in rural area (Ravindra and Garg, 2005). Contaminated water can be unsuitable for various purposes and its remediation is difficult, time-consuming and expensive. It may be harmful for human health as well as Environmental health (Chatterjee *et al.*, 2009)

After many years of research, water quality standards are put in place to ensure the suitability of efficient use of water for a designated purpose. Water quality analysis is to measure the required parameters of water, following standard methods, to check whether they are in accordance with the standard.

SCOPE AND OBJECTIVES

SCOPE AND OBJECTIVES

Water plays essential role in all forms of life and it is the fundamental requirement for people. Water is not only used for drinking or cleaning but for recreation people seek water's edge, such as a solvent, cleanser, coolant, a compound from which every organism is created. Though the surface of the earth is mostly consisting of water, only a small part of it is usable, which makes this resource very limited. This precious and limited resource, therefore, must be used with prudence. Nowadays, water pollution has become global concern. Extraordinary urbanization terrified up water pollution. Water quality may be determined by chemical composition of water. If the chemical composition is in permissible limits, it is safe to use (Sudha and Sangeetha, 2017) Water is perhaps the most precious natural resource after air.

As water is required for different purposes, the suitability of it must be checked before use. Also, sources of water must be monitored regularly to determine whether they are in sound health or not. Poor condition of water bodies is not only the indictor of environmental degradation, it is also a threat to the ecosystem. In industries, improper quality of water may cause hazards and severe economic loss. Thus, the quality of water is very important in both environmental and economic aspects. Thus, water quality analysis is essential for using it in any purpose (Roy, 2019)

Literature overview reveals that no specific work has been done so far to evaluate the qualitative aspect of different water resources of Keelavaippar village, Thoothukudi District for domestic purpose. Hence, the present research work has

under taken to investigate the physicochemical characteristics of different water resources i.e., well water, bore water and lake water of this area with the following objectives

- Collection of water samples from different resources like well, bore and lake.
- Evaluation of physical characters like colour, odour, turbidity, pH, conductivity and total dissolved solids in well water, bore water and lake water.
- Analysis of chemical characters like total alkalinity, total hardness in all the collected water samples.
- Estimation of nitrate, chloride, fluoride, sulphate, calcium, magnesium, sodium and potassium of water samples from selected three water resources
- Investigation of presence of heavy metals such as iron and manganese in collected water samples.

AREA OF STUDY

AREA OF STUDY

Keelavaippar is a Village in Vilathikulam Block in Tuticorin District of Tamil Nadu State, India. It is located 31 KM towards North from District headquarters Thoothukudi. It is near to Bay of Bengal. There is a chance of humidity in the weather.

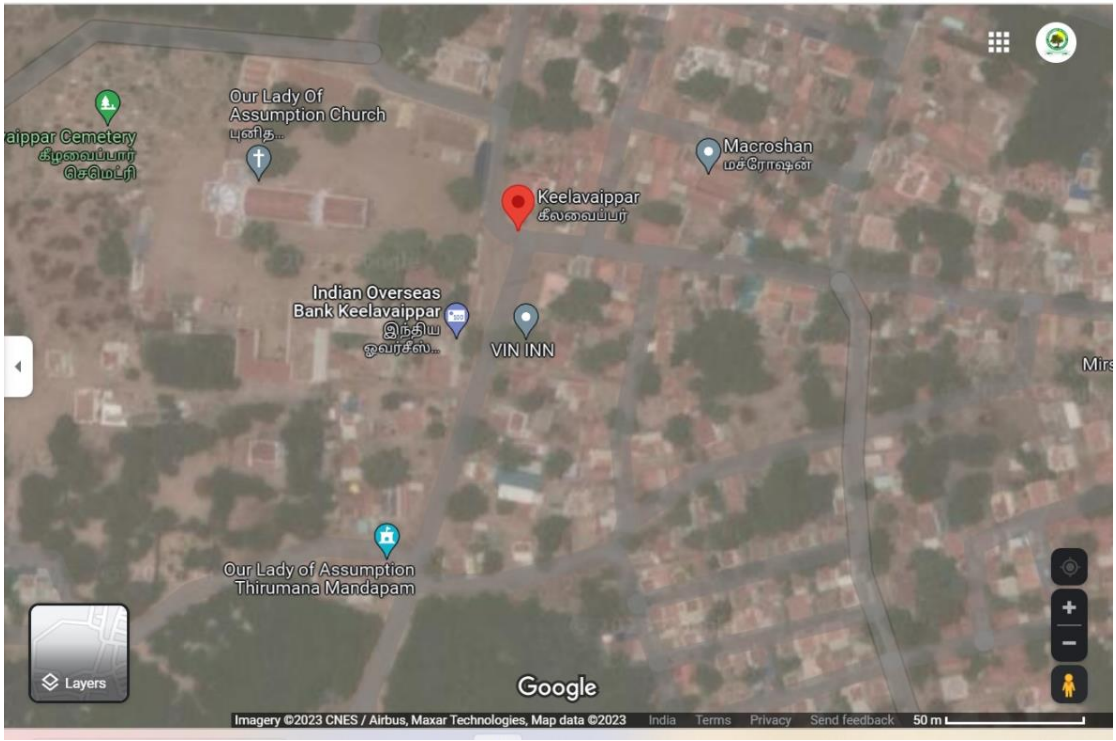

Figure 1: Study Area- Keelavaippar village (Google Map)

REVIEW OF LITERATURE

REVIEW OF LITERATURE

Water is abundant on our planet and it is essential for the survival of all form of life. The fresh water is a finite and limited resource128, 29 (Patel and Bhatt, 2010; Bouwer, 2000). The aggregate sum of worldwide water, just 2.4% is appropriated on the fundamental land, of which just a small portion can be utilized as fresh water. The accessible fresh water to man is hardly 0.3-0.5% of the aggregate water accessible on the earth and along these lines, its reasonable utilization is imperative (Appelo and Postma, 2005). Water is indispensable and one of the precious natural resources of this planet (Shahnawaz and Singh, 2009). In addition to various domestic purposes, water is required for irrigation, power generation, sanitation and industries. To ensure safe groundwater for drinking in urban and rural areas, a multi faced and comprehensive approach is necessary. The cooperation of government, health staff, industrial management and the people is needed to preserve drinking water quality in our villages. With beginning of life in earth there was no pollution. Nature was in perfect balance. Issue of pollution began with civilization of man (Chatterjee *et al.,* 2009). The demands of water supply have been increasing tremendously due to result of exploding population resulting in urbanization, industrialization, agriculture etc.

The concentration of salts depends upon the environment, and source of the ground water. The concentrations of dissolved constituents are found to be higher in groundwater than the surface waters. Polluted water used for drinking purposes leads to many diseases which are not water-borne but due to excess salts (Sabata and Nayar, 1995). The quality and type of mineral matter dissolved in water depend

on the physical structure and chemical composition of rocks. Industrial waste discharged into the aquatic system change the physicochemical properties of water such as hardness, conductivity, pH value, total dissolved solids (TDS), chemical oxygen demand (COD) and dissolved oxygen (DO) etc. Discharge of industrial effluents, domestic sewage and solid waste dump causes the groundwater to become contaminated and create health problems (Raja, 2002).

The expanding demand of water from fast growth of industries has put pressure on limited water resources (Ramesh and Soorya, 2012). Groundwater is extreme, most suitable fresh water resource with about adjusted concentration of the salt for human utilization (Tewari *et al.*, 2010). The groundwater pollution is in particular area and there is no planned readymade arrangement. Industries serve as another originator of chemicals or groundwater contamination. The greater part of these sources of groundwater contamination is localized or non-point sources such as solid waste disposal point, leakage from landfills, seepage from well and underground tanks disposal. The mining exercises are additionally a major role in groundwater contamination. Drainage from underground mines and filters from mine tailing contribute the same.

The significance and importance of groundwater has been all inclusive surely known by every one of the nation's confronting the water related issues. In India with creating financial aspects, the ideal improvement, effective use furthermore, viable administration of their water assets ought to be the predominant technique for monetary development, yet in late year's unscientific administration and utilization of this assets for different reason practically perpetually has made

undesirable issues afterward, water logging and saltiness in the instance of agribusiness utilize and environment contamination of different breaking points as an aftereffect of mining, businesses and city utilize (Kumar and Kumar, 2013). In India, the greater part of the population is reliant on groundwater as the main source of drinking water supply. The groundwater is accepted to be comparatively much clean and free from contamination than surface water (Patil and Patil, 2010).

The quality of water can be influenced by various pollutants such as, chemical, biological and physical. Microorganisms, infections, substantial metals, nitrate and salt have discovered their way into water supplies (WHO, 2007). The water contamination happens when a waterway is unfavourably influenced because of the expansion of a lot of materials to the water (Atta and Razzak, 2015). Ground water crisis is not the result of natural factors. It has been created by human activity quite a bit of sick wellbeing which impacts humankind, particularly in the developing nations can be followed to pool or traced to lake of safe and whole some water supply (Shyamala, 2009).

Ground water contains various ions and salts in high concentration, therefore using such type of water as drinking water then it leads to various water-borne diseases. Unsafe drinking water contributed to various health problems in developing countries for example, the one billion or more occurrences of diarrhea that happen yearly (Mark *et al.*, 2002). High concentration of chloride is due to the intrusion of domestic wastes and disposals by human activities (Jha and Verma, 2000). The changes in quality of groundwater response to variation in chemical,

physical, and biological environments through which it passes (Singh *et al.*, 2003; Vasanthavigar and Vasudevan, 2010).

The usage of water from ages has prompted its over abuse combined with the developing population along with enhanced way of life as an outcome of technological innovations (Todd, 1986). This contamination of groundwater is not far from the indecencies of modernization, therefore in this way, quality of groundwater is deteriorating at a speedier pace due to contamination extending from septic tanks (Banerji, 1983), land fill leachates, domestic sewage (Sharma H and Kaur, 1995; Subba Rao and Subba Rao, 1995) agricultural runoff, agricultural fields(Datta and Gupta, 1996;Handa, 1986; Rajankar *et al.,* 2013) and industrial wastes (Rengaraj and Ramalingam, 1996). Reddy (2003) have reported that the high concentration of iron in groundwater due to rusting of casing pipes, disposal of scrap iron in open areas, non-usage of bore wells for a long time, contamination due to industrial activities etc. The pH value of the water dependent upon the relative quantities of calcium, carbonates and bicarbonates (Shrivastava and Patil, 2002).

Zafar (1966) have carried out that the pH value of the water appears to be dependent upon the relative quantities of calcium, carbonates and bicarbonates. The water tends to be more alkaline when it possesses carbonates. The carbonate alkalinity was observed to be absent indicating that the total alkalinity recorded was due to accumulation of bicarbonate only (Trivedy and Goel, 1986; Jameel and Hussain, 2007; Richard and Diagnosisa, 1954). Fluoride in drinking water causes mild type of dental fluorosis (Dinesh, 1999; Gupta *et al*., 1993; Susheeia, 1993; Yadav and Lata, 2004). Saralakumari and Rao (1993). reported that a significant

part of the fluoride entering the human body is obtained from drinking water. Gopal and Gosh (1985), have studied that fluoride samples which exceeded the acceptable limit are not recommended for consumption without treatment. Fluoride is considered as an essential element though health problems may arise from either deficiency or excess amount (Rao, 2006; BIS, 1991).

Naturally, chloride happens in a wide range of waters. The contribution of chloride in the groundwater is due to minerals like mica, apatite and hornblende and also from the liquid inclusions of igneous rocks (Das and Malik, 1988; Nash H and McCall, 1995; Brown *et al*.,1970). Human excreta, especially the urine, contain chloride in an amount equal to the chlorides consumed with food and water increases the amount of chloride in municipal wastewater to about 15 mg/l above that of the carriage water in lotic systems (Sawyer and McCarty, 2000). The amount of the chloride present in the ground water samples was within the permissible limit. High chloride makes water salty in taste, which is unacceptable for human consumption (Durvey, 1991; Ganesh and Kale, 1995; Garg and Kavita *et al.,* 2004). Contamination of groundwater depends on the geology of the area and it is fast in hard rock where extensive cavern systems are below the water table (Rao, 1997; Singh, 1982). Rao and Suryanarayana (2005), has point out that groundwater chemistry has been used as a tool to outlook water quality for different purposes. Calculation of water quality index is an important technique for separating groundwater quality and its suitability for drinking purposes (Tatawat and Chandel, 2007; Tripathi and Singh, 1996).

WQI (Mangukiya *et al*., 2012; Mariappan *et al.,2005;* Rajankar et *al.*, 2013; Singh *et al.,* 2013), is characterized as a method of rating that gives the composite impact of individual water quality parameters on the general quality of water for human utilization. Shivasharanappa and Mallikarjun (2011), made investigation on assessment of ground water quality characteristics and Water Quality Index (WQI) of Bidar city and its industrial area, Karnataka State.

Ground water pollution (Chhatwal Gurdeep Raj, 2002; Rao and Yadav, 2004) by the heavy metals has turned into a striking issue for most recent two decades as consequences of release of industrial effluent, untreated domestic waste and increasing use of agrochemicals, i.e. fertilizers and pesticides in cultivation.

The water for utilization comes to people in various structures and from various sources. There have been two principal sources of drinking water; one is surface water and other one is groundwater. In villages the main source of drinking water is groundwater available through hand pumps or bore wells (Ray and Sanyal, 2001). Groundwater contamination in contrast to others is very critical, as a decentralized source of drinking water and other services for millions of rural and urban people, groundwater as a natural resource plays a crucial role which, accounts for nearly 80% of the rural domestic water needs and 50 per cent of the urban water needs in India (Kumar *et al.,* 2005).

Various research workers have carried out broad studies in the related field. Karunakaran (2009) have made statistical study on physicochemical characteristics of groundwater in and around Namakkal, Tamilnadu, India and suggested the necessity to purify the available water resource prior to utilization. Patil and Patil

(2010) carried out physicochemical Analysis of selected groundwater samples of Amalner Town in Jalgaon District, Maharashtra, India and reported that the need of some treatment for minimization of the different parameters. Das Gupta *et al*. (2001) have made assessment of drinking water quality of River Brahmani. Raja *et al*. (2002), assessed the physico- chemical parameters of some groundwater samples of Kotputli Town Jaipur, Rajasthan.

Gupta *et al*. (2010) have studied the physico-chemical analysis of drinking water quality from 32 locations in Delhi and reported that most of the water quality parameters slightly higher in the wet season than in the dry season. Drinking water is the major problem faced in the urban areas. Datta *et al*. (1996) have carried out assessment of groundwater contamination from fertilizers in Delhi and reported that the conditions prevailing in the urban area make the water pullulated. Raja and Venkatesan (2010), have studied groundwater pollution and its impact in and around Punnam Area of Karur District, Tamilnadu, India and reported that the water samples are highly polluted, hence suggested to exercise all the necessary precautions before the water is used for drinking and irrigation. Otherwise, it may lead to much adverse health effect. Shrivastava and Tyagi, (2003) have made study of physicochemical characteristics of waterbodies around Jaipur that Jalmahal lake and found most polluted having high alkalinity, free CO_2, hardness and pH but a low level of DO, endosulfan and zinc contents in Jalmahal lake well also high, thus making it unsuitable for biota and fish and contrarily, Ramgarh lake shown high DO, low alkalinity, free CO_2, hardness pH, endosulfan and zinc concentrations were relatively low throughout the year.

Craig and Anderson (1979) carried out study on the effects of urbanization on groundwater quality. Ghosh *et al*. (2010) investigated assessment of health risks associated with fluoride-contaminated groundwater in Birbhum district of West Bengal. Dass *et al*. (2001) carried out a study on fluoride and other water quality parameters of ground water of district Agra (U.P.). Hemant *et al*. (2012) reported study on seasonal variation in groundwater quality of Sagar city. Murli *et al*. (2011) evaluated the ground water quality in Coimbatore South Taluk, Coimbatore district. Loganathan *et al*. (2011) investigated status of groundwater at Chennai city. Keshvan and Parameswari (2005) made investigations on ground water quality in Kancheepuram. Belkhiri *et al*. (2010) investigated groundwater quality and its suitability for drinking and for agricultural use in Ain Azel plain Algeria. Khaiwal and Garg 2(006) studied distribution of fluoride in groundwater and its suitability assessment for drinking purpose.

Mishra *et al*. (2010) worked out the comparative study of physico- chemical and microbial parameters on Lotic and ground-waters in selected outlying areas of central Gujarat. Nagarajappa *et al.* (2011) assessed physico-chemical analysis of underground water of Harihara taluk of Davanagere district, Karnataka. Mishra and Patel (2011) have made study on the pollution load in the drinking water of Rairangpur, a small tribal dominated town of North Orissa. Mumtazuddin *et al*. (2012) have carried out the physico-chemical analysis of groundwater of the Budhi Gandak belt in Muzaffarpur district. Muthukumaravel *et al*. (2010). have reported the evaluation of groundwater quality in Perambalur. Mohan and Choudhary (1998). have made investigations on hydrochemistry and quality assessment of ground water quality in Jind city.

Kaur and Singh (2011) have made assessment for different groundwater quality parameters for irrigation purposes in Bikaner city, Rajasthan. Aghadeh and Mogaddam (2010) carried out assessment of groundwater quality and its suitability for drinking and agricultural uses in the Oshnavish Area, Northeast of Iran. Niranjan *et al.* (2011) carried out groundwater quality assessment of Wailpalli Nalgonda.

Patil *et al*. (2010) made study on the physicochemical characteristics of ground water of Gulbarga city, Karnataka. Ravichandra and Chandana (2006) evaluated groundwater pollution in Bakkannaplem, Visakhapatnam. Ramakrishna *et al.* (2009) studied groundwater quality in slums of Visakhapatnam. Rao *et al.* (2012) carried out assessment of ground water quality for application in Kakinada. Subba Rao *et al.* (2012) carried out an assessment of quality of drinking water at Srikurmam in Srikakulam District, Andhra Pradesh. Rao and Suryanarayana (2005) assessed ground water quality in a coastal area, Andhra Pradesh. Ratnakanth Babu (2011) carried out on assessment of groundwater pollution in parts of Guntur District. Raviprakash and Rao (1989) studied the chemistry of ground water in Paravada area with regard to their suitability for domestic and irrigational purposes. Gorde and Jadhav (2013) carried out assessment of water quality parameters.

Mukherjee *et al*. (2005) made work on assessment of groundwater quality in the South 24-parganas, West Bengal coast. Sabahi *et al*. (2009) studied the characteristics of leachate and groundwater pollution at municipal solid waste landfill of Ibb city, Yemen. Saha *et al.* (2008) made investigation on geochemical evolution of groundwater in the Pleistocene aquifers of South Ganga Plain, Bihar. Kumar (1993) reported among water quality parameters for ground water in Barmer

district. Sharma *et al.* (2005) have studied quality status of groundwater of Sanganer tehsil in Jaipur district. Singh *et al*. (2013) evaluated hydrogeochemical processes and groundwater quality in the Jhansi district of Bundelkhand region. Somashekar *et al*. (2000) evaluated the groundwater chemistry of Channapatna Taluk, Bangalore rural district. Subba Rao (1993) studied the environmental impact of industrial effluents on ground water in regions of Visakhapatnam Industrial Complex.

Suryanarayana (1995) carried out effect of groundwater quality on health hazards in parts of Eastern ghats. Tiwari and Singh (2014) carried out the hydrogeochemical investigation and groundwater quality assessment of Pratapgarh district, Uttar Pradesh. Banerji (1983) made investigation on importance of evolving a management plan for groundwater development in the Calcutta region of the Bengal basin. Naik and Purohit (2001) reported Studies on water quality of river Brahmani in Sundargarh district, Orissa. Rao *et al*. (1996) worked on factors controlling groundwater quality in parts of Srikakulam District, Andhra Pradesh.

Industrial effluents contain heavy metals which have long lasting adverse effects on human health. Today, heavy metals are as often as possible present in our water, soil and air because of broad utilization of their compounds (Gupta *et al.*, 2014). Heavy metal pollution in groundwater causes major health effects on human beings. Banerjee *et al.* (2011) reported heavy metal contaminants in underground water at Indo Bangla Border Districts of Tripura, India. Mohan *et al.* (1998). studied heavy metals in the groundwater of non-industrial area. Mohapatra and Singh (1999) reported trace metals in drinking water from different sources in old capital city of Cuttak. Removal of zinc from wastewater by adsorption reported by

Shrivastava *et al.* (2007). Preliminary studies on heavy metals in ground water of Mandeep has been carried out by Verma *et al.* (1995).

In recent years the water resources are depleting at an alarming rate due to careless misuse, consequently need of water resource management and protection strategies are on prime interest (Chatterjee *et al.*, 2009; Bhopal *et al.*, 2012; Cristina *et al.*, 2012*;* Srivasthava *et al.*, 1994). High concentration of fluoride in drinking water caused hazardous effects studied by many workers in different parts of India. Ashley and Burely (1994) made investigation on controls on the occurrence of fluoride in the groundwater in the Rift Valley of Ethiopia. Banerjee (2015) reported groundwater fluoride contamination. Ghosh *et al.* (2010) carried out assessment of health risks associated with fluoride contaminated groundwater in Birbhum district of West Bengal.

In India most of the states are facing acute water shortage. In these states top priority is recharging the aquifers through rainwater harvesting. Scientists developed so many techniques and modelling for interpretation of water quality in terms of suitability for domestic, drinking, industrial and irrigation purpose. LSI, AI and RI (Prajapati and Mathur, 2005) are the most popular among these all. Water quality data presented in terms of % Na, SAR, ESP and RSC shows the suitability of water for irrigation purpose. In recent years remote sensing and GIS (Machiwal and Jha, 2015) technology is developed to assess the water properties. Remote sensing data are well used in water resource evaluation and management.

Seasonal pollution assessment through comparative hydro biolological studies in river Jojari at Salawas, Jodhpur has been discussed by Vishoni *et al*.

(2005). Sinha and Saxena (2006) worked on statistical assessment of underground drinking water contamination and effect of monsoon at Hasanpur, J.P. Nagar, Uttar Pradesh. Hemant and Limaye (2011) made study of seasonal variation in groundwater quality of Sagar city.

Abdul Saleem *et al.* (2012) assessed correlation-regression model for physicochemical quality of groundwater in the South India city of Gulbarga. Chandra Sekhar and Satya Prasad (2005) studied regression models for assessment of dissolved pollutants in Krishna River. Dash *et al.* (2006) studied a correlation and regression study on the ground water quality in rural areas around Angul-Talcher industrial zone. Jeyaraj *et al*. (2001) reported correlation among water quality parameters for ground water samples of Bharathi Nagar of Tiruchirapalli City.

MATERIALS AND METHODS

MATERIALS AND METHODS

The three different water samples (lake water, bore water, well water) were collected from Keelavaippar Village in Polythene Bottles. The Water samples were immediately brought in to Laboratory for the estimation of various Physico-chemical parameters like colour, odour, turbidity, pH, TDS, free CO2, Hardness, Alkalinity, Chlorides, Phosphate, Nitrate, Calcium, Magnesium, Sodium, Potassium, Iron, Manganese, Ammonia, Nitrite, Fluoride and Sulphate were estimated in the Laboratory by using APHA (2012) procedures.

Turbidity

Determine turbidity as soon as possible after the sample is taken. Gently agitate all samples before examination to ensure a representative measurement. Sample preservation is not practical, begin analysis promptly. Refrigerate or cool to 4°C, to minimize microbiological decomposition of solids, if storage is required. For best results, measure turbidity immediately without altering the original sample conditions such as temperature or pH. Turbidity is measure with the help of Digital turbidity meter. This calibrated with the help of standard NTU.

Determination of pH

The pH of water is measured with the help of pH meter which gives grounds to judge the properties of the water especially with reference to the presence of carbon and bicarbonate of sodium, calcium and magnesium.

Electrical conductivity:

Procedure:

1. Read the temperature of the sample and adjust the temperature knob of the conductivity meter accordingly.
2. Shift the selector switch to 1000 and adjust to CAC Mark.
3. Take the sample in a beaker and immerse the conductivity cell in it.
4. After connecting the cell terminals to the sockets switch to X 100 and repeat the step described above.
5. Record the deflection at this level.
6. Shift the selector switch to X 10 and perform the steps described above.
7. Disconnect the cell terminals, was with distilled water and put off the instrument.

Total Dissolve Solids (TDS)

Total dissolve solids are those which are present in water after filtering water through 1 micro meter pores. The procedure is carried out by heating the sample at 1030º C in the oven till drying of evaporated dish.

$$\textbf{TDS (mg/ l)} = \textbf{Wf} - \textbf{Wi x 1000 x 1000/ Volume of sample}$$

Where,

Wi- Initial weight of evaporated dish

Wf- Final weight of evaporated dish

Determination of Alkalinity:

Reagent:

a) Sulphuric acid (0.02 N)

b) Sulphuric acid (0.01 N)

2.8 ml of conc. H_2SO_4 diluted to 1 litre using distilled water. Take 200ml from this using distilled water. Take 200ml from this stock solution and dilute to 1 litre using distilled water.

c) Phenolphthalein indicator

1 g of phenolphthalein is dissolved in 100ml of ethyl alcohol. After complete dissolution add 100ml of distilled water. Add NaOH reagent (0.2272) in drops till a faints pink colour appears.

d) Methyl orange indicator

0.1 g of methyl orange is dissolved in 200ml distilled water.

Procedure:

1. Estimation of alkalinity should be done immediately after the collection of sample.
2. In 50ml of sample, add 2-3 drops of phenolphthalein indicator.
3. If the solution shows pink colour, titrate it against sulphuric acid.
4. Appearance of slight pink colour indicates the presence of hydroxides or carbonates, whereas colourless sample confirms the presence of Co2.
5. Note down the colourless and point as P.
6. Now add 2-3 drops of methyl orange indicator to the same flask and proceed with the titration till the solution turns from yellow to orange.
7. Record the values at t and calculate the alkalinity the formula

Alkalinity due to bicarbonate = t × 1000/ volume of sample.

Alkalinity due to carbonate = P × 1000/ volume of sample.

Determination of Total Hardness:

Reagent:

a) Ammonia buffer solution

Dissolved 13.5 g of ammonium chloride in 114 ml of conc. ammonium hydroxide make up to 200 ml using distilled water.

b) Erichrome black T indicator

Dissolve 0.5 g of Erichrome black T dye in 100 ml of ethyl alcohol .

c) EDTA Solution (0.001 m)

3.723 g of sodium salts of EDTA in little distilled water and make up to 100 ml use polythene bottle for storage.

Procedure:

1. In 50 ml sample add 1 ml of ammonium buffer solution and 4 drops of Erichrome black T indicator.
2. Now the solution turns wine red colour.
3. Titrate it against EDTA Solution till the colour changes from wine red colour to blue.
4. Record the end point, repeat the procedure to attain constant value.
5. Calculate the total hardness using the formula.
6. Total hardness = E × 1000/ V.

Estimation of Nitrate:

Reagent:

a) 30% Sodium chloride

b) 10 ml conc. H_2SO_4

c) 0.5 ml Bruncines sulphate acid

Procedure

1. 2 ml of 30% Sodium chloride solution is added to 10 ml of water sample taken in a 250 ml volumetric flask.
2. To this add 10 ml of conc. H_2SO_4 and 0.5 ml 0.5% of Bruncines sulphanilic acid.
3. Place the flasks in cool water bath.
4. Mix thoroughly and then boil in a hot water bath for 20 minutes.
5. Allow it cool and read the absorbance at 410 nm.
6. Distilled water is used as blank.
7. Draw a standard given by plotting the concentration of nitrate solution against absorbance value.

Determination of Chloride:

Reagent:

a) Standard sodium chloride solution

b) Nitric acid 0.1N

c) Sodium hydroxide 0.1N

d) Reagent for chloride concentrations below 700 mg/I

e) Standard mercuric nitrate titrant

f) Mixed indicator reagent

g) Strong standard mercuric nitrate titrant 0.141 N

Procedure:

1. Use a 100 ml sample that the chloride is less than 10 mg. Add 1 ml indicator acidifier reagent.

2. For highly alkaline or acid waters, adjust pH to about 8 before adding indicator acidifier reagent. Titrate with 0.41 1 mercuric nitrate to a definite purple end point.
3. The solution turns from green blue to a few drops before the end point. Determine the blank titrating 100 ml distilled water containing 10 mg of sodium bicarbonate.

$$\text{Chloride, mg/I(V2)} = \frac{(V1-V2) \times N \times 35.450 \times 1000}{20}$$

Where

V1 = volume in ml of silver nitrate used by the sample,

V2 = volume in ml of silver nitrate used in the blank titration,

V3 = volume in ml of sample taken for titration and

N = normality of silver nitrate solution

Determination of Fluoride:

Reagents

a) Fluoride stock solution

To a 100 ml volumetric flask, add 0.221 g of anhydrous sodium fluoride. Make up to mark using distilled water.

b) Fluoride standard solution

Transfer 100 ml of the stock solution in a 1000 ml volumetric flask using distilled water. Make up to mark one ml of this solution contains 0.01mg fluorine.

c) Acid zirconium Alizarin solution

i) Dissolve 0.7 g sodium alizarin sulphate in 100 ml distilled water.

ii) Sulphuric acid solution

Carefully add 70 ml of conc. Sulphuric acid to 700 ml distilled water. Mix well and cool. Add solution (ii) to (i) stir well and finally add solution. Make up the mixed solution to a final volume of 1000 ml.

d) Sodium arsenate solution

To 100 ml distilled water add 1 g of sodium arsenate stir well and store it in amber bottle.

Procedure

1. Prepare several dilutions of fluoride standards solution into a series of 50 ml Nessler's tubes with their concentrations ranging from 1 ml to 12 ml at 2 ml interval.
2. Shift the pH of each solution to neutral pH (pH7) and make up the volume of each tube to 50ml using distilled water.
3. Take 50ml of distilled water as blank and the prepared sample in separate tubes.
4. Add 1 ml of acid zirconium alizarin solution to the standards sample and blank. Stairwell and measure their absorbance.
5. Draw a standard graph by plotting their absorbance value against concentrations.
6. From the calibration curve evaluate the concentration of fluoride in the sample.

Note

Residual chlorine interferes with the analysis and can be eliminated using sodium arsenate.

Determination of Sulphates:

Reagents

a) Methyl red indicator

Dissolve 10 g of barium chloride in 100ml of distilled water. Filter it through whatman No:1 filter paper and store for use.

b) Silver nitrate solution

Dissolve 8.5 of silver nitrate powder in little nitric acid solution. Make up to 500 ml using distilled water.

c) Hydrochloric acid (50%)

Add distilled water and conc. Hydrochloric acid in the ratio 1:1.

Procedure

1. Transfer 100 ml of sample into a 250 ml volumetric flask.
2. To this add 2-3 drops of methyl red indicator.
3. Slowly add hydrochloric acid to the flask content and observe for the colour change from red to orange. (This indicator the acidic pH of the solution).
4. Add still more hydro chloride acid to the flask and boil it.
5. Now add warm barium chloride solution so as to favour the precipitation process.
6. Continue to add it till the precipitation is completed.
7. Again heat it in a hot water bath for about 2 hours. Do not cool it. Immediately filter it through ash less filter paper.
8. Flush the filter paper with distilled water many times.
9. This should be repeated till the filtrate does not contain traces of chloride.
10. Every time after washing the filter paper, check the presence of chloride in the filtrate by filtering it against silver nitrate solution. Absence of turbidity indicates nil chloride content and at this stage transfers the filter paper to a silica crucible.

11. Ignite it in a muffle furnace at 80°C for 1 hour. After one hour take the weight of the precipitate.

Estimation of Calcium:

Reagent:

a) Ammonium buffer : Dissolved 5 g of ammonium chloride in 80 ml of liquid ammonia.

b) Erichrome black T : A fine mixture of sodium chloride and Erichrome black T is made in proportion of 200:1

Procedure:

1. 5 ml of the sample was taken in conical flask and 5 ml of sodium hydroxide was added followed by a pinch of muroxide indicator.
2. It was titrated against Ethylene diamine tetra acetic acid until the colour changes from pink to purple this titrate value was taken as B.

Estimation of Magnesium:

Reagent:

a) Ethylene diamine tetra acetic acid (0.1)

Dissolved 8.143 g of Ethylene diamine tetra acetic acid in 500 ml of distilled water. In order to avoid probable decomposition, it was better to keep the solution in polythene bags.

b) Muroxide indicator (ammonium purpurate)

A solid mixture of muroxide and sodium chloride in the ratio 1:100

Procedure:

1. 5 ml of the water sample was taken in a conical flask and 5 ml of ammonium buffer was added.

2. It was diluted to 100 ml with distilled water and a pinch of Erichrome black T was added.
3. The solution was warmed to 60°C and it was titrated against Ethylene diamine tetra acetic acid from the burette.
4. The end point from salmon red to blue. The titrate value is taken as A.

Calculation:

Consumption of EDTA to the calcium and magnesium = A

Consumption of EDTA due to the calcium alone = B

Consumption of EDTA due to the magnesium = A-B

Factors value F for calcium = 2 mg

Amount of calcium in the sample = F × B × 1000/ sample volume.

Amount of magnesium in the sample = A × (A × B) × 1000/ sample volume.

Determination of sodium and potassium:

Reagents:

a) Double distilled water
b) Sodium stock solution

Procedure:

1. Take approximately 3 g of sodium chloride powder, dry in it hot air oven at 140°C for about one hour.
2. Then from that amount weigh accurately 2.5422 g and dissolve it in one litre of double distilled water.
3. 1 ml of this solution contains 1 mg of sodium.

Estimation of Dissolved Oxygen

Reagent

a) Manganous sulphate,

b) alkali mixture,

c) starch indicator,

d) Con.H2SO4,

e) sodium thiosulphate [0.025N].

Procedure

1. The sample collecting bottles [250ml] must below the surface of the water body while filling.
2. The water sample allowed to over flow in order to avoid entrapping of air bubbles and stopper. The bottle is opened and 1ml of manganous sulphate and 1ml of alkali mixture are added.
3. The bottle is restoppered and carefully tilted when a white precipitate of manganese hydroxide is formed and settled at the bottom.
4. The stopper is removed and 1ml of con.H_2SO_4 is added. The bottle is tilted carefully until a clear straw coloured solution is formed due to the liberation of iodine.
5. Take 100ml of the solution from the bottle and is transferred to 250 ml conical flask. This is titrated with 0.025 N sodium thiosulphate until a pale yellow colour formed.
6. At this stage 3-4 drops of starch indicator is added and the titration is continued until the disappearance of the blue colour.
7. The final volume of sodium thiosulphate solution in note.

Estimation of Chemical Oxygen Demand

Reagent

a) Potassium dichromate solution

b) Ferrous ammonium sulphate solution

c) Ferrion indicators

d) Mercuric sulphate

e) H_2SO_4.

Procedure

1. Take 5ml of the sample in a standard flask. For effluents with high COD, reduce the size of sample by diluting it with distilled water.
2. Make sure that the final volume is 20 ml, for turbid samples, blend the solids using homogenizer and then add the required reagents for digestion.
3. To this, add 2.5 ml of 0.025N potassium permanganate solution.
4. Shake well and add a pinch of mercuric sulphate powder. Addition of mercuric sulphate should be based on the chloride content of the sample, i.e. it should be added in the ration of 10:1.
5. See to it that the sample is golden orange in colour, place that flask in a cool water bath and add 30 ml of sulphuric acid solution. Allow it to stand for 30 minutes.
6. Now digest the content of the flask in a reflux condenser for 2 hours. Following the digestion process, put off the mantle and allow the water to circulate for another 10 minutes.

7. Then detach the unit, dilute it to 37.5ml using distilled water. Add 2-3 drops of ferroin indicator and titrate it against 0,1N ferrous ammonium sulphate solution.
8. Observe for the colour change from bluish green to blue to red. The end point is very sharp and as soon as the solution shows bluish green colour, perform the titration carefully and slowly. run blank using distilled water.

Determination of Iron:

Reagents:

a) Concentrated hydrochloric acid

b) Hydroxylamine hydrochloride solution

10 mg of hydroxylamine hydrochloride dissolved in distilled water. Make up to 100 ml.

b) Ammonium acetate buffer solution

100 mg of ammonium acetate dissolved in 60 ml distilled water. To this add 280 ml of glacial acetic acid.

c) Phenolphthalein solution

50 mg of 1, 10 – phenanthroline monohydrate dissolved in 50 ml distilled water. Ensure complete mixing and heat up to 80°C in a water bath.

Standard iron solution

Dissolve 1.404 g of FAS in 20 ml of sulphuric acid diluted with 50 ml distilled water. Add $KMnO_4$ solution in drops till faint pink colour appears. Make this solution to 1.1 keep this stock solution to prepare standard iron solution from 1-5 mg.

Procedure:

1. In 50 ml of sample, add 1 ml of HCL and 1 ml of hydroxylamine hydrochloride solution.
2. Mix well and heat the contents till boiling point.
3. Put off the flame when the content is half of its original volume.
4. After cooling, add 2 ml each of ammonium acetate buffer solution and phenanthroline solution.
5. Dilute the content with distilled water and make up to 100 ml.
6. Keep this flask for ten minutes and measure the absorbance on Spectrophotometer (510) using distilled water as blank.
7. Repeat the same procedure for standard iron solution series and note down their respective absorbance at 510 nm.
8. Using these values draw a standard curve.
9. Evaluate the total iron content by comparing sample values with the different dilution of known standard solution.

Determination the Manganese:

Reagents

a) Manganese stock solution

Prepare 1% sulphuric acid solution. To this add 1 g of pure manganese metal. Stir well and ensure complete dissolution. Make up this solution to one litre. One ml of this solution contains 1 mg manganese.

b) Manganese working solution

Transfer 5 ml of manganese stock solution to a 500 ml volumetric flask. Make up to the mark using distilled water. One ml of this solution contains 1 mg manganese.

c) Special reagent

d) Mix 200 ml of concentrated nitric acid to 100 ml of distilled water. To this add 3% 5 g of mercuric sulphate 100ml of 85% phosphoric acid and make up to 500 ml using distilled water.

e) Ammonium per sulphate crystals

Procedure

1. Prepare several dilutions of manganese working solution in a series of beaker with their concentrations ranging from 5 ml to 50ml at 5ml interval. Make up the content of each beaker to 100ml using distilled water.
2. Take 100ml of distilled water as blank.
3. Similarly pipette out 100ml sample in a beaker.
4. To all the beakers add 5ml of special reagent.
5. Continue to heat untill the content of each beaker is reduced to 40ml.
6. Now add 1 g of ammonium per sulphate to each beaker and boil for another minute.
7. Carefully remove the beakers from the hot plate and cool them under running tap water.
8. Measure the absorbance value of all the solution, sample, standards and blank in Spectrophotometer at 545 nm.
9. Draw a standard curve by plotting the absorbent value of standards against their concentrations.
10. From the standard curve, evaluate the concentrations of manganese in the sample.

RESULTS AND DISCUSSION

RESULT AND DISCUSSION

Water is life, however good quality drinking water is still a dream for most the population. Hence determining the water quality for human consumption and recreational purpose is very essential. The consumption of high fluoride water causes floristic and skeleton disorder whereas high nitrate causes methaemoglobinaemia disease (Spalding and Exner, 1993). All the quality parameters have equal importance; on the other hand, fluoride and nitrate values are most important due to more hazardous impact on human health. Water quality has major impact on soil quality and crop yield. This investigation is the first attempt to analyse the quality of different water resources.

WATER QUALITY PARAMETERS

Physical Parameters

Table 1 shows the result of physical parameters like colour, odour, turbidity, pH, electrical conductivity and Total dissolved solids.

Colour

Colour in water is primarily a concern of water quality for aesthetic reason. Coloured water gives the appearance of being unfit to drink, even though the water may be perfectly safe for public use. On the other hand, colour can indicate the presence of organic substances, such as algae or humic compounds. More recently, colour has been used as a quantitative assessment of the presence of potentially hazardous or toxic organic materials in water (Tsair-Fuh Lin *et al.*, 2019). In the present study, all the selected water samples were colourless.

Odour

Human detect many more tips of odour than tastes. Organic materials discharged directly to water, such as falling leaves, runoff, etc., are sources of tastes and odour-producing compounds released during biodegradation. odour in source water include decaying vegetation, algae, moulds and actinomycetes. Odour are usually associated with the presence of specific organic compounds released by the source agent which give rise to “earthy” or “musty” taste or odour. Chlorine and the by-products of chlorination can also cause complaints of taste or odour. Domestic plumbing materials and arrangements and in some circumstances water mains may also impart a noticeable taste or odour (Tsair-Fuh Lin *et al.*, 2019). Fortunately, all the three selected water samples were odourless.

Turbidity

The presence of suspended materials such as clay, slit, finely divided organic material, plankton, and other inorganic materials in water is called turbidity. Turbidity is a measure of the clarity of water. Low-turbidity water is clear, while high turbidity water is cloudy or murky (Reham and Abu Shmeis, 2018). Turbidity of well water, bore well water and lake water were 0.7, 0.8 and 0.5 NTU respectively which were lower than maximum permissible limit (5 NTU) of IS 10500.

pH Values

The pH is considered as an important ecological factor and provides an important information for geochemical equilibrium. The variations in pH values may be due to increase or decrease of human and other biological activities. The permissible limit of pH values for drinking water is specified as 6.5 to 8.5 as per IS

10500. The pH values of well water, bore well water and lake water were 6.85, 7.08 and 7.66 respectively. Generally, pH of water is influenced by geology of catchments area and buffering capacity of water. The factors like air temperature bring about changes the pH of water. Most of bio-chemical and chemical reactions are influenced by the pH. The reduced rate of photosynthetic activities reduces the assimilation of carbon dioxide and bicarbonates which are ultimately responsible for increase in pH (Kamble *et al.*, 2009)

Electrical Conductivity

Electrical conductivity measures the amount of dissolved mineral content in water. Conductance is not too harmful but water with higher conductance is not suitable for drinking, irrigational and other purpose. Conductance generally varies according to the season. The values of conductance of well water, bore well water and lake water were 3003 µS/cm, 7261 µS/cm and 1035 µS/cm correspondingly. The result indicated that, the number of free ions is higher in bore well water.

Total Dissolved Solids (TDS)

Water of high TDS is not suitable for use in boilers and hence restricted industrial use. Normally ground water has a higher total dissolved solids load compared to surface water. The TDS of well water, bore well water and lake water were 2042 mg/L, 4938 mg/L and 693 mg/L. High TDS value was observed in bore well water. As per the salinity classification suggested by Robinove *et al.* (1958) lake water is non saline (TDS < 1000), well water is slightly saline (TDS =1000-3000) and bore well water is moderately saline (TDS=3000-10000).

Chemical Parameters of Water

Total Alkalinity

Alkalinity of water is its capacity to neutralize a strong acid. According to IS 10500 the maximum allowable concentration of total alkalinity for drinking water is 600 mg/L. Alkalinity in natural waters is due to free hydroxyl ions and hydrolysis of salts formed by weak acids and strong bases such carbonates and bicarbonates. Total alkalinity of well water, bore well water and lake water were 356 mg/L, 440 mg/L and 156 mg/L (Figure 2). Total alkalinity of all samples was below the permissible limit. It is itself not harmful to human being (Pande and Sharma, 1999).

Total Hardness

Hardness is the measure of calcium carbonate in the sample. The hardness in water may deliver from dissolved CO_2, release by bacteria found in water. The major cations imparting hardness are calcium and magnesium. Hardness is the property of water, which prevents the lather formation with soap and increase the boiling point of water. Figure 3 shows that he total hardness of well water, bore well water and lake water were 900 mg/L, 2200 mg/L and 240 mg/L. According to IS 10500 the maximum permissible total hardness is 600 mg/L. Total hardness of both well water and bore well water samples exceeded permissible limit. Durfor and Becker (1964) have classified water as soft (0-60 mg/L), moderate (61-120 mg/L), hard (121-180 mg/L) and very hard (>180 mg/L) based on their hardness. On the basis of this classification it has been observed that all the water samples tested are very hard water. So, action of soap and detergents is very difficult in it. The anions responsible for hardness are carbonates, bicarbonates, sulphate and

chloride. Hardness is temporary if it is associated mainly with carbonates and bicarbonates, and permanents if with sulphate and chloride. The high concentration of total hardness may cause heart disease and kidney problem (Jain *et al.,* 1997).

Nitrate

Domestic sewage, industrial effluents, natural run-off and agricultural wastes are the important sources of it. Nitrate is one of the critical nutrients for the growth of algae and helps accelerating the eutrophication. Well water, bore well water and lake water showed nitrate content of 51 mg/L, 58 mg/L and 34 mg/L (Figure 4). The maximum allowable limit of nitrate in drinking water as per IS 10500 is 45 mg/L. Nitrate ion concentration is very important in drinking water because if it exceeds 45 mg/L it causes blue babies' syndrome (Methaemoglobinaemia) in children (Almasri and Kaluarachchi, 2004; Bohlke, 2002). Of the selected samples, well water and bore well water exceeds maximum permissible limit.

Chloride

The most important source of chloride in natural waters is the discharge of sewage. Chloride occurs naturally in all types of waters. In natural fresh waters, its concentration remains quite low. Chloride may present naturally in groundwater and may also originate from diverse sources such as weathering, leaching of sedimentary rocks and infiltration of seawater etc. (Rout and Sharma, 2011). The chloride concentration in well water, bore well water and lake water were 740 mg/L, 250 mg/L and 250 mg/L correspondingly (Figure 5). The values observed are within the permissible limit of 1000 mg/L as per IS 10500.

Fluoride

Water containing high fluoride concentration is not suitable for drinking water purpose because fluoride causes mottling of teeth, skeletal fluorosis, forward bending of vertebral column, defloration of knee joints (Westcot and Ayers, 1984). The fluoride ion concentrations in the study were 0.6, 0.8 and 0.4 mg/L in well water, bore well water and lake water (Figure 6). As per IS 10500 maximum fluoride concentration in drinking water is 1.50 mg/L.

Sulphate

Sulphate content of selected water samples is depicted in Figure 7. The maximum allowable limit of sulphates in drinking water as per IS 10500 is 400 mg/L. The sulphate ion concentration in well water, bore well water and lake water 129 mg/L, 210 mg/L and 75 mg/L respectively. The sulphates content may be contributed due to bio chemical, anthropogenic sources and industrial processes etc. Sulphate is a naturally occurring anion found almost in all kinds of water bodies. This is also an important anion imparting hardness to the waters. The sulphate ion produces cathartic effect upon human beings when it is present in excess. All the samples found to be well within permissible limit

Calcium and Magnesium

The calcium and magnesium are the most abundant elements in the groundwater. Calcium may dissolve readily from carbonate rocks and lime stones or be leached from soils. However, dissolved Mg^{2+} concentration is lower than Ca^{2+} in the groundwater. Other sources include primarily industrial and municipal discharges. Calcium is an essential nutritional element for human being and aids in maintaining the structure of plant cells and soils. Mg^{2+} is

a constituent of bones and is essential for normal metabolism of Ca^{2+}. Its deficiency may lead to protein energy malnutrition (Rout and Sharma, 2011). Calcium and magnesium content of selected water samples are presented in Figure 8. 248, 560 and 62 mg/L of calcium was found to be present in well water, bore well water and lake water and 67, 192 and 20 mg/L of magnesium were present in well water, bore well water and lake water respectively. Maximum allowable calcium and magnesium limit is 200 mg/L and 100 mg/L correspondingly (IS 10500). Higher amount of calcium and magnesium might be due to the heavy discharge of organic matter through birds' guano. Higher amount of calcium might be due to the rapid oxidation of organic matter in the substrate.

Sodium

Practically all sodium compounds are water soluble and tend to remain in aqueous solution. Water in contact with igneous rocks will dissolve sodium from its natural source. Higher concentration of Na+ ion in drinking water may cause heart problems. Higher Na^+ ion in irrigation water may cause salinity problems. Excessive amount of Na^+ ion in groundwater normally affects the palatability of water (Rout and Sharma, 2011). Sodium values of well water, bore well water and lake water were 300 mg/L, 750 mg/L and 110 mg/L (Figure 9). The maximum value of sodium is examined in bore well water sample and the minimum value of sodium measured in lake water. The permissible limit of sodium in drinking water as prescribed by BIS is 50 mg/l. On comparison with BIS standards, Na^+ concentration of all samples was found to be higher than the permissible limit.

Potassium

Potassium is an important cation and plays a vital role in intermediately metabolism. K^+ is an essential nutrient for both plant and human life. However, ingestion of excessive amounts may prove detrimental to human beings (Rout and Sharma, 2011). Well water, bore well water and lake water contained 38 mg/L, 88 mg/L and 15 mg/L of potassium (Figure 10). The maximum value of potassium examined in bore well water.

Dissolved Oxygen (DO)

Dissolved oxygen is a most important aquatic parameter, whose existence is essential to aquatic fauna. It plays an important role in life process of animals. The condition in case of dissolved oxygen (DO) is slightly complicated since in contrast to other pollutants, the quality of water is enhanced if it contains more oxygen. An ideal DO value of 5.0 mg/l is the standard for drinking water (Bhanja et al., 2000). In natural waters, DO values vary according to the physicochemical and biological activities. The data reveals that, 0.25 mg/L, 0.27 mg/L and 0.1 mg/L of DO values were observed in well water, bore well water and lake water (Table 2). DO levels below 3 milligrams per liter (mg/L) are of concern and waters with levels below 1 mg/L are considered hypoxic and usually devoid of life (USEPA, 2022). In water bodies, DO levels fluctuate periodically, seasonally and even as part of the natural daily ecology of the aquatic resource. As DO levels drop, some sensitive animals may move away, decline in health or even die.

Chemical Oxygen Demand (COD)

Chemical oxygen demands (COD is an important parameter for oxygen required to degradation of organic matter. Table 2 shows the chemical oxygen demand of selected water samples. COD of well water, bore well water and lake

water were 33.8mg/L, 33.8 mg/L and 31.3 mg/L High level of biochemical oxygen demand and Chemical oxygen demand is due to the presence of chemicals that may be organic or inorganic caused by the inflow of domestic, livestock and industrial waste that contains elevated levels of organic pollutants (Abaidooand *et al*., 2015)

Heavy Metals

Heavy metals in drinking water pose a threat to human health. Populations are exposed to heavy metals primarily through water consumption, but few heavy metals can bioaccumulate in the human body (e.g., in lipids and the gastrointestinal system) and may induce cancer and other risks (Chowdhury *et al.,* 2016). Heavy metals like Fe and Mn were non detectable in all the selected water samples.

Table 1: Physical Parameters of selected water samples

S.No	Parameters	Samples		
		Well water	**Bore water**	**Lake water**
1.	Colour	Colourless	Colourless	Colourless
2.	Odour	Odourless	Odourless	Odourless
3.	Turbidity (NTU)	0.7	0.8	0.5
4.	pH	6.85	7.08	7.66
5.	Electrical Conductivity (µS/cm)	3003	7261	1035
6.	Total Dissolved Solids (mg/L)	2042	4938	693

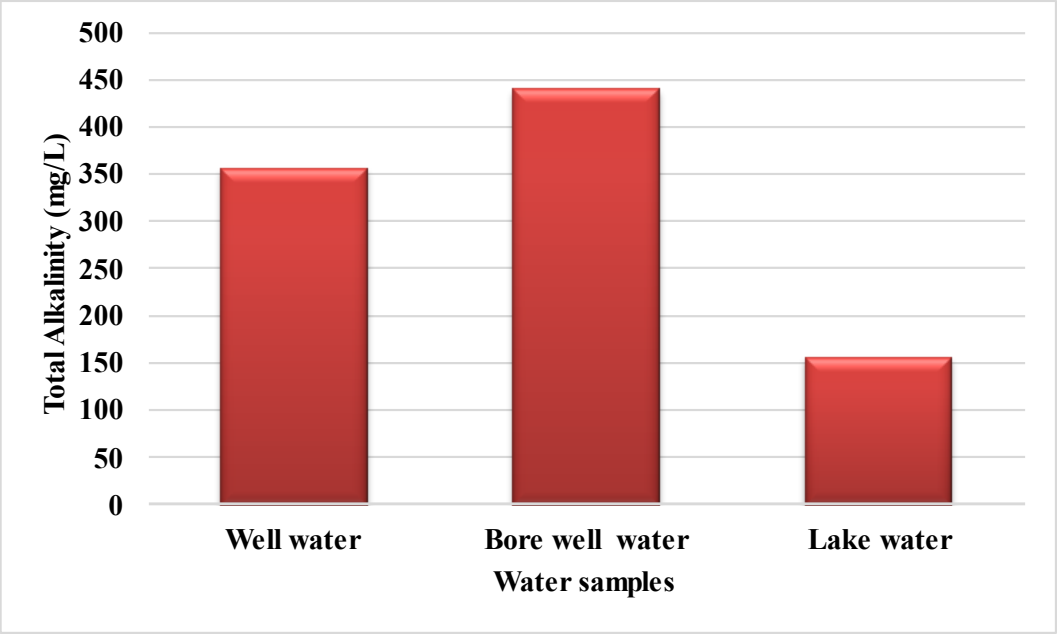

Figure 2: Total alkalinity of selected water samples

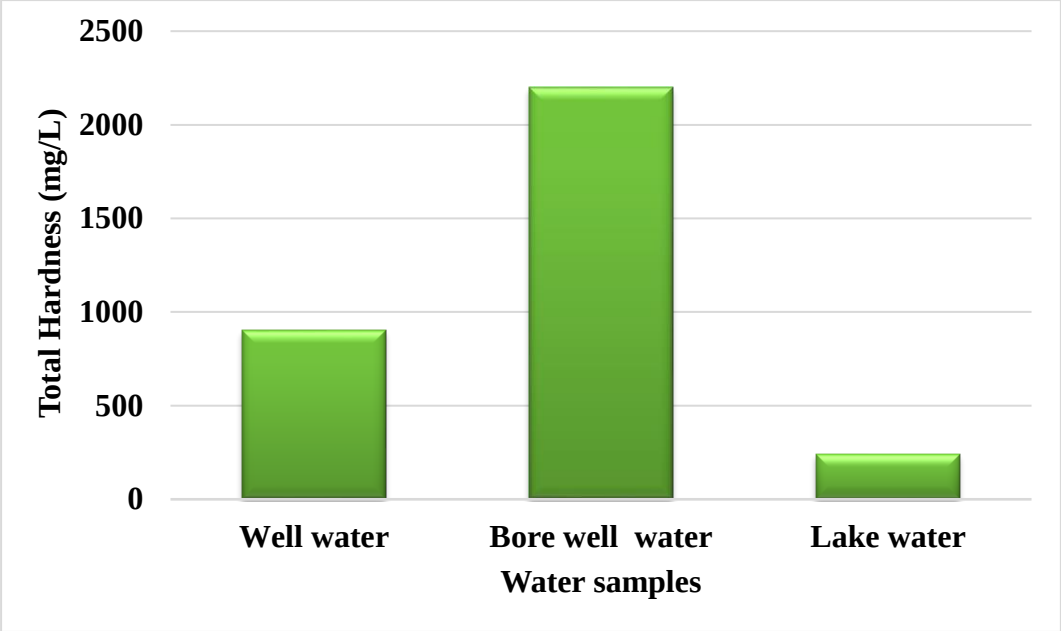

Figure 3: Total hardness of selected water samples

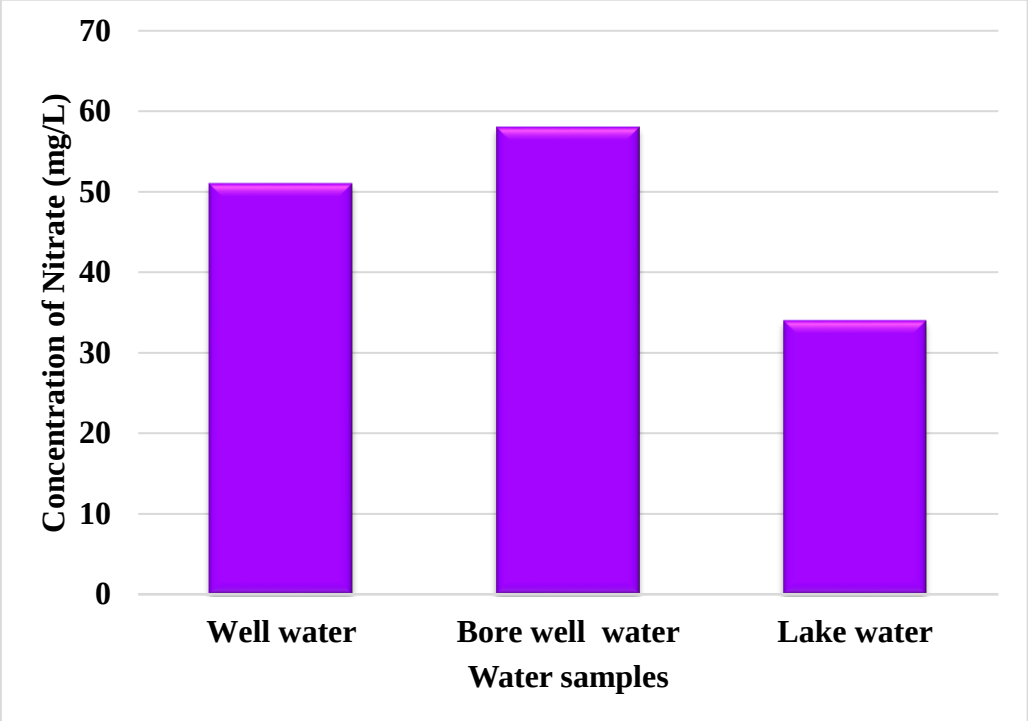

Figure 4: Nitrate content of selected water samples

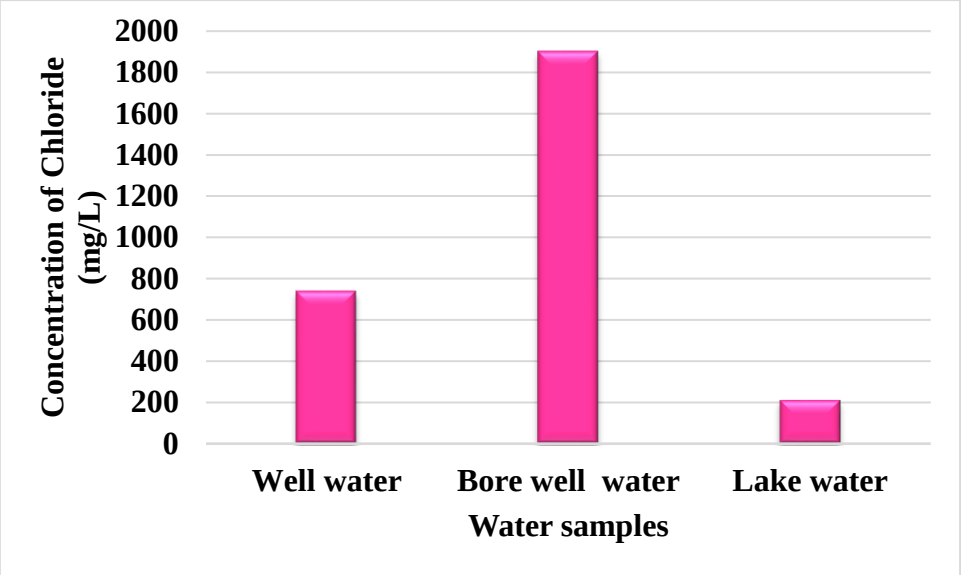

Figure 5: Chloride content of selected water samples

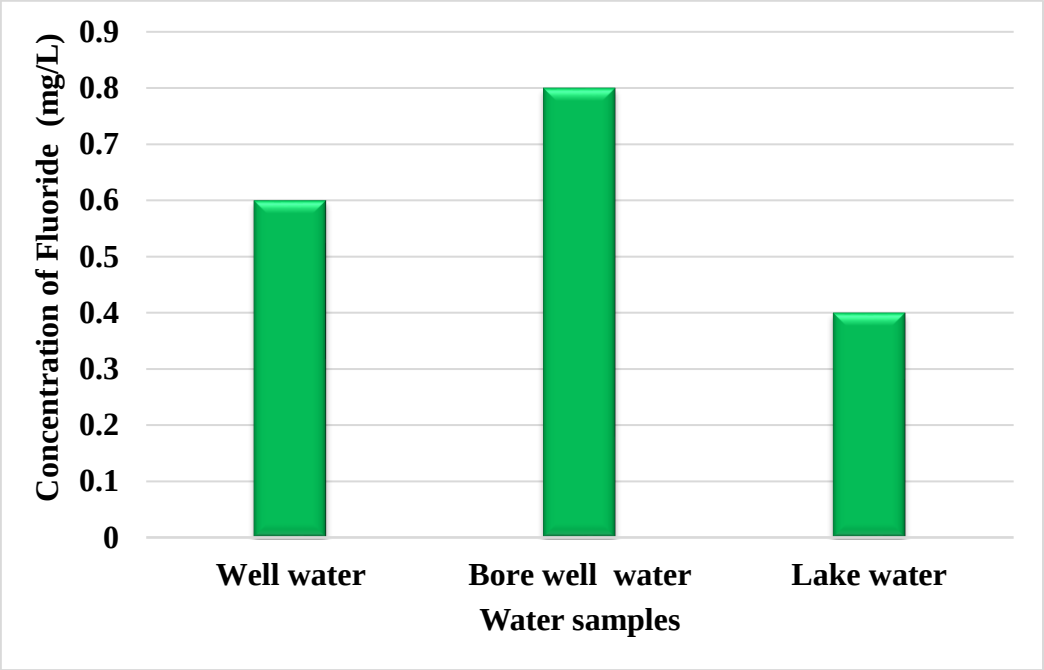

Figure 6: Fluoride content of selected water samples

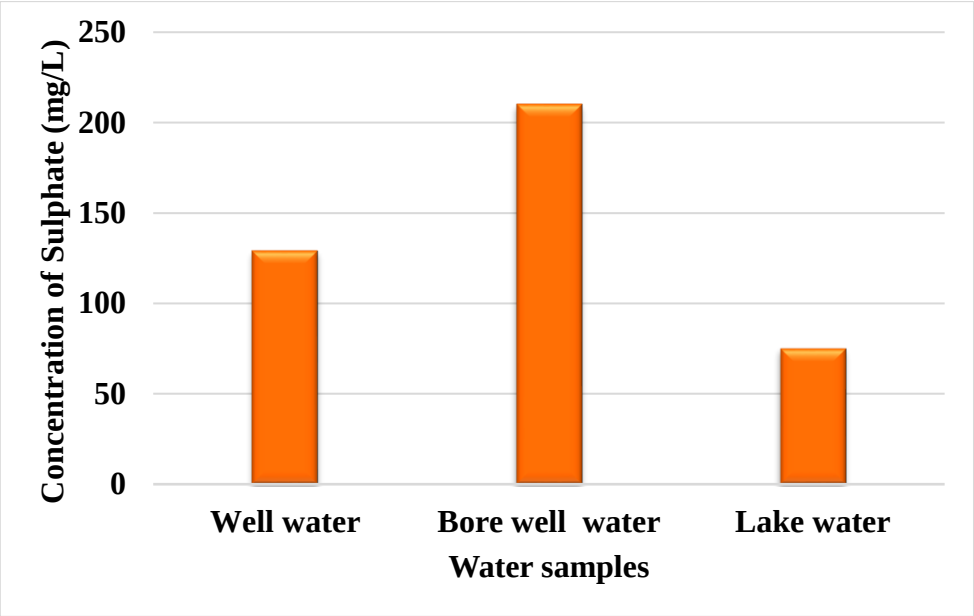

Figure 7: Amount of sulphate content of selected water samples

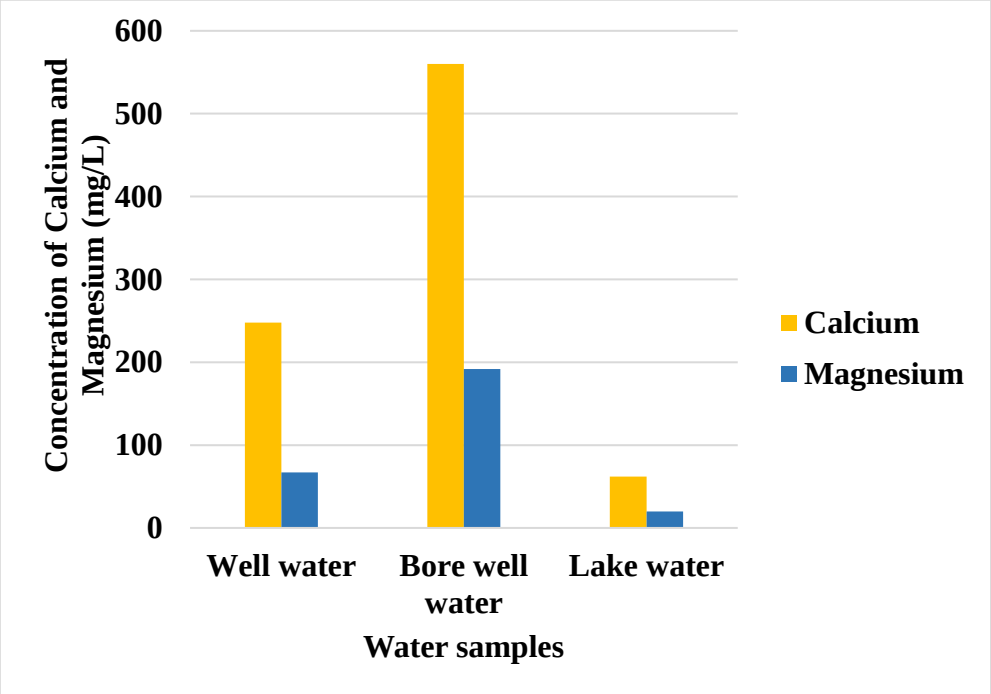

Figure 8: Amount of calcium and magnesium of selected water samples

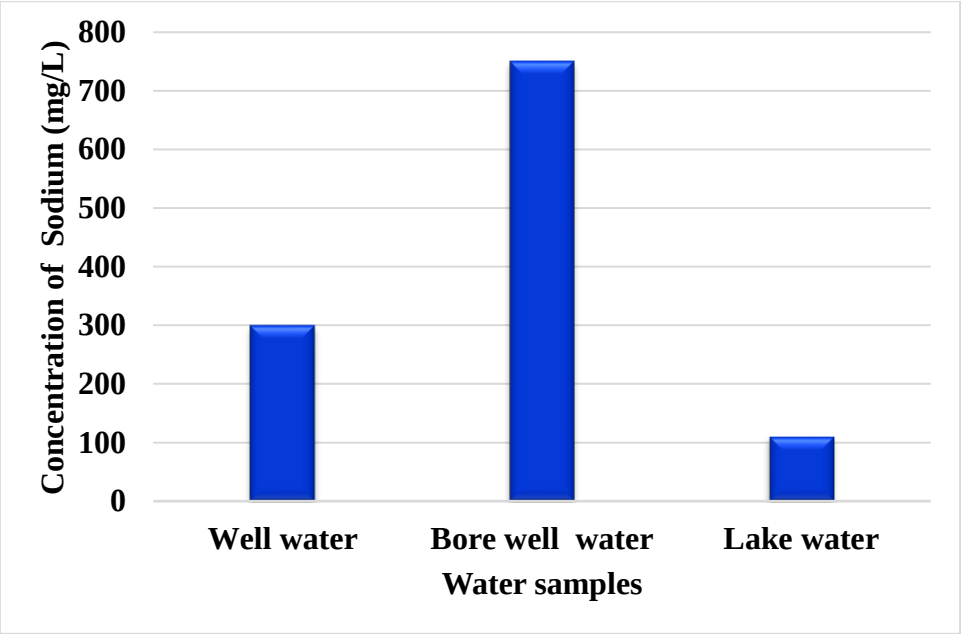

Figure 9: Sodium content of selected water samples

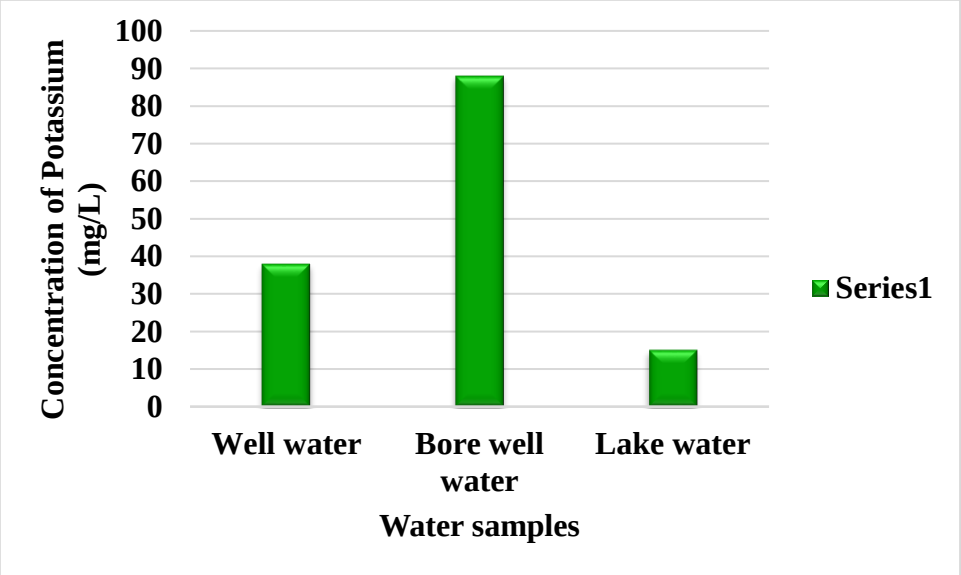

Figure 10: Potassium content of selected water samples

Table 2: BOD and COD of selected water samples

S.No	Water Samples	DO (mg/L)	COD (mg/L)
1.	Well water	0.25	33.8
2.	Bore well water	0.27	33.8
3.	Lake water	0.1	31.3

SUMMARY AND CONCLUSION

SUMMARY AND CONCLUSION

In the present study characterization of the physiochemical parameters of three different water samples such as well water, bore well water and lake water from Keelavaippar village of Thoothukudi district was carried out. To assess the quality of water each parameter was compared with the standard desirable limits prescribed by Bureau of Indian Standard (BIS). From the study it can be concluded that groundwater is safe for drinking purposes from the point of view of levels of pH, electrical conductivity, sodium, potassium, fluoride and sulphate. But other parameters are found to be higher than maximum permissible IS values and the total hardness varied in between 240-2200 mg/L, which indicates that all the water samples are very hard. Fortunately, heavy metals like iron and manganese are absent in al the tested water samples. The values of the parameters are applicable to changes, according to the time of collection, because the water in lake is always flowing thus giving new samples every time.

Hence, the current study suggested that water samples from studied area were not suitable for drinking, washing and any other purposes. So, it is recommended to the cantonment localities to soften the tube well water before consumption. Proper planning should be executed before drawing the well water and bore well water. People awareness campaigning should be organized by the government and non-government organization. Ground water must be pretreated so as to ensure less health threats. Programs should be implemented to monitor the bore wells and handpumps exceeding the limitation of guidelines. Domestic waste affect groundwater quality so proper monitoring is needed.

Sanitation system must be improved. The benefits of cleanliness on human health need to be understood. Chemical treatment can be given to separate out unwanted dissolved chemicals materials.

Finally, it is proposed that the further research should be carried out for detailed mapping and hydrological studies for existing water sources to show flow lines and hydrogeochemical survey in that area. It is also necessary to find out the source of contaminants which is due to soil types, industrialization, water chemistry and other human activities.

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SUSTAINABLE COLOURATION OF COTTON FABRIC USING NATURAL DYES EXTRACTED FROM PLANT SOURCES

A dissertation submitted to

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MASTER OF SCIENCE IN BOTANY

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CERTIFICATE

It is certified that this short term project work entitled **"SUSTAINABLE COLOURATION OF COTTON FABRIC USING NATURAL DYES EXTRACTED FROM PLANT SOURCES"** submitted to **St. Mary's College (Autonomous)** affiliated to **Manonmaniam Sundaranar University** in partial fulfillment of the requirements for the degree of **Master of Science in Botany** and is a record of work done in the Department of Botany, St. Mary's College (Autonomous), Thoothukudi during the year 2022 – 2023 by **Lawanya. G**, Reg. No. 21APBO05.

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I do hereby declare that this dissertation entitled **"SUSTAINABLE COLOURATION OF COTTON FABRIC USING NATURAL DYES EXTRACTED FROM PLANT SOURCES"** submitted by me in partial fulfillment for the award of the degree of **Master of Science in Botany**, is the result of my original and independent work carried out under the guidance of **Ms. S. Pauline Jenifer, M.Sc., B.Ed., SET,** Assistant Professor of Botany, St. Mary's College (Autonomous), Thoothukudi and it has not been submitted elsewhere for the award of any other degree.

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Station: Thoothukudi

LAWANYA. G

Date: 12-04-2023

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LAWANYA. G

CONTENT

Introduction

INTRODUCTION

Natural dyes replaced the synthetic dyes in the past year due to its eco-friendly in nature. Now it is currently being favoured by consumers because of its benefits like eco-friendly, less toxic, usage in children's wear, favour to wearer skin etc.. Natural dyes which are used in colouring of clothes, food substrate, leather, etc.. Natural dyes can be obtained from various parts of plants including leaf, barks, roots, flowers. The natural dyes were extracted from natural plants for alternate to synthetic dyes. The parts of some trees like, ebony tree, turkish red pine tree, mangrove tree, eucalyptus tree, walnut tree, sirisis tree and etc., have the ability to dye the textile materials. Mostly, the bark of the tree able to produce dye. The synthetic dyes create bad impact on water resources. For processing of a one ton of textile material, might have to use as much as 230-270 tons of water. The natural dyes are great alternators of synthetic dyes. Natural dyes are not commercially used one which is fully consumer favour (Ansari and Thakur, 2000).

The natural dyes obtained from fruits, vegetable, leaves, bark, flowers, roots etc., are less toxic and allergenic than synthetic dyes. Using natural dyes to products becoming important for avoiding the skin related issue. Textile chemical processing industry is one of the major environmental polluters. There are two ways to control the environmental pollution of the textile processing industry. The first one is to use eco-friendly products like natural dyes, mordants etc., and another one is to build a proper and effective effluent treatment plants. Natural dyes produce the different and unique shades that can be obtained from roots, insects, minerals (Bhattacharya and Santhosh, 2002). In spite of various advantages of natural dyes, it also has some disadvantages like, availability of dye source (Tree, Vegetables) might be found for some particular

season and poor fastness properties. The natural dyes are unable to form the strong bonds with textile materials, it needs a crosslinking agent or intermediate materials like mordant to improve the substantivity of the dyeing. Various mordants are used to improve the fastness properties of the natural dyes. Natural and synthetic mordants are available in the market. It is better to use natural mordant for eco-friendly. The banana sap is a natural mordant used to improve the fastness properties of the dyeing process and it is a cost effective mordants (Bhyuyan *et al.*, 2012).

Presently there is a great demand for the use of natural colours throughout the world due to non-biodegradable and carcinogenic nature associated with synthetic dyes (Chungkrang and Bhuyan, 2020). They are not produced any undesired by-products and at the same time they help in regenerating the environment, therefore natural dyes are the safe dyes (Ahuja *et al*., 2015). Government of Germany was the first to take initiative to put ban on azo-dyes for manufacturing, dyeing and importing textiles and other consumer goods dyed with these dyes from January 1, 1995 by the act of German Legislation (Consumer Goods Ordinance). Netherlands followed a ban with effect from August 1, 1996 on similar lines. European Union is likely to impose ban on these toxic dyes shortly. India has also banned the use of specific azo-dyes and under notification sufficient legal teeth‖ had been given for taking panel action against those who use these dyes (Kapoor and Pushpangadan, 2001).

Natural dyes are important since they are better than synthetic dyes in many ways. Undeniably, the natural dyes are healthier products, purely because they do not comprise chemicals damaging to fitness. These dyes are collected from nature and no need to apply manufacturing process to prepare them. These dyes are easily decomposed in nature after using and they do not pollute the environment while

destroying them after end use. The natural dyes are used mainly in coloration of textiles, food, drugs and cosmetics. Small quantities of dyes are also used in coloration of paper, leather, shoe polish, wood, cane, candles and such other products requiring coloration (Gulrajani, 2001). Natural dyes are known to be used since historic times for colouring food substrate, leather, as well as common textile fibers like cotton, wool and silk. However due to the advent of synthetic dyes and their good fastness properties in comparison to natural dyes, the use of natural dyes have suffered drastically.

In the present scenario there has been a rise in concern of eco-friendliness and sustainability of the products used by the consumers for which natural dyes are again starting to experience slight rise in popularity (Samanta and Agarwal, 2009). Now a days environment is deteriorating at an alarming rate especially by industrial pollution. Dyeing of paper industry has also in race of deterioration of environment, when traditional natural dyes were replaced by synthetic dyes. Synthetic dyes are designed to resist chemicals and improve the quality of the product but are obstinate in the environment (Aminoddin and Haji 2010). Most synthetic dyes are carcinogenic, highly toxic in nature and cause allergic dermatitis, skin irritation and mutation to humans (Srivastava *et al*., 2004). In recent years, environmental contamination by synthetic dyes is a serious problem due to their negative ecotoxicological effects (Saha *et al*., 2010).

The contamination of water due to synthetic dye molecules causes damage to the environment and has adverse effects on public health (Kiran *et al*., 2009) The discharge of highly coloured synthetic dye effluents into inland and coastal waters is an environmental problem of growing concern (Padmavathy *et al.,* 2003). During the dyeing process loss of colourants to the environment can reach 10-50 percent (Ben e*t*

al., 2012) Textile dyeing industry at present uses excessive number of synthetic dyes to meet the required coloration of global consumption of textiles due to cheaper prices, wider ranges of bright shades, and considerably improved fastness properties in comparison to natural dyes. But the production of synthetic dyes is dependent on petrochemical source, and some of these dyes contain carcinogenic amines. The application of such dyes causes serious health hazards and influences negatively the eco-balance of nature. Moreover, many countries already imposed stringent environment standards over these dyes. For instance, Germany has banned the azo dyes. In this situation, a higher demand is put towards the greener alternatives or agricultural residues. As a result, natural dyes are among the promising options for developing a greener textile dyeing process and such interest is reflected to the increased number of recent publications. Plant leaves are potential sources of natural dyes because of their easy availability and abundant nature (Deo and Paul, 2000).

In the present scenario, due to the excelling advantages of natural dyes, it is becoming an enticing option over synthetic dyes. Natural dyes are biodegradable, non-toxic, environment friendly, aesthetically appealing resulting in employment generation and utilization of wasteland, easy extraction of colours by boiling the plants, berries, leaves, bark or flower heads in water, Focus towards the utilization of the vast diversity of natural resources of colour pigments for their use in food materials, pharmaceuticals and textiles, in place of their synthetic counterparts. This trend is aimed at safeguarding human health as well as protecting and prolonging life on Earth. Detailed scientific studies with natural dyes have established that in most cases their properties are comparable to those of synthetic dyes. Therefore, if natural dyes have to be commercialized, they need to conform to the same stringent standards of

performance that are applied to synthetic dyes. It thus follows that much more research and developmental effort needs to go in this area. The technology of extracting and utilizing natural dyes in the modern textile industry is relatively new and is still being improved upon. In India, textile manufacturers are not yet finding proper enticement in switching over to natural dyes for being more expensive than their synthetic counterparts. Synthetic dyes have replaced almost all the natural colouring matters. However, the art and craft of producing natural dyed textiles is being practiced in every corner of the country by a handful of expert crafts-persons. (Balut *et al*., 2014)

Dyes derived from natural materials were the only dyes available to mankind for the colouring of textiles until the discovery of the first synthetic dye in 1856. In the last decades of the 20th century, environmental issues revived consumer interest in natural dyes. Natural dyes suffer from the inherent disadvantages of standardized application and standardization of the dye (Agarwal *et al*., 2003).

The art of using natural materials for colouring textiles was very old in India, Egypt and Mesopotamia. The use of natural dyes for textiles other than body tattooing and cave painting was in boom till the starting of 19th century. Now the use of these natural sources for various uses is taking a boom. Due to the lack of proper documentation, identification and use of the natural sources became a problem (Shahin *et.al*., 2014).

The specific objectives were to analyze the aqueous extraction process of the dyes, to explore the possibilities of producing fashionable hues from the dyes using different mordants, to compare between unmordanted and mordanted dyed fabrics, to analyze the color values, and to assess the color fastness properties of dyed fabric. Dyeing can be carried out in an alkaline bath, acidic bath or in a neutral bath. There are

various reports available on different methods of mordanting on different fibers such as cellulosic, protenic and synthetic for dyeing with different natural dyes. Various kinds of shades like black to brown, green to yellow to orange, etc can be obtained by application of different mordants. Dyeing of cotton and silk with henna, indigo, marigold etc is reported. (Gulrajani *et al*, 1992). There is a growing interest in the revival of natural dyes in textile colouration. In contrast, natural dyes are environmental friendly, exhibit better biodegradability and generally have a higher compatibility with the environment than synthetic dyes. The process is economically viable as the raw materials are available at low cost and so cost of production is also very low. Similar findings were reported in Marigold, China rose and Bixa flower (Ibrahim *et al*, 1997).

The main advantage of natural dyes are,

- Minimal Environmental Impact – Because they come from natural sources, natural dyes are not harmful to the environment, which makes it so appealing for consumers. Natural dyes are biodegradable and disposing them don't cause pollution.
- Renewable – Natural dyes are obtained from renewable sources that can be harnessed without imposing harm to the environment.
- Color pay-off – If you're going for a soft hue or soothing shade, natural dyes can help you achieve that look.
- Safe – Some natural dyes, such as carmine found in lipsticks, will not cause harm or health problems when ingested.
- No staining -Numerous natural dyes have the advantage that, while having low wash fastness ratings, they do not stain nearby materials during washing since the dye is not strongly attracted to the cloth.

The present study shows that the plant samples are collected from the excessive

parts apart from the main use. In *Jatropha* the flowers are collected, mainly this plant was used to make biodiesel from the seeds and leaves the rest of the parts will be considered as waste material in industries. In *Vitis*, the damaged fruits are collected among the fresh fruits. In *Plectranthus*, the dry leaves are selected for making dye. And in *Nerium*, the flower which was withered in the ground are collected to make a dye. The collected plant samples will not cause any pollution to the environment. Economically it was comfortable for doing research. More samples are available in local areas. They are eco-friendly.

Scope and Objectives

SCOPE AND OBJECTIVES

One of nature's greatest treasures is colour. They surround us and are found all throughout the planet. Fabrics are naturally colourless or creamy/off-white. Thus, colouring fabrics makes sense to make them stylish and appealing. The term "colouring" refers to substances having distinctive colours (functional groups that bear colour) that can add colour to other substrates like food, textiles (fibres), fabric, paper, etc. Natural dyes coming from various sources have been used by humans since the beginning of humanity. Natural textile dyeing is both an art and a science, and it is fundamental to human culture. Both experience and technique are necessary for a perfect performance. Natural dyes are employed in paintings as pigments in addition to for colouring textiles.

By keeping the above scope in mind the following objectives were set for the present study.

- Extraction of dyes from selected samples using Aqueous, Acidic, and Alkaline medium
- Dyeing of cotton cloth and thread
- Dyeing of extracted dye with different mordants.
- Assessment of light fastness and washing fastness for each dyed sample.
- Spectrometric analysis of the extracted dyes.
- Preliminary phytochemical screening of dyes

Review of Literature

REVIEW OF LITERATURE

Plants are one of the main sources of natural textile dye. The present study was employed to identify the fabric dyeing capacity of different plant samples using alum as the mordant. The extracted dye was successfully applied and it imparted soothing shades on the selected textile fabric (Abdu Zubairu *et.al.,* 2015).

Artificial dyes are toxic, harmful and carcinogenic. There is an increase in the demand of natural dyes due to their therapeutic properties and no known side effects. The current research aimed at extracting dyes of different colours from natural plant sources. The dyes were tested for their anti-microbial ability and were found to be inhibitory to common organisms like *S. typhi, C. diphtheriae, S. aureus*, etc. Phytochemical tests confirmed presence of several important metabolites like Phenols, Tannins, Terpenoids, and many more. Dyes tested for anti-oxidant activity using the FRAP assay and antioxidant levels ranging (Sivakumar *et.al*., 2011).

Natural dyes are biodegradable, eco-friendly and free from hazardous chemicals. The study was conducted to extract the natural dye from the leaves of *Polyalthia longifolia*. The optimum time for dye extraction was found to be 60 min at 90 ¼C and the dye yield and M: L ratios were 18% and 1: 10 respectively. Phytochemicals found in the extract were alkaloids, triterpenoids, tannins, saponin, anthraquinones, and glycosides. The colour fastness to wash properties of the dyed silk fabrics using different mordants were good (Sun *et al*., 2015).

Dyeing is an ancient art practiced during the Bronze Age in Europe. Synthetic dyes impart strong colors but cause carcinogenicity and inhibition of benthic photosynthesis. Natural dyes from *Beta vulgaris* (Beetroot), *Spinacia oleracea*

(Spinach), *Ixora* (Jungle geranium), *Brassica* (Purple Cabbage), and *Tagetes erecta* (African Marigold) were used in the study. Wool takes up less dye than cotton (Adeel *et.al*.,2009).

Synthetic dyes are harmful for humankind and cause skin diseases and disorders. The demand for natural dyes which do not cause any side effects rose. Dyes extracted from plant source (bark, flowers, leaves and fruits) are more preferable than other natural sources in textile industry. Natural dyes also provide UV protection (Teli *et.al*.,2014).

Natural colourants are used mainly for textile dyeing. Less research is done in this field. For red shades beetroot dye is the potential source. In this article we review the extraction and application of natural dye with special reference to Beetroot Dyes and their application on textile products (Vankar, 2009).

The screening of phytochemicals was carried out to analyse the chemical compositions of selected plant extracts. *Alkanna tinctoria, Quercus infectoria*, and *Thuja* were chosen for laboratory experiments in the present investigation and revealed to note the presence of tannins and phenols (Vasundhara *et.al.,* 2016).

Pterocarpus santalinus powder extract was examined as a natural textile dye. The selected cotton fabric was dyed with the extract at 800 C for 60 minutes using dye bath of pH 9 and MLR of 1:40 to produce good colour strength (Venkidusamy and Arunkumar, 1997).

Cotton fabrics were dyed with natural dyes derived from the crude bark extracts of *Albizia coriaria, Morinda lucida, Syzygium cordatum and Vitellaria paradoxa* from Mukono and Mbale districts of Uganda. Natural dyes are becoming important in

industry for their less toxic tendencies as compared to synthetic dyes (Ashis and Adwaita, 2011).

Catechu is a natural dye used as a dye and an antibacterial substance. Cotton and viscose fabrics were dyed with catechu natural dye at different conditions such as pH, dyeing time and temperature. The dyed fabrics have higher antibacterial activity and are resistant to washing up to 25 times in a row (Bhattacharya *et.al*.,2002).

There is an attempt to test the inherent bio colorant property and medicinal characteristics of *Castanopsis indica* via green technique using water and ethanol as solvent. The results revealed the color of fabric with water extracted *C. indica* dye with organic and synthetic mordant is to be bright compared to dull color from ethanol extract (Bhuyan and Saikia, 2004).

Lemon leaves are utilized in many cultures. The health benefits of citrus leaves are mainly attributed to the presence of bioactive compounds. Phytochemicals present in the leaf powder of citrus include terpenoids and anthocyanin. Preliminary analysis of sample citrus leaves shows the pH, weight of moisture, content and ash content. Infrared spectroscopy was done to determine functional groups in molecules (Babel and Gupta, 2016).

The phytochemical analysis reveals the presence of phytoconstituents that could be useful as dye for smart windows in the chromophore of the plant. The dye pigment grown on glass from the extracts exhibited low absorbance and high reflectance at transmittance set at 400 nm. (Chattopadhyay *et.al.*,1997).

Red rose is a one of the most important ornamental plant mainly growing in garden and rich in red and pink pigments. The dyeing pigments present in flowers of

red rose were extracted by using four different solvent extraction methods .The three different mordant were used. The result revealed that different shades of pink an yellow colour were obtained from the dye to mordant .The colour dye extracted from red rose flower can be used for coloration of cotton ,silk and wool fabrics **(**Gopika *et al.,* 2018).

The dye potential colourants obtained from the marigold evaluated by colorin pure cotton fabrics ,yarns of the pure cotton and wool various metal salts were used as mordant extract dye on the fabrics and yarns fastness tests of dyed clothes was undertaken .The colour shades differences .The maximum strength of the dye was found in the ethanol-water-mixture solvent .The surface colour of the dyed fabrics and yarns was not affected by washing .The different colour shades were obtained for mordant used .Good light fastness ,wash fastness and rubbing fastness were observed of fabrics mordanted ferrous sulphate. These finding reveal that tageteserectal can be used in textile industry for dyeing purpose **(**Selvam *et.al*., 2015).

Merina *et al.,(*2016) made an attempt has been made to utilize petal parts of chrysanthemum flower and peel of Badam fraie to extract dye for application on the fabrics .The natural dye was extracted from the flower of chrysanthemum with two different colours. To the experimental results the performance was better with that all the natural dye which can be the better alternative of synthetic dyes.

According to Thilagavathi and Rajendrakumar (2005) *Celosia argenteal* is a species of the genus celosia belonging to the family amarantheceae .The present work highlights the use of *Celosia argenteal* aqueous flower extract as acid base indicator in acid base titrations .The equivalence point obtained by the flower extract .The natural

indicator was found be a very useful ,economical simple and accurate for the acid.

Singh *et al*., 2006 founded that natural dyes are obtained from natural sources such as plants ,insects and minerals .All the plant based dye sources are more important for textile dyeing as it provides both dye as well as frequence. This paper review the available for floral dye sources application and extraction of colourant from flower and effect of different mordant.

Samanta and Agarwal (2009) studied the dyeing of bleached jute and cotton fabrics with mordants using Jack fruit wood extract. It is observed that the application of 10-20 per cent myrobolan followed by 10- 20 per cent of Al2(S04)3 or FeS04 in sequence have been identified as two most prospective mordanting systems. The study on the effect of dyeing process variables on surface colour strength indicates that the 90 min dye 33 concentration, and 15 gpl common salt are the optimum values with minor differences among the different fibre mordant systems studied. Colour fastness to washing, rubbing and exposure to sunlight, in general, and dyeing-pH sensitivity, in particular, for selective fibre-mordants-dye systems have also been assessed and compared. Dyeing at pH 11.0 for both the double pre-mordanting systems offers overall good colour fastness properties.

Vasundhara *et al.,* (2016) extracted organic colour from kokum (*Gracinia indica*) fruit rind to dye textiles such as cotton, jute, silk and polystyrene. The fruit rind is used in three different forms for dye extraction: 1. fresh rind, 2. dry rind (the fresh rinds dried in hot air oven at 50ºC for 5 days), 3. sugar rind (the rinds are soaked in sugar for a week and kokum juice was extracted and dried in hot air oven at 50ºC for 5 days. The extracts are found to be rich in anthocyanins and were quantified as 79.93,

85.03 and 7.83 mg/kg, respectively. One set of fabrics were dyed without mordant and the other set was mordanted 20 with 2% ferrous sulphate for 30 minutes; then all the fabrics were dyed with the dye extracts for 60 minutes. Mordanted fabrics produced better shades than un-mordanted samples. Jute exhibited best shades than other fabrics dyed.

Mamta *et al*., (2017) defines "Cosmetotextiles" as a concept that (fabric) releases cosmetic ingredients to the skin of the wearer. The functions of cosmetotextiles include moisturizing, UV protection, skin whitening, anti-wrinkle treatment, aromatic, refreshing and relaxing. The methods followed to impart cosmetic effects on textiles are dyeing and finishing (microencapsulation). The cosmetic ingredients include bio-active materials like aloe vera, ginseng, chitosan, squalene, fruits, flowers, extracted essential oils, vitamin E, etc. The cosmetotextiles might have future scope if developed through research.

The oldest natural dye for textile, used by humans is indigo dye obtained from the plant *Indigofera tinctoria* opines Samanta *et al.,* (2009). It is the most popular dye that ruled the world from the middle of seventeenth century to the end of nineteenth century. The Indian subcontinent has played a major role in the history of indigo. The indigo plant grows favourably in tropical climate rather than temperate climates. The emergence of the indigo plantations in the seventeenth century has been a marked phase in the history of indigo states Kumar (2012). Indigo plantations were introduced in India during eighteenth century and in the beginning of nineteenth century. Bengal became a major supplier of indigo to the entire world. Then the indigo plant was cultivated in different parts of India. Indigo dye became more popular with the denim fabrics manufacture.

Sonia John *et al.*,(2018) stated that the extracts of *Ixora lutea* (Rubiaceae) as an effective antimicrobial agent and a natural dye. The preliminary phytochemical analysis of the stem and leaf extracts in various solvents (polar and non-polar) revealed the presence of alkaloids, carbohydrates, phenols, tannins, saponins, reducing sugar, triterpenoids and steroids. The flower showed great antibacterial and antifungal properties. the extraction of natural dye from the flower and their application on textiles. The extracted dye along with the mordants gave varying shades of colors on the fabric.

Sandeep Bains *et al*., (2003) studied the dyeing of cotton with peach leaves using different mordants. The dyeing was carried out at optimized dyeing conditions namely, dye material, extraction time, dye material concentration and dyeing time and using combinations of mordants such as alum: chrome, alum: copper sulphate, alum: ferrous sulphate, chrome: copper sulphate, chrome: ferrous sulphate, copper sulphate: ferrous sulphate in the ratio of 50:50, 25:75 and 75: 25 respectively. The dyed samples were evaluated for colour fastness to washing, rubbing, perspiration and light. The dyeing of cotton at optimized conditions resulted in good to very good colour fastness to light (rating range 5-6), fair to excellent colour fastness to washing (rating range 3-4/5), good to excellent colour fastness to rubbing (rating range 4-5) and poor to fair colour fastness to perspiration (rating range 2-3) as found by evaluation of the colour fastness of the dyed samples by prescribed methods. The shades obtained were khaki, greenish khaki, bamboo light, platinum blonde, beige, shallow to dark shallow, greyish military green, mouse grey and brownish grey to dark brownish grey.

Sudhakar and Ninge Gowda (2005) analysed the degummed silk fabric dyed with the flower extract of *Spathodea companulata* along with varying concentrations of different mordants. Colour values with respect to K/S and CIE L* a* b* were

influenced by the mordants and the mordanting techniques. Pre-mordanting was found to be better in the case of stannous chloride whereas meta-mordanting was found better in case of potassium aluminium sulphate and tannic acid with respect to colour values. The unmordanted dyed samples exhibited good fastness to washing, rubbing and perspiration, barring light. A very slight improvement in fastness to light was recorded with the use of tannic acid as mordant.

Susan *et al.,* (2006) studied the effect of mordants alum, chrome ferrous sulphate and copper sulphate on colour fastness properties of cotton dyed with Kilmora dye. It was found that different mordants improved the colour fastness of Kilmora dye on cotton.

Silk fabric was dyed with natural dye extracted from eucalyptus leaves by Mongkholrattanasit *et al*., (2011). The fabric was dyed with mordants such as alum, ferrous sulphate, copper sulphate and stannous chloride and also without any mordant. The optimum results were obtained with dyeing at 90ºC for 60 minutes at pH 4. The samples dyed with ferrous sulphate mordant produced dark greyish-brown shade while with other mordants yellowish-brown shades were obtained. The dyed samples exhibited fair to good fastness properties and good to excellent UV protection properties. Sharma and Jahan (2003) used the natural dye obtained from barks of peepal (Ficus religiosa) tree to dye silk fabrics with myrobalan and cow dung as natural mordants and metal mordants namely, alum, ferrous sulphate, potassium dichromate, stannous chloride and nickel sulphate. The dye produced different shades with each of the mordants mentioned above and the fastness properties were found to be good. The dye is free from heavy metals and hence free from skin problems. Punrattanasin *et al,*. **(2013**) dyed silk fabrics with natural dye extracted from mangrove bark with mordants

like alum, ferrous sulphate, copper sulphate and stannous chloride. The dye produced different shades with different mordants and the optimum conditions for dyeing are 90ºC for 60 minutes at pH 3.

Cotton yarn was dyed by Cristea *et al*. (2006) with natural dyes such as weld (*Reseda luteola*), woad (*Isatis tinctoria*) and madder roots (*Rubia tinctoria*) that produce yellow, blue and red colours respectively. In addition to the dyes, antioxidants and UV absorbers like cafeic acid, gallic acid, vitamin C, phenyl salicylate, benzophenone and vitamin E to enhance the light fastness of the dyed cotton yarn. Since the UV light from the sun is the major cause of fading of dyes, the UV absorbers and antioxidants act as neutralizers for the destructive effect of UV light. Kamel et al. (2011) dyed the cationized cotton fabric with natural dye, cochineal, by conventional heating and ultrasonic methods. The cotton fabrics were cationized by padding the fabric with fresh solution of 3-chloro-2-hydroxypropyltrimethyl ammonium chloride. The cationization and ultrasound technique improved the colour strength of the fabric. Ticha et al. (2016) used natural dyes derived from red cabbage to dye modified cotton fabrics treated with cationic agent at 50ºC for 60 minutes, where cationization improved the dyeability of cotton further. It is identified natural Dye from *Ixora coccinea* the dye potential of the extract was evaluated by dyeing on cotton fabrics under the normal dyeing conditions and tested for their colour fastness to washing properties.. Secondly, mordanting with the different metal salts exhibited variation in color hue because of their ability to form coordination complexes with the dye molecules, which resulted in different shades to cotton fabrics. These findings revealed that the extract of floral petals of Ixora coccinea (Linn). can be used for cotton fabric coloration.

Gopika *et.al* **.,** (2018) stated that Water hyacinth is reported to be used for several

purposes. In this study, the extracts of water hyacinth flowers were tested as a potential source of natural dyes. Three different methods were followed for the extraction of the dye. Potassium dichromate ($K_2Cr_2O_7$), copper sulphate ($CuSO_4$), oxalic acid ($C_2H_2O_4$), stannous chloride ($SnCl_2$) and ferrous sulphate ($FeSO_4$), each at a concentration of 6 % of the dye were used as mordants. The dyed clothes were washed and checked for the fastness. This study proves that the flowers of water hyacinth could be used as a source of natural and eco-friendly dye with potential for a range of applications.

Materials and Methods

MATERIALS AND METHODS

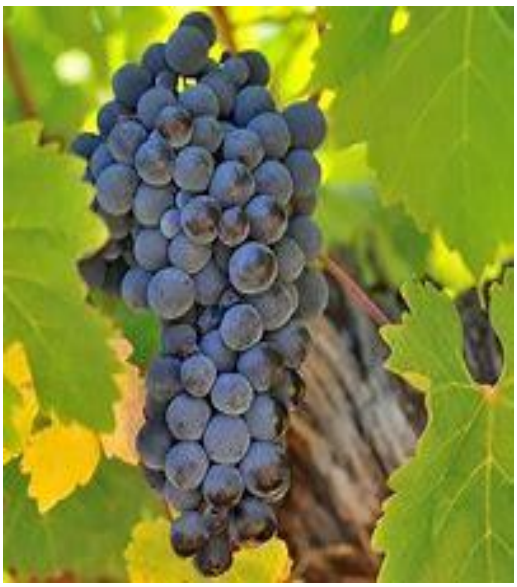
Selection of dye materials:

The dye materials are collected freshly from various locality. Mostly it was collected from the Southern region. The following dye yielding plants were selected for obtaining raw dye materials for this study

S.No.	Botanical Name	Common Name	Family	Dye yielding part
1.	*Jatropha gossypifolia*	Bellyache bush	Euphorbiaceae	Flower
2.	*Vitis vinfera*	Grapes	Vitaceae	Fruit
3.	*Plectranthus scutellarioides*	Painted neetle	Lamiaceae	Leaves
4.	*Nerium oleander*	Nerium	Apocyanaceae	Flowers

Plate 1

Jatropha gossypifolia



Vitis vinifera

Plectranthus scutellarioides

Nerium oleander

Description about the samples:

Jatropha gossypiifolia

Jatropha gossypiifolia, commonly known as bellyache bush, black physicnut or cotton-leaf physicnut, is a species of flowering plant in the spurge family, Euphorbiaceae. The species is native to Mexico, South America, and the Caribbean islands, but is currently spread throughout the tropics. It is declared noxious weed in Puerto Rico and is naturalised in northern Australia, including Queensland where it is listed as a Class 2 declared pest plant. It grows to 2.5–4 m (8.2–13.1 ft) high. The three lobed leaves are purple and sticky when young and become bright green with age. The small red flowers with yellow centres appear in clusters. The flower is represented in Plate 1

Uses:

Several human and veterinary uses in traditional medicine are described for different parts like leaves, stems, flowers etc., The most frequent reports concern its antihypertensive, anti-inflammatory, antiophidian, analgesic, antipyretic, antimicrobial, healing, antianemic, antidiabetic, and antihemorrhagic activities pesticide, insecticide, vermifuge, ornamentation, and even its use in religious rituals.

Vitis vinifera

It is a liana growing tall at a fast rate. Leaves are alternate, palmately lobed, deciduous, with 3 to 5 pointed lobes, coarsely prickly-toothed leaf margins and a heart-shaped foot, 5–20 cm long and broad. The vine attaches to supports by tendrils. The stems, called twigs, grow through their tip, the cauline apex. Their flowers, small and greenish to white, are grouped in inflorescences and their fruits, of different shapes

depending on the subspecies, are berries grouped in clusters. The calyx is single-leaf with 5 short, deciduous teeth. The corolla consists of five petals, fused at the top and base, and then falls off in its entirety. Opposite to the petals there are five stamens interspersed with glands. The upper ovary bears a very short style with a button-shaped stigma. The fruit is a berry and it is shown in Plate 1

Uses:

Grapes are the richest source of anti-oxidant. Grapes are very useful for the eyes. They are also good for liver and boost the immune system. Ripe grapes are good for the heart, soothing and body controlling pitta body energy and pacifying it ill effects. Due to the presence of Vitamin E content it helps to glow the skin.

Plectranthus scutellarioides

The *Plectranthus scutellarioides* is an evergreen perennial herbaceous plant. Stems are almost quadrangular section, tall up to about 90 cm, erect or ascending and hollow. Tiny translucent hairs are present on the stem, branches, petioles and on the foliar lamina. The leaves, quite variable in shape, dimension and colour, are opposite, with a usually ovate shape with toothed edges, 4-14 cm long. The upper page is velvety, very small yellowish glands are present in the lower one. The inflorescences in terminal cymes, up to 50 cm long, carry flowers with an about 1,2 cm long corolla, bilabiate, obtuse and trifid upper lip, of white or pale blue colour, the lower one, acute and semi-bifid, of violet blue colour. The fruit is achenes and the leaves are shown in Plate 1

Uses:

It is used to treat asthma, High blood pressure, dry eyes, erectile dysfunction, heart function, glaucoma and obesity.

Nerium oleander

Oleander grows to 2–6 metres (7–20 feet) tall, with erect stems that splay outward as they mature; first-year stems have a glaucous bloom, while mature stems have a greyish bark. The leaves are in pairs or whorls of three, thick and leathery, dark-green, narrow lanceolate, 5–21 centimetres (2–8 inches) long and 1–3.5 cm (3⁄8–1+3⁄8 in) broad, and with an entire margin filled with minute reticulate venation web typical of eudicots. The leaves are light green and very glossy when young, maturing to a dull dark green. The flowers grow in clusters at the end of each branch; they are white, pink to red, 2.5–5 cm (1–2 in) diameter, with a deeply 5-lobed fringed corolla round the central corolla tube. They are often, but not always, sweet-scented. The fruit is a long narrow pair of follicles 5–23 cm (2–9 in) long, which splits open at maturity to release numerous downy seeds. The flowers are shown in Plate 1

Uses:

Nerium Oleander has traditionally been used in the treatment of cardiac illness, asthma, diabetes mellitus, corns, scabies, cancer, and epilepsy, and in wound healing as an antibacterial/antimicrobial. However, limited quality clinical trials are available to support these uses.

Selection of cotton material:

Cotton threads:

Cotton thread was brought from Durga store, Thoothukudi. Cotton thread is compatible with fabric made from yarn of plant origin such as cotton and linen and for rayon (made from plant substance) because it has similar shrinkage

characteristics. It is not suitable for more synthetics. Cotton thread is best foe sewing the fabrics. Cotton thread has a distinct texture and non-reflective matte finish that allows it to blend into the fabric better.

Cotton cloth:

Cotton cloth was collected from Local textile shop in Thoothukudi. Cotton cloth absorbs approximately 25 times its weight in water, which makes it an easy fabric to dye. The fibres actually breathe by quickly soaking up and releasing moisture. The combination of dye molecule in powdered mix sticks best to fibres that are also very attracted to water. The main molecule is cotton is also known as the polymer cellulose is very attracted to water.

Selection of mordants:

Mordants form the link between dyestuff and fibre that allows the dye with no affinity for the fibre to be fixed. Among the mordant used for fixing natural dyes, metallic mordants are most common therefore copper sulphate and ferrous sulphate were selected for the present work. Mordant not only give the dye the affinity, but in many cases they produce different colours and improve the fastness of the dye. There are many plants which may yield a colour that is brilliant and pleasing but fades easily, unless fixed by using a mordant.

Chemicals used for the study:

Copper sulphate (CuS04) and ferrous sulphate (FeS04) were used as mordant for fixation of the dye and also to obtain different shades. Other chemicals like sulphuric acid (H2S04), sodium hydroxide (NaOH), hydrogen peroxide (H,0,),sodium carbonate

(Na,CO,) were also used for desizing of cotton . For extraction of the dye, sodium carbonate (Na2C03) and hydrochloric acid (HC1) were used for alkali and acid extraction respectively. The chemical used for colour fastness test of the dyed cotton were sodium carbonate (Na,C03), and sodium chloride (NaCl).

S.no	Name of the chemical	Molecular formula	Purpose
1.	Sulphuric acid	H_2SO_4	For desizing of cotton Thread and cloth.
2.	Hydrogen peroxide	17 H_2O_2	For bleaching cotton thread and cloth
3.	Sodium carbonate	Na_2CO_3	For bleaching cotton thread and cloth and extraction of dye
4.	Hydrochloric acid (5%)	HCL	For extraction of dye
5.	Ferrous sulphate	$FeSO_4$	For mordanting
6.	Copper sulphate	$CuSO_4$	For mordanting
7.	Acetic acid	CH_3COOH	For colour fastness test
8.	Sodium chloride	Na CL	For colour fastness test

Methods of dye extraction:

Selected natural dyes were extracted by aqueous, acidic and alkaline media as detailed below.

Aqueous method

2 g dye materials were mixed in 100 ml of water and heated in 80° to 90°C for different time periods. The extract was then filtered through sieve. The extraction time was determined based on maximum optical density of the extract obtained after particular period of extraction.

Acidic method

Acidic solution was prepared by adding of 1 ml of cone. HC1 solution in 100 ml of water. 2 g dye material was then added and heated at 80° to 90°C for different periods. Dye solution was filtered after it got cooled. The optical density of the extracted dye was determined spectrophotometrically.

Alkaline method

1 per cent solution was prepared with the addition of 1 g sodium carbonate in 100 ml of water. 2g dye material was added and heated at 80° to 90°C for different periods. The cooled dye solution was then filtered. Extraction time was optimized depending upon the maximum optical density of the extracted dye solution.

Mordanting method

All the three mordanting methods *viz.*, pre, simultaneous and post -mordanting methods were followed for the present study.

Pre- mordanting method

In this method the yarns were mordanted in the first stage and then dyed in the second. First optical density of the extracted dye liquor was recorded. 5, 10, 65 15 and

20% aqueous solution of Copper sulphate and Ferrous sulphate were prepared by dissolving appropriate amount of CuSO4 and Feso4 in water. Cotton samples were then treated in each of the mordant solutions for various time periods (15, 30, 45 and 60 min.) and then dyed in the prepared dye bath for time variations like 15, 30, 45, 60 and 90 min. for each dye. Optical density of the dye liquor was recorded. Samples were then washed, rinsed and dried in shade.

Simultaneous mordanting:

In this method, the mordants and the dyes were applied simultaneously in the same bath and treatments were done for optimizing dyeing and mordanting time with the help of previously standardized pre-mordanting method. Here, mordant was added to the dye bath during dyeing by lifting yarn and mixed properly and then boiling the yarn in the dye solution with occasional stirring. Optical density of the dye solution was recorded after completion of the treatment. Treated yarn sample was then washed, rinsed and dried in shade.

Post mordanting:

In this method the yarns were first dyed and then mordanted. The method is basically similar to pre-mordanting method and similar concentrations of dyes and mordants were used. The treatments were done for optimizing dyeing and mordanting time with the help of previously standardized pre-mordanting method.

Concentration of mordant :

Four concentrations of each mordant were used for the present work. 5. 10, 15 and 20 percent for alum and 1, 2, 3 and 4 percent for rest of the mordants (copper sulphate and ferrous sulphate). Higher concentration of alum was used as it is less harmful compared to the other mordants. Concentration of mordants were also optimized for the study.

pH of the dyeing media:

Since pH effects the dye absorption and fixation, it has been decided to carry out the dyeing in both alkaline and acidic media. The pH of the dyeing bath is known to effect dye absorption as well as characters of the dyed Cotton. Alkaline condition adversely affects the silk yarn quality but improves dye extraction and dyeing of cotton thread and cloth. Hence, the pH of the dyeing solution was kept at 9.5 for cotton thread and 6.0 for cotton cloth. The pH was adjusted either with sodium carbonate or 5% acetic acid.

Dying time and temperature:

The dyeing time for both cotton and silk were determined based on maximum dye absorption by the yarn tested. The selected dyeing temperature for silk and cotton yams were 60-70°C and 80-90°C, respectively.

Determination of absorption (%)

A fixed concentration of dye solutions was maintained. The absorbance of the solution was recorded before and after dyeing of the samples at particular wavelength in each case. An average of three measurements at each concentration was recorded. The dye absorbance was calculated as given below.

$$\% \text{ dye absorption} = \frac{\text{OD of the dye liquor before dyeing} - \text{OD of the dye liquor after dyeing}}{\text{OD of the dye liquor before dyeing}} \times 100$$

Solid dye content (%) of extracted dye solution:

For determining the solid content of the dye extracts, three extraction media, i.e . aqueous, acidic and alkaline and ethanol solutions were used for extraction of the dye. A measured quantity of extracts was taken in a pre-weighted petridish and the contents

were dried in an oven at 100±5°C till completely dried residue was obtained. The material was kept in a desiccator to cool down and then weighted

The solid content of the extracted dye solution was obtained as follows ;

$$\% \text{ of solid content} = \frac{W2 - W1}{\text{wt of the solution}} \times 100 \text{ wt of the solution}$$

W1 = Weight of the petridish

W2 = Weight of the petridish + solid

Determination of fastness properties of dyed cotton:

All the dyed yarn samples were evaluated for colour fastness to washing, rubbing or crocking (dry and wet), sunlight, and perspiration by the standard procedures.

Procedure

1. The yarn samples were cut into 6 cm long pieces.
2. A set of standards and the specimens to be tested on the card board with an opaque cover made of the same material across one half of each of the standards and specimens were mounted on the exposure rack.
3. The exposure rack was then placed facing to south and sloping at an angle of45°C.
4. The specimens and standards were then exposed simultaneously to light from 9 am to 5 pm.
5. Samples were evaluated for colour change after 48 h of exposure; the intervals of exposure being 8, 16, 24, 32, 40 and 48 h.

Wavelength scan for the selected dyes:

Diluted extracts of dye samples were used for wavelength scanning for each dye, as it is difficult to get maximum optical density value of dye solution obtained by optimized extraction procedure. Dye solution obtained by optimized extraction procedure was diluted 10 times to get an optical density value of around 0.3. The diluted solution was then subjected to wavelength scanning.

Standard wave length (nm) ranges for different visible colours:

Wavelength of light absorbed (nm)	**Absorbed light**	**Visible colour**
400-435	Violet	Yellowish green
435-480	Blue	Yellow
480-490	Greenish blue	Orange
490-500	Bluish green	Red
500-560	Green	Purple
560-580	Yellowish green	Violet
580-595	Yellow	Blue
595-605	Orange	Greenish blue
605-750	Red	Bluish green

Determination of fastness properties of dyed fabrics:

All the dyed samples were evaluated for colour fastness to washing and exposing to sunlight, by the standard procedures.

Colour fastness to light (ASTM Standard, 1968)

For assessing the resistance of the colour of textiles to the fading action of sunlight, the ASTM method (AATCC Method 16B-1964) was followed.

A specimen from the textile to be tested and a standard dyeing or dyeings were exposed simultaneously under specified conditions for sufficient time to produce "just appreciable fading" of the test specimen or the standard. Colour fastness was then rated in terms of the relative fastness of the specimen and the standard.

The dyed samples were cut into 6 cm long pieces. A set of standards and the specimens to be tested on the card board with an opaque cover made of the same material across one half of each of the standards and specimens were mounted on the exposure rack. The specimens and standards were then exposed simultaneously to light from 9 am to 5 pm. Samples were evaluated for colour change after 48 h of exposure; the intervals of exposure being 8, 16, 24, 32, 40 and 48 h.

Interpretation of results

The effect on the colour of the test specimens were expressed in the International Gray Scale by grading the samples 1 to 6.

1 - Very poor

2 - Poor

3 - Fair

4 - Very fair

5 - Good

6 - Very good

Preliminary phytochemical analysis: (

1.Test for Alkaloids :

To 2ml of dye extract, 2ml of concentrated sulphuric acid was added. Then few dops of Mayer's reagent was added. Presence of green colour are white precipitate indicates the presence of alkaloids.

2. Test for Tannins:

To 1ml of dye extract , 2mlof 5% Ferric chloride was added. Formation of dark blue are greenish black indicates the presence of tannins.

3. Test for saponins :

To 2ml of dye extract. 2ml od distilled water was added and shaken in a graduated cylinder for 15 mins. Formation of foams indicates the presence of saponins.

4.Test for flavonoids

To 2ml of dye extract 1ml of 2N Sodium hydroxide was added . Presence of yellow colour indicates the presence of flavonoids.

5.Test for Glycosides :

To 2ml of dye extract ,2ml of chloroform and 10% ammonia solution was added. Formation of pink colour indicates the presence of Glycosides.

6.Test for cardiac glycosides:

To 0.5 ml of dye extract, 2ml of glacial acetic acid and few drops of 5% Ferric chloride was added. This was under layered with ml of concentrated sulphuric acid. Formation of brown ring at the interface indicates the presence of cardiac glycosides.

7.Test for terpenoids :

To 0.5ml of dye extract , 2ml of chloroform and concentrated sulphuric acid was added carefully. Formation of red brown colour at the interface indicates the presence of terpenoids.

8. Test for phenols :

To 1ml of dye extract, 2ml of distilled water followed by few drops of 10% ferric chloride was added. Formation of blue or green colour indicates the presence of phenols.

9.Test for Anthroquinones:

To 1ml of dye extract few drops of 10% ammonia solution was added. Appearance of pink colour precipitate indicates the presence of anthroquinones**.**

10. Test for Phlobatannins :

To 1ml of dye extract, few drops of 2% HCL was added. Apperance of red colour indicates the presence of phlobatannins.

Results and Discussion

RESULTS AND DISCUSSION

The results of the experimental works carried out along with discussion and the results achieved were included in this chapter keeping in view the objectives of the study and the data generated were recorded systematically.

Colour of the dye exacts using different solvents:

The various colour dye extracted from the selected plant samples are given in Table 1 and Plate 2. In *Jatropha gossypifolia* the colour extracted from aqueous solution is light green colour and the colour was not absorbed more in mordant material. From acid solution, the pink colour dye was extracted and light pink colour was uptake in the mordant material and from alkaline solution the dark green colour was extracted and the mordant material absorbed green colour. Similar results were observed and supported positively by Akilesh and Nandita, 2015, during their work in *Jatropha gossypifolia.*

In *Vitis vinifera,* the colour extracted from Aqueous solution is bright purple and the colour was absorbed in mordant material is purple. From acid solution, dark red colour dye was extracted and bright pink colour was uptake in the mordant material and from alkaline solution the dark green colour was extracted and the mordant material absorbed yellowish green. Moreover the colour extract from this plant was about dark purple to purple in aqueous, Dark red to bright pink in acid and dark green to yellowish green in alkaline.

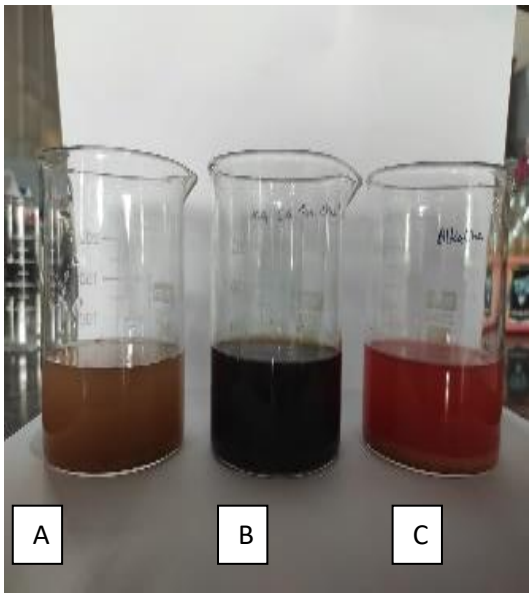
In *Plectranthus scutellarioides* the colour extracted from aqueous solution is brown and the colour was absorbed in mordant material is pale brown. From acid solution, reddish orange colour dye was extracted and pale pink colour was uptake in

Table 1 : Colour of the dye exacts using different solvents

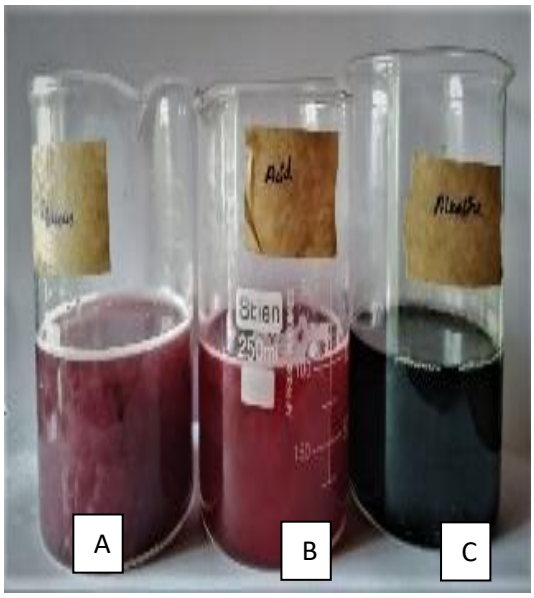
S. No.	Plant sample	Solvent	Colour absorbed before mordant	Colour absorbed after mordant
1.	*Jatropha gossypifolia*	Aqueous	Light green	Pale green
		Acid	Pink	Light pink
		Alkaline	Dark green	Green
2.	*Vitis vinfera*	Aqueous	Bright purple	Purple
		Acid	Dark red	Bright pink
		Alkaline	Dark green	Yellowish green
3.	*Plectranthus scutellarioide*	Aqueous	Brown	Pale brown
		Acid	Reddish orange	Pale pink
		Alkaline	Dark green	Green
4.	*Nerium oleander*	Aqueous	Green	Pale green
		Acid	Bright pink	Pale pink
		Alkaline	Dark green	Yellowish brown

Plate 2

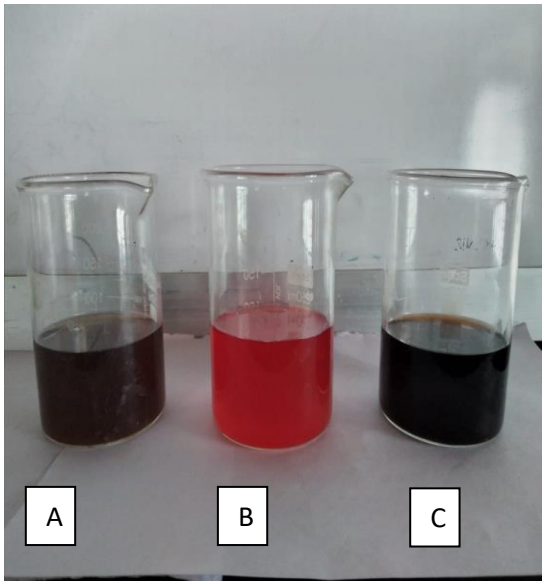
Colour of the dye exacts using different solvents



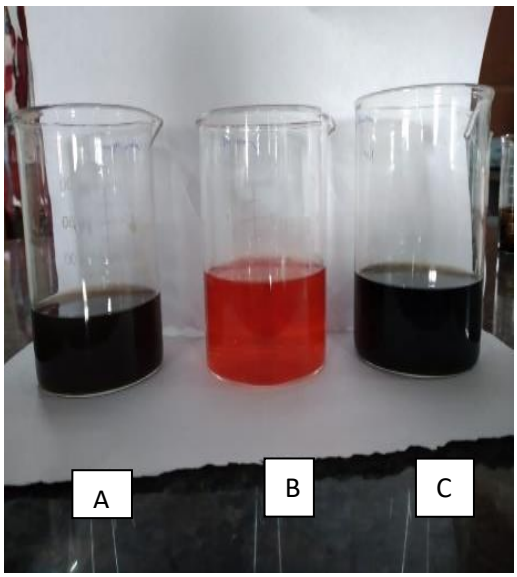
Jatropha gossypifolia



Vitis vinifera



Plectranthus sctellarioides



Nerium oleander

A – Aqueous, B- Acid, C- Alkaline

the mordant material and from alkaline solution the dark green colour was extracted and the mordant material absorbed green colour. Moreover the colour extract from this plant was about brown to pale brown in aqueous, reddish orange to pale pink in acid and dark green to green in alkaline.

In *Nerium oleander,* the colour extracted from aqueous solution is green and the colour was absorbed in mordant material is pale green. From acid solution, bright pink colour dye was extracted and pale pink colour was uptake in the mordant material and from alkaline solution the bright green colour was extracted and the mordant material absorbed yellowish brown. Moreover the colour extract from this plant was about green to pale green in aqueous, bright pink to pale pink in acid and dark green to yellowish brown in alkaline.

Similar results were observed in *Syzygium cumini* seed endosperm dye when extracted with different extracts by Mariselvam *et al*., 2017.

Absorption of the dye in cotton cloth and thread:

The absorption of the different colour dyes on the cotton cloth and thread is shown in Plate 3 – 6. When compared to all the dyes extracted, the cotton thread and cotton cloth did not absorb any colour from the aqueous dye extract of *Jatropha gossypifolia* flower. This may be due to the light colour dye obtained from this flower. The highest absorption is noted in all the acidic and alkaline extracts and especially in cotton threads when compared to the cotton cloth. This may be due to the scourging effect of the cotton cloth.

When natural dyes are extracted using different methods, the most suitable one is using aqueous method. The natural dyes have the capacity to ooze out even when

Plate 3

Dye obtained in cotton cloth and thread

Jatropha gossypifolia

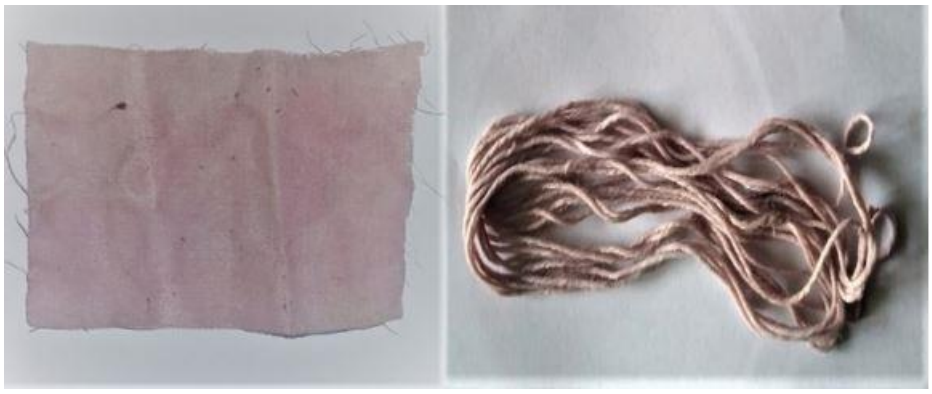
Acid extract of dye

Alkaline extract of dye

Plate 4

Dye obtained in cotton cloth and thread

Vitis vinifera



Aqueous extract of dye

Acid extract of dye

Alkaline extract of dye

Plate 5

Dye obtained in cotton cloth and thread

Plectranthus sctellarioides

Aqueous extract of dye

Acid extract of dye

Alkaline extract of dye

Plate 6

Dye obtained in cotton cloth and thread

Nerium oleander

Aqueous extract of dye

Acid extract of dye

Alkaline extract of dye

kept in normal water (Gokhale *et al*., 2004). Cotton fabrics are needed to be scrouged properly so as to make them lighter to absorb the dye (Siva, 2007).

Determination of absorption percentage:

The absorption percentage of various dye extracts of the selected samples are given in Table 2. From this it is observed that the maximum absorption was seen in the cotton threads when compared to the cotton fabrics. In all the extracts of the samples, the maximum absorption was seen in the alkaline extract followed by acid extract.

This helps to understand the required method of extraction for the dyeing process of specific fibres. Similar results were observed in the textile coloration of cotton using marigold flower by Jothi, 2008. Meena *et al.,* 2013 also observed the similar results.

Determination of solid dye content:

The solid dye content percentage of every extract of the various samples are shown in Table 3. It is interesting to note that the dyes showed the maximum per cent of solid dye content in alkali extraction media and the minimum in aqueous extraction. The highest solid dye percentage was seen in the alkaline extract of *Nerium oleander* flower with 51.2% The least solid dye extraction percentage was seen in the aqueous dye extract of *Jatropha gossyipifolia* flower with 11.2%

This was in agreement with the findings of Dayal and Dobhal (2001) on "Natural dyes from some Indian plants". He has also reported that the highest solid dye content (%) was possible with alkaline media (29.0%) and the lowest amount of solid dye content i.e. 10.6% was reported with water extraction followed by acid (12.5%).

Table 2: Absorption percentage of dye extract on each fabric

S. No.	Plant sample	Solvent	Absorption percentage	
			Cotton cloth	Cotton thread
1.	*Jatropha gossypifolia*	Aqueous	0.8	1.2
		Acid	7.56	12.05
		Alkaline	9.62	18.02
2.	*Vitis vinfera*	Aqueous	10.75	23.25
		Acid	11.25	52.02
		Alkaline	8.75	80.75
3.	*Plectranthus scutellarioide*	Aqueous	9.35	11.26
		Acid	10.58	61.5
		Alkaline	7.32	75.2
4.	*Nerium oleander*	Aqueous	10.26	23.78
		Acid	6.23	51.2
		Alkaline	5.75	50.21

Table 3: Solid Dye content (%) of extracted dye solution

S. No.	Plant sample	Solvent	Solid Dye content (%)
1.	*Jatropha gossypifolia*	Aqueous	11.2
		Acid	26.5
		Alkaline	42.6
2.	*Vitis vinfera*	Aqueous	20.1
		Acid	31.1
		Alkaline	45.3
3.	*Plectranthus scutellarioide*	Aqueous	11.7
		Acid	15.5
		Alkaline	44.3
4.	*Nerium oleander*	Aqueous	21.1
		Acid	36.2
		Alkaline	51.2

Determination of wavelength:

The optical density of the extracted dye was tested under spectrophotometer at 430 to 660 nm. And as a result the peak value was noted and drawn as a graph shown in Figure 1

In *Jatropha gossyipifolia,* by testing wavelength of the dye the peak value was noted in aqueous solution is 490 nm in acid solution 520 nm and in alkaline solution 610nm. The colour was more absorbed in the cotton thread.

In *Vitis vinifera*, by testing wavelength of the dye the peak value was noted in aqueous solution is 500 nm, in acid solution 490 nm and in alkaline solution 600nm. The colour was more absorbed in the cotton thread.

In *Plectranthus scutellarioide*, by testing wavelength of the dye the peak value was noted in aqueous solution is 490 nm, in acid solution 530 nm and in alkaline solution 600nm. The colour was more absorbed in the cotton thread.

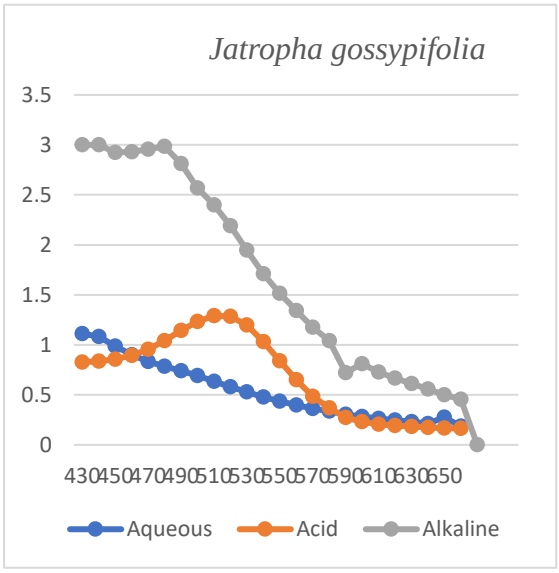
In *Nerium oleoander,* by testing wavelength of the dye the peak value was noted in aqueous solution is 440 nm, in acid solution 560 nm and in alkaline solution 600nm. The colour was more absorbed in the cotton thread.

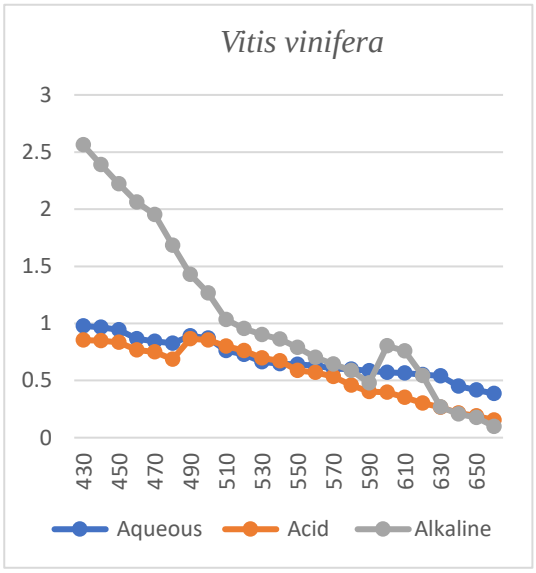
The yellow dye obtained from annatto seeds has also been studied at 350 nm wavelength (Shirwaikar *et al.,* 2011).

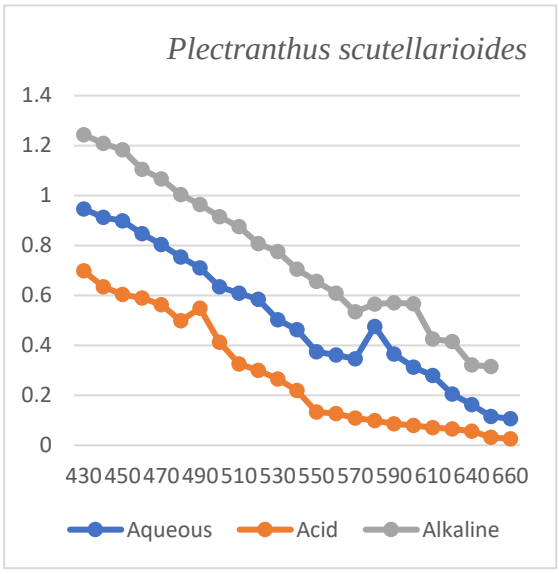
Colour fastness grade value:

The colour fastness grade value of each fabric using different dyes extracted from the plant samples are given in Table 4. The highest fastness grade value was obtained in the aqueous extract of Vitis vinfera fruit with 82.45% which is closely followed by alkaline extracts of Nerium oleander, *Plectranthus scutellarioide*

Figure 1: Wavelength determination of each sample







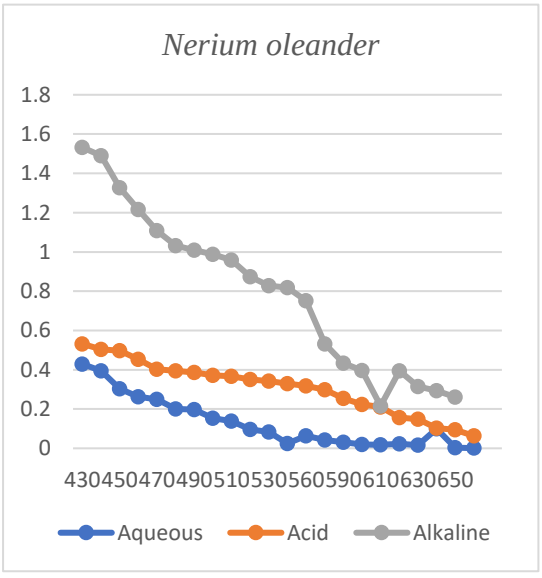


Table 4 : Colour fastness grade value

S. No.	Plant sample	Solvent	Percentage	Grade
1.	*Jatropha gossypifolia*	Aqueous	1.772%	Poor
		Acid	23.68%	Fair
		Alkaline	51.44%	Good
2.	*Vitis vinfera*	Aqueous	82.45%	Very good
		Acid	55.42%	Good
		Alkaline	73%	Very Good
3.	*Plectranthus scutellarioide*	Aqueous	43.36%	Fair
		Acid	12.81%	Poor
		Alkaline	73.32%	Very good
4.	*Nerium oleander*	Aqueous	43%	Fair
		Acid	57.85%	Good
		Alkaline	74.46%	Very good

Table 5: Preliminary Phytochemical screening of the extracted dyes

S.No.	Test	*Jatropha gossypifolia*			*Vitis vinfera*			*Plectranthus scutellarioide*			*Nerium oleander*		
		Acid	Aqueous	Alkaline	Aqueous	Acid	Aqueous	Acid	Acid	Alkaline	Aqueous	Acid	Alkalin
1.	Alkaloids	+	+	+	+	+	+	+	++	+++	++	+	++
2.	Terpenoids	++	+	+++	++	-	++	+++	+++	+	+++	+++	+++
3.	Flavonoids	+	+	+++	-	-	-	++	+	+++	++	+	+++
4.	Tannins	+++	+++	++	-	-	-	+	++	+++	+++	+++	+
5.	Glycosides	-	-	-	-	-	-	-	-	-	-	-	-
6.	Cardiac Glycosides	-	-	-	++	+++	+	-	-	-	-	-	-
7.	Phloabtannins	+	+	+	+	+	+	-	-	-	-	-	-
8.	Steroids	+	+	+	+	+	+	+++	+	++	+	++	+++
9.	Saponins	+	+	+	+	+	+	-	-	-	-	-	-
10.	Phenols	++	++	++	+++	+++	++	++	+	+++	++	++	+++

(+) – indicates presence; (-) indicates absence

and *Vitis vinfera* with 74.46% 73.32% and 73% Similar results were observed by Zhang *et al.,* 2016 in their work with urea-free printing of cotton fabrics.

Preliminary Phytochemical screening of dyes

The results of the preliminary phytochemical screening of the dye samples is shown in Table 5. It is observed that except the glycosides, all the other secondary metabolites are widely present. An overall analysis of phytochemical analysis shows that the secondary metabolites are present in the selected dye samples from four different plants which was used for the studies. It proved that the medicinal properties were present on the dye, when these extracted dyes are used in textile industries, the fabric does not leach out easily and mostly the skin diseases are reduced. Mirunalini *et al* 2010 proved that the presence of phenols and alkaloids are attributed to their medicinal properties.

Summary and Conclusion

SUMMARY AND CONCLUSION

Natural dyes are eco-friendly and their extraction methods are economic and less time consuming. Keeping the above objective in mind the four different samples *Jatropha gossypifolia* flower, *Vitis vinfera* fruit, *Plectranthus scutellarioides* leaves and *Nerium oleander* flower were used for the present study. All the samples used were collected from waste yards. When extracted with aqueous, acid and alkaline soultions they showed the different colours varying from green to yellowish green, Brown to yellowish brown and Pink to purple. The wavelength of the dye material was mostly about 490, 520 and 600 nm to obtain the above colours.

For mordant less amount of chemicals are used. The concentrations was also used in low percentage. This may not cause any severe infection to the skin. In textiles the natural dye fabrics are mostly preferable because they reduces the infection of many skin diseases.

It is interesting to note that the dyes showed the maximum per cent of solid dye content in alkali extraction media and the minimum in aqueous extraction. The highest solid dye percentage was seen in the alkaline extract of *Nerium oleander* flower with 51.2% The least solid dye extraction percentage was seen in the aqueous dye extract of *Jatropha gossyipifolia* flower with 11.2%

The grade was given by doing the colour fastness test, and as a result the grades are varied from Poor to very good. The highest fastness grade value was obtained in the aqueous extract of Vitis vinfera fruit with 82.45% which is closely followed by alkaline extracts of Nerium oleander, *Plectranthus scutellarioides,* and *Vitis vinfera* with 74.46% 73.32% and 73%

When compared to cotton cloth and cotton thread the cotton thread was absorbed more amount of colours. Among these four plant samples *Vitis vinifera* gives the betters results in three different solvents.

The present studies also indicates that the dye material also have the medicinal properties, this was proved by phytochemical analysis. Under this study it showed that the dyes have the secondary metabolites like terpenoids, tannins, steroids, phenols and alkaloids.

Although natural dyes have several advantages there are some limitations as well. Tedious extraction of colouring component from the raw material, low colour value and longer time make the cost of dyeing with natural dyes considerably higher than with synthetic dyes. Some of the natural dyes are fugitive and need a mordant for enhancement of their fastness properties. Some of the metallic mordants are hazardous. Also there are problems like difficulty in the collection of plants, lack of standardization, lack of availability of precise technical knowledge of extracting and dyeing technique and species availability.

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FLORAL DIVERSITY OF ATHIMARAPATTI IN THOOTHUKUDI

A dissertation submitted to

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MASTER OF SCIENCE IN BOTANY

By

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CERTIFICATE

It is certified that this short-term project work entitled **"Floral Diversity of Athimarapatti in Thoothukudi"** submitted to **St. Mary's College (Autonomous)** affiliated to **Manonmaniam Sundaranar University** in partial fulfillment of the requirements for the degree of **Master of Science in Botany** and is a record of work done in the Department of Botany, **St. Mary's College (Autonomous)**, Thoothukudi during the year 2022-2023 by **Lingammal M (21APBO06).**

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I do hereby declare that this dissertation entitled **"Floral Diversity of Athimarapatti in Thoothukudi"** submitted by me in partial fulfillment for the award of the degree of **Master of Science in Botany**, is the result of my original and independent work carried out under the guidance of **Dr. Sr. A. Arockia Jenecius Alphonse,** Assistant Professor of Botany, St. Mary's College (Autonomous), Thoothukudi and it has not been submitted elsewhere for the award of any other degree.

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CONTENT

INTRODUCTION

INTRODUCTION

Biodiversity is referred as the composition of living organisms including plants, animals and microbes inhabiting the terrestrial, aquatic and other habitats of a region or a country. United Nations Environment Programme (UNEP) described biological diversity as the variety and variability of all animals, plants and microorganisms and the ecological complexes of which they are a part. Biodiversity is very indeed to the functioning of various ecosystems. Each species in the world plays a unique role within an ecosystem and every species is dependent on other species for food, shelter and other purposes. Even the loss of a single species can make an impact on ecosystem as well human life.

India is a country rich in a wide variety of Biodiversity. Most of the plants that grow here serve a high medicinal purpose **(Kalisdha *et al.*, 2013).** In India, from the pre-medieval age, holds a possession over natural medicine. Traditional uses of floristic diversity are the foremost vital part of indigenous information system, which is widely practiced by human populations all across the world. This knowledge has been transferred orally from generation to generation.

Floristic studies are nothing but exploring the region by identifying plants and grouping them, data collection of plants present in the region and counting of them. These studies have gifted mankind with the knowledge of plants which are economically important and of high medicinal value. The angiosperms fulfill major needs of human life such as foods, medicines, shelter, cloths and other luxuries **(Kamini Dubey and Sweta Prakash, 2021).** Tamil Nadu is one among the twenty-eight states of India is found with rich floral diversity region. **Irwin *et al.* (2014)**

revealed that there are about 5674 angiospermic species in Tamil Nadu state, which include 212 taxa that are endemic to the state.

Mankind has been utilizing plants for food and medicinal purpose. Therefore, various aspects of plants towards health, economic value, sustainable utility, their conservation, floral assessment and documentation are necessary. India is a rich centre of plants diversity. All types of flora and fauna are elements of biodiversity and influenced by various climatic conditions such as temperature, availability of moisture in the form of humidity and precipitation and variation in physiographical conditions – soil, altitude, slope, etc. **(Suba *et al*., 2014; Sukumaran and Parthiban, 2014).**

The great wealth of biological diversity in tropical regions is due to the myriad environmental conditions existing there. Interest in biodiversity has recently increased in response to the damage caused to ecosystems by anthropogenic activities **(Merigot *et al*., 2007).** It is well known that floristic composition is determined by environmental factors. However, the composition influences biodiversity patterns at regional scales and further reflects both anthropogenic and natural disturbances. Therefore, floristic characteristics and biodiversity patterns are often influenced by environmental factors and anthropogenic disturbances **(Liu *et al*., 2009).**

Conservation of biodiversity is essential for the proper functioning of ecosystems and for the maintenance of the environmental services they provide **(Lopez-del Toro *et al*., 2010).** However, high rates of tropical deforestation and habitat destruction frequently cause the local extinction of plant and animal species. Flowering plants are by far the most numerous, diverse and successful extant plant group containing well over 90%of all land plant species alive today **(Simpson, 2006).**

In India, dicots are represented by 2282 genera and 12750 species whereas, monocots are represented by 702 genera and 4250 species. Dicots accounts for c.75% flowering plants in terms of both genera and species **(Brummit, 1992).**

Documenting basic patterns of biodiversity is fundamental for prioritizing areas for conservation and management action **(Villasenor *et al*., 2007).** Taxonomic inventorying is essential for exploring unexplored genetic resources. Floristic study of smaller areas is more important in comparison with that of larger areas. Smaller areas can be explored thoroughly with critical field observations to find out any additional species which might have been left out from earlier studies. Due to fragmentation of habitats and ecosystems because of various developmental activities many earlier reported taxa might have become extinct, rare and endangered. Study of the existence, size, structure and locality of such taxa deserves more importance.

Floristic explorations and taxonomic studies can provide efficient and convenient information about the nomenclature, distribution, ecology, utility of various plant species, and thus about an ecosystem. Taxonomy is an integrated and perhaps, intuitive science of identifying, naming and classifying plants. This may be considered as the oldest of sciences in the world, as the primitive man had to distinguish the plants that he can eat safely, from those which are poisonous and inedible. Floristic studies help us to understand the basic aspects of biology such as speciation, isolation, endemism and evolution.

A scientific study on medicinal and floristic diversity of the hill group along with traditional knowledge on medicinal plants, which are used by native people in healthcare system, is warranted. Therefore, the importance and scope of the present study is imperative for the conservation and sustainable utilization of plant resources and traditional knowledge for the betterment of future generation without prompting

the use of present generation. The primary goal in the present work is to document the angiosperms associated with Athimarapatti, Thoothukudi District, Tamil Nadu, India that may lead to formulating steps to conservation of natural resources associated with it. The study is also aimed to explore the diversity indices and economic importance of the plants from the study area.

SCOPE AND OBJECTIVE

Natural resources survey like floristic study plays an important role in the economic development of developing country like India. Vegetation is the most precious gift and nature has provided to us as meeting all kinds of essential requirements of the humans in the form of food, fodder, fuel, medicine, timber, resins, and oil, etc. Scientists have been trying to bring to the attention of people and their governments the importance of maintaining biodiversity of planet Earth and of carrying out our daily lives in a fashion that ensures our offspring will inherit a cleaner, greener, more ecologically sustainable world. Floristic studies acquire increasing importance in recent years in response to the need of developing and under developing countries to assess their plant wealth. Keeping all the benefits of floristic studies in mind, the present study is an attempt to realize the target the Mullakadu with the following objectives.

- To make through Floristic survey of flowering plants.
- To provide short description of the dominant families in Dicotyledons (Polypetalae, Gamopetalae, Monochlamydeae) and in monocotyledons
- To provide a list of medicinal plants along with their usage and also to document environmentally, economically useful and ornamental plants.
- To evaluate the present status of the study site and suggest methods for conservation.

REVIEW OF LITERATURE

Vascular plants, an important component of vegetation, must be constantly monitored and managed inorder to direct successional processes towards maintaining species and habitat diversity **(Attua and Pabi 2013; Naidu and Kumar, 2016). Swamy *et al.* (2000)** and **Kothandaraman and Sundarapandian (2017)** explored the flora of low-level evergreen forests of the Western Ghats of Kanyakumari district. **Meena (2007)** listed an exhaustive floristic spectrum of Marundhuvalmalai, a tropical scrub jungle of Western Ghats of Kanyakumari district. Intensive explorations of **Kabeer and Nair (2009)** led to the excellent description of 447 species and 19 intraspecific grass taxa under 136 genera and 19 tribes in Tamilnadu, which form the major collections from Kanyakumari district.

Neelamegam *et al.* (2015) made a baseline survey to evaluate the status, structure, composition, diversity and utilization of home garden plants in two villages one located in a rural area (Arumanalloor) near Nagercoil and the other (Konam) a town municipality of Nagercoil, Kanyakumari district and to understand the impact of socio-economic conditions of households on home garden structure. **Sreekanth *et al.* (2012)** studied the genetic diversity of the teak (*Tectona grandis*) population in Southern Western Ghats. **Karuppusamy and his co-workers (2013a)** enlisted some of the endemic *Caralluma* sp. of Kanyakumari district in their monumentalwork *Caralluma in India*.

Arul *et al.* (2013) reported a new distributional record of *Stylosanthes scabra* from Alamparai hills of Kanyakumari district. While preparing the flora

Plants of the Western Ghats, **Ganeshiah *et al.* (2013)** listed many rare and endemic floristic elements from Western Ghats of Kanyakumari district. Recently, **Karuppusamy (2013b)** and his research team reported the evergreen species *Cleidion nitidum*, so far thought to endemic to Andaman and Nicobar Islands, from Thadagamalai in Kanyakumari district.

Sporadic floristic explorations of Kanyakumari district were made by several researchers recently. **Anderson (2006)** studied the floristic wealth of Adakkadu hills and **Rajeshwari (2009), Anusha (2010), Jani (2011)** and **Vasanthi (2011)** collected the herbaceous plants of Pechiparai, Nagercoil, Marunthuvalmalai and Thovalai, respectively. **Jebaselvi (2010)** surveyed the herbaceous flora of teak and rubber plantations of the district. **Jashbar and Brintha (2011)** botanized the plants of Mothiramalai and Ponmalai hillock, respectively, while the vascular plant diversity of Udayagiri fort of Kanyakumari district was explored by **Sukumaran and Parthiban (2014). Kensa *et al.* (2014)** gave a delightful description of the ornamental flora of an urban environment of Kanyakumari district.

Gajurel *et al.* (2007) studied wild edible plants of Dibang Biosphere Reserve, Arunaehal Pradesh. It has been found that 150 wild plant species used for various purposes of which 80 species are wild edible. **Ghosh and Das (2007)** studied Plants of ethnobotanical significance for the tea Garden Workers in Terai and Duars of Darjeeling. They reported 133 dicotyledonous and 33 monocotyledonous and 4 pteridophytic plants of ethnobotanical significance. Ethnobotanical observations in Pench National Park (Maharashtra) were recorded by **Chaturvedi and Panhi Kumar (2007).**

Sharma *et al.* (2000) reported 694 species which are endemic to Maharashtra. According to annual report of BSI (2001), out of about 17500 species, 5285 species of 140 genera of angiosperm is endemic to India. **Yadav and Sardesai (2002)** compiled 340 endemic plants in the flora of Kolhapur District. **Pawar *et al.* (2006)** studied floristic composition and quantitative assessment of plant resources of Western Ghats from Dhule and Nandurbar District. Quantitative assessment of plant resources from Pamer and Sangamner Tahsils of Ahmednagar District was studied by **Shendage *et al.* (2007).**

Endemic species and their potential value of plant resources are gradually shaping the future of several regions. However, since past hundred years the trends observed in the loss of plant diversity is our biggest concern. Despite all the efforts made to conserve plant diversity, the situation today is still very alarming. **Takhtajan (1986)** stated that India to be cradle for the origin of cultivated plants. On similar lines, **Vavilov (1992) and recently Khoury *et al*. (2016)** stated that India to be prime pocket for the origin of pulses. The rich diversity of edible plants has been valued and consumed consciously by the Indian villagers. According to **Nayar (1996),** 5725 species under 147 genera of angiosperms i.e., 33% are endemic. Documentation on wild flora of Indian region carried out by **Singh and Arora (1978)** showed that around 250 species occur in the western and nearly 300 species occur in the eastern Himalayan ranges. This diversity includes 258species of edible fruits, 121 species of green leafy vegetables, 37 species of roots and tubers, and 20 species of edible flower buds. Amongst the 214 threatened species occurring in the Himalayas, nearly 37 are exploited, being medicinal herbs of commercial value and need priority action for conservation **(Arora and Nayar, 1984).**

Urbanization is one of the major reasons for the destruction of the natural vegetation. This ongoing growth of urban agglomerations leads to far-reaching changes in biodiversity, including the loss of forests and other natural areas **(Kumar *et al*., 2010; Von der Lippe and Kowarik, 2008).** Urbanized areas can also harbour a high number of threatened species **(Sodhi *et al*., 2010).** The destruction of tropical forests and habitats causes global biodiversity degradation **(Singh, 1998).**

Floristic diversity study helps us to evaluate the floristic wealth and its prospects of an area. Floristic inventories help us perceive biological aspects such as endemism, evolution, speciation and isolation **(Elourard *et al*., 1997).** It helps to assess the country's plant wealth, distribution and status **(Ellis, 1987).** There are 15000 species of flowering plants in India belonging to 2250 genera and 315 families, contributing 6% of World's flowering plants **(Nayar, 1977).** The present number of flowering plants in India is found to be 18666 species under 2991 genera and 251 families **(Mao, 2019).**

The total number of angiosperms reported from the Dibrugarh District presents 462 species belonging to 334 genera and 106 families show a similar agreement with other floristic studies previously carried out in different regions of India. An inventory of the native flowering plants in the East Siang district of Arunachal Pradesh presented 508 taxa belonging to 348 genera and 102 families **(Taram *et al.*, 2020).** Higher plant diversity in East Kameng district of Arunachal Prdesh reported 215 species of higher plants belonging to 165 genera and 70 families **(Tag *et al.*, 2012).** Floristic diversity assessment and vegetation analysis of Upper Siang district of Northeast India reported 1003 taxa belonging to 110 families and 529 genera **(Choudhary *et al.*, 2012).** A study of Angiospermic diversity in the Bhadrak

region of Odissa showed 383 species including 262 native and 121 non-native species belonging to 282 genera under 93 families **(Panda *et al.*, 2020).**

Biodiversity reflects variety and variability within and among living organisms, their associations and habitat-oriented ecological complexes. India is one of the 12 "megadiversity" countries in the world. Mankind has been utilizing plants for food and medicinal purpose. Therefore, various aspects of plants towards health, economic value, sustainable utility, their conservation, floral assessment and documentation are necessary. India is a rich centre of plants diversity. All types of flora and fauna are elements of biodiversity and influenced by various climatic conditions such as temperature, availability of moisture in the form of humidity and precipitation, and variation in physiographical conditions – soil, altitude, slope, etc. **(Arul *et al.*,2013; Ghildiyal and Juyal, 2012; Suba *et al.*, 2014; Sukumaran and Parthiban, 2014).**

The great wealth of biological diversity in tropical regions is due to the myriad environmental conditions existing there. Interest in biodiversity has recently increased in response to the damage caused to ecosystems by anthropogenic activities **(Merigot *et al.* 2007).** It is well known that floristic composition is determined by environmental factors; however, the composition influences biodiversity patterns at regional scales and further reflects both anthropogenic and natural disturbances. Therefore, floristic characteristics and biodiversity patterns are often influenced by environmental factors and anthropogenic disturbances **(Liu *et al.*, 2009).**

Conservation of biodiversity is essential for the proper functioning of ecosystems and for the maintenance of the environmental services they provide **(Lopez-del Toro *et al.*, 2010).** However, high rates of tropical deforestation and habitat destruction frequently cause the local extinction of plant and animal species.

Flowering plants are by far the most numerous, diverse and successful extant plant group containing well over 90%of all land plant species alive today **(Simpson, 2006)**. In India dicots are represented by 2282 genera and 12750 species whereas monocots are represented by 702 genera and 4250 species. Dicots accounts for c.75% flowering plants in terms of both genera and species **(Brummit, 1992).**

Natural resources survey like floristic study plays an important role in the economic development of developing country like India. Vegetation is the most precious gift, nature has provided to us as meeting all kinds of essential requirements of the humans in the form of food, fodder, fuel, medicine, timber, resins, and oil etc. **(Gaur, 1999).** Plant communities play a pivotal role in sustainable management by maintaining biodiversity and conserving the environment **(Farooquee and Saxena, 1996).** Floristic diversity refers to the variety and variability of plants in a given region. It refers to the number of types or taxa in a given region or group. It can be measured at any level from overall global diversity to ecosystem, community, species, populations, individuals and even to genes within a single individual.

Floristic study and diversity assessments are necessary to understand the present diversity status and conservation of biodiversity. Floristic study is a necessary prerequisite for much fundamental research in tropical community ecology, such as modeling patterns of species diversity or understanding species distributions **(Phillips *et al.*, 2003).** Floristic studies acquire increasing importance in recent years in response to the need of developing and under developing countries to assess their plant wealth **(Vediya and Kharadi, 2011).** Floristic explorations and taxonomic studies can provide efficient and convenient information about the nomenclature, distribution, ecology, utility of various plant species, and thus about an ecosystem.

Taxonomy is an integrated and perhaps, intuitive science of identifying, naming and classifying plants. This may be considered as the oldest of sciences in the world, as the primitive man had to distinguish the plants that he can eat safely, from those which are poisonous and inedible. The gravity of the situation is so severe due to variety of reasons, the foremost being habitat destruction at an alarming rate leading to loss of biodiversity essential for the sustenance of life on earth. Thus, conservation of biodiversity has gained prime consideration all over the world since the Earth, Summit at Rio de Janeiro in 1992. Floristic studies help us to understand the basic aspects of biology such as speciation, isolation, endemism and evolution. Flora of any area is not fixed up. It changes from time to time. Various ecological factors, mostly biotic, change the floristic components. The total number of species may be changed; dominant species may be replaced with other species; the floristic composition, i.e.; family: genus: species ratio may be changed.

Documentation of existing green spaces of an area is important to determine existing resources and to set target for future improvements. Floras serve as the basic reference of the plant biota of an area; they are critical tools that serve botanists, conservationists, ecologists, foresters, gardeners, agronomists, researchers, and the general public. The botanical exploration of an area and writing a flora to summarize that information was seen as a basic societal need leading to the discovery of economically valuable information. Among the plants inhabiting the earth, the angiosperms or flowering plants are one of the major groups of extant seed plants and arguably the most diverse major extant plant group on the planet with at least 2,60,000 living species classified in 453 families **(Judd *et al.*, 2002).**

In India, the angiosperms comprising a total of 17,817 species constitutes 38.15% of floral diversity of the entire country. According to current estimates, the Indian flora represents nearly 12% of the global floral diversity (excluding viruses). The significance of the Indian flora is further evidenced by the number of species of wild relatives of crop plants in different regions of the country. Since time immemorial man has been using plant resources to meet his daily requirements. The immense diversity which occur in flowering plant provides economic benefits as they are important sources of food, fodder, timber, medicine, fibres, spices, dyes and tannins, beverages, gums and resins etc. **(Singh *et al.*, 2002).**

MATERIALS AND METHODS

PLANT COLLECTION AND IDENTIFICATION

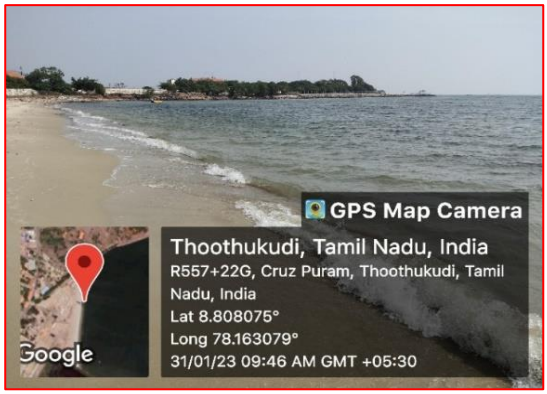
From the month of December, 2022 to March 2023, Athimarapatti village was regularly visited and every plant was photographed and collected in plastic bags for identification. With the aid of Flora of the Presidency of Madras, the plants were identified. In addition to data on taxonomic place (family), vernacular name, common name, life form, and folk medicinal uses, the species entries were supplemented. The life type was graded according to the system suggested by Raunkiær into herbs, shrubs, grasses and trees (annual, biennial or perennial).

ETHNOBOTANICAL SURVEY

The ethnobotanical study of the village includes the testing of knowledge about the usage of common medicinal plants. Based on the literature review, about 14 categories of common ailments like Kidney Disease (KD, Skin disease (SD), Hair problem (HP), Stomach problem (SP), Respiratory disease (RD), Cold (C), Cough (CO), Diuretic (D), Tooth ache (TA), Head ache (HA), Fever (F), Body heat (BH), Insect Bite (IB) and Menstural issues (MI) were selected for the present survey. A questionnaire was prepared using Google forms to collect the ethnobotanical applications of the plants in the village. The link is https://forms.gle/CrHEk1xTXWTosWBH7. The link was then circulated among the residents of the chosen village. Choices were given to each respondent to select the medicinal plants and their uses. There were up to 124 members participated in the survey.

PLATE 1: SAMPLING STATIONS OF THOOTHUKUDI COAST

STATION I: VEMBAR



STATION II: THERESPURAM

STATION III: PUNNAKAYAL

QUANTITATIVE ANALYSIS: (Umair *et al.*, 2017)

The ethnobotanical data was analyzed using different quantitative indices including Informant Consensus Factor (ICF), Use value (UV), Relative frequency citation (RFC), Fidelity level (FL), Relative popularity level (RPL), Rank order priority (ROP). Data were reported in proportions and percentages.

INFORMANT CONSENSUS FACTOR (ICF).

ICF value describes informants' consensus on the medicinal plant consumption species, and evaluates variability in mode of utilization against reported diseases. Before calculating ICF value, ailments are broadly categorized into different categories. The maximum ICF value i.e. close to 1 indicates that well known species are used by a large proportion of local communities due to their authenticity regarding diseases. However, low ICF index close to 0 specifies that the informants use this species randomly to treat reported diseases. The ICF value was calculated using the formula as described earlier.

$$ICF = \frac{N_{ur} - N_t}{N_{ur} - 1}$$

Where,

"Nur " is the total number of use reports for each disease category

"Nt" indicates the number of species used in said category.

USE VALUE (UV)

Use value (UV) determines the relative importance on uses of plant species. It is calculated using the following formula as follows

$$UV = \frac{\sum U_i}{N}$$

Where,

"UV" indicates use value of individual species,

"U" is the number of uses recoded for that species and

"N" represents the number of informants who reported that species.

RELATIVE FREQUENCY OF CITATION (RFC)

Relative frequency of citation (RFC) signifies the local importance of each species in a study area. This index is determined by dividing the number of informants citing a useful species (FC) by total number of informants in the survey (N). RFC is calculated by the formula,

$$RFC = \frac{FC}{N} \quad (0 < RFC < 1)$$

FIDELITY LEVEL (FL)

FL is the percentage of informants who mentioned the uses of certain plant species to treat a particular ailment in a study area. The FL index is calculated using formula,

$$FL\ \% = \frac{NP}{N} \times 100$$

Where,

'Np' is the number of informants that claimed a use of certain plant species for a particular disease

'N' is the total number of informants citing the species for any disease.

The maximum FL indicates the frequency and high use of the plant species for treating a particular ailment by the informants of the study area.

RELATIVE POPULARITY LEVEL (RPL)

RPL is the ratio between number of ailments treated by a particular plant species and the total number of informants for any disease. However, plant species with comparable FL may vary in their healing potential. A correction scale is therefore introduced, in which all the encountered plant species are divided into popular and unpopular groups. The relative popularity level (RPL) assumes a value 0 and 1.0, with '1' being complete popularity of a plant for major ailments and '0' no ailments treated by a plant species. When all plant species are frequently used to treat some major ailments, popularity index would be maximum (1.0); then decrease towards zero as the relative popularity of the species diverge away from popular side. For popular plant species, the RPL value is rationally selected to equal unity (i.e. equal to 1), while RPL value is less than 1 for unpopular plant species. The relative popularity level (RPL) of the plant species is calculated and designated as popular or unpopular. The RPL value may be determined for each specific plant in accordance with its exact position on graph.

RANK ORDER PRIORITY (ROP)

ROP is a correction factor, used for appropriate ranking of the plant species with different FL and RPL values. The ROP is derived from FL; by multiplying RPL and FL values as follows.

ROP = FL x RPL

RESULTS AND DISCUSSION

FLORISTIC ANALYSIS

Flowering plants are the most numerous, diverse and successful extant plant group containing well over 90% of all land plant species alive today **(Simpson, 2006).** In India, dicots are represented by 2282 genera and 12750 species whereas monocots are represented by 702 genera and 4250 species. Dicots accounts for c.75% flowering plants in terms of both genera and species **(Brummit, 1992).** Species richness is measured on samples carefully chosen in a particular area. Such data are important for prioritizing conservation strategies since they allow identification of geographic regions of the world wide exceptional or with very poor diversity **(Krishnamoorthy 2003).**

Floristic studies help us to understand the basic aspects of biology such as speciation, isolation, endemism and evolution. Flora of any area is not fixed up. It changes from time to time. Various ecological factors, mostly biotic, change the floristic components. The total number of species may be changed; dominant species may be replaced with other species; the floristic composition, i.e. family: genus: species ratio may be changed. The present study revealed that the study area is housing more taxa, however, identified only 90 taxa **(Table 1).** These 90 species were found to be dispersed in 32 families in Dicot and 2 families in Monocot, 66 genera in Dicot and 4 genera in Monocot, 86 species in Dicot and 4 species in Monocot **(Fig 1).**

GENERIC AND SPECIES DIVERSITY

Figure 3 and Table 2 show generic and species diversity of 34 families studied. Out of 34 families, Cucurbitaceae and Euphorbiaceae have more generic

diversity compared to other families. 5 genera in Euphorbiaceae, 7 genera in Cucurbitaceae, 4 genera in 3 families such as Asteraceae, Malvaceae and Solanaceae, 3 genera in 7 families, 2 genera in 2 Families and 20 families are monogeneric. Maximum species diversity occurs in the family Solanaceae (9 species), Cucurbitaceae (9 species), Euphorbiaceae (7 species), Malvaceae (6 species) and Convolvulaceae (5 species).

DIVERSITY OF HABIT FORMS

Different life forms of species are shown in **Table 3**. In general, the vegetation is dominated by herbs (56%) followed by Shrubs (25%), Climbers (18%) and Tree (1%). The percentage distribution of habit forms in different class is shown in **Fig.2.**

USE REPORT AND USE CATEGORIES:

Based on the literature review, about 6 categories of common ailments like Skin disease (SD), Hair problem (HP), Cold (C), Head ache (HA), Fever (F), and Menstrual issues (MI) were selected for the present survey and about 23 plant species were given in the survey to note down their usage. Among which, about 11 species were surveyed to cure skin diseases which is followed by head ache and menstrual issues reported by 10 as shown in **Table 4**. Many researchers previously documented the ethnobotanical surveys also stated that, the skin disorder reports higher number of plants **(Khajoei and Khosravi, 2014).** Due to poor dietary conditions and unsafe drinking water, this ailment category is one of the most common problems in the study area and in fact in other parts of the world. Dermatological infections such as scabies, small pox, lesion odour and burns are other prevalent ailments in the study area which are treated with medicinal plants by Malayali ethnic people. Similarly, **Morvin *et al.*, (2014)** reported that medicinal plants for the treatment of dermatological infections had a high prevalence in Kerala, India.

TABLE 1: CHECK LIST OF ANGIOSPERM DIVERSITY IN THE STUDY AREA

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
1.	*Ricinus communis* L.	Castor bean	Euphorbiaceae
2.	*Jatropha gossypiifolia* L.	Bellyache bush	Euphorbiaceae
3.	*Chrozophora brocchiana* (Vis.) Schweinf.	Poison bush	Euphorbiaceae
4.	*Euphorbia peplus* L.	Petty spurge, milk weed	Euphorbiaceae
5.	*Euphorbia serpens* Kunth.	Matted sandmat	Euphorbiaceae
6.	*Croton bonplandianus* Baill.	Aathuppoondu	Euphorbiaceae
7.	*Euphorbia prostrate* Aiton.	Prostrate sandmat	Euphorbiaceae
8.	*Stachytarpheta jamaicens* (L.)	Blue porterweed	Verbenaceae
9.	*Lantana camara* L.	Arch man	Verbenaceae
10.	*Lantana aculeate* L.	Largeleaf lantana	Verbenaceae
11.	*Phyla nodiflora* (L.) Greene	Turkey tangle, Frog fruit	Verbenaceae
12.	*Acacia tortilis* (Forssk).	Umbrella thorn	Leguminosae
13.	*Senna occidentalis* (L.) Link	Antbush, Coffee senna	Leguminosae
14.	*Galactia spp.*	Milk peas	Leguminosae
15.	*Cassia angustifolia* Vahl.	Senna, Indian senna	Caesalpiniaceae
16.	*Pergularia daemia* (Forsskal) Chiov.	Veliparutthi	Asclepiadaceae
17.	*Oxystelam esculentum* R. Br.	Rosy milkweed vine	Asclepiadaceae

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
18.	*Calotropis procera* (Aiton) Dryand.	Apple of sodom	Asclepiadaceae
19.	*Calotropis gigantea* (L.) Dryand.	Bleaching powder	Asclepiadaceae
20.	*Ziziphus spina-christi* (L.) Desf.	Christ's thorn	Rhamnaceae
21.	*Tridax procumbens* (L.)	Coatbuttons, kinarruppacan	Compositae
22.	*Parthenium hysterophorus* L.	Santa Maria feverfew	Asteraceae
23	*Stoebe plumose* L.	Silver stoebe	Asteraceae
24.	*Sida spinosa* L.	Arrow leaf sida	Malvaceae
25	*Sida cordifolia* L.	Heart leaf sida	Malvaceae
26	*Abelmoschus ficulneus* (L.)	White wild musk mallow	Malvaceae
27	*Hibiscus calyphyllus* Cav.	Hibiscus	Malvaceae
28	*Abutilon indicum* (L) Sweet.	Monkey Bush	Malvaceae
29	*Tribulus terrestris* L.	Puncture Vine	Zygophyllaceae
30	*Luffa cylindrica (*L.) M. Roem	Galka, Bath sponge gourd	Cucurbitaceae
31	*Citrullus lanatus* (Thunb.) Matsum & Nakai	Water melon	Cucurbitaceae
32	*Momordica cymbalaria* (Hook & Fenzl)	Little wild gourd	Cucurbitaceae
33	*Cucumis melo* L.	Muskmelon, sweet melon	Cucurbitaceae
34	*Momordica charantia* L.	Balsam pear; Bitter gourd	Cucurbitaceae

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
35	*Citrullus colocynthis* (L.) Schrad	Vine of sodom	Cucurbitaceae
36	*Mukia maderaspatana* W.	Madras pea pumpkin.	Cucurbitaceae
37	*Lagenaria siceraria* (Molina) Standley.	Bottle gourd,	Cucurbitaceae
38	*Cucurbita maxima* Duch.	Winter squash	Cucurbitaceae
39	*Achyranthes aspera* L.	Prickly chaff flower	Amaranthaceae
40	*Celosia cristata* L.	Cockscomb	Amaranthaceae
41	*Cardiospermum halicacabum* L.	Balloon vine	Sapindaceae
42	*Physalis peruviana* L.	Cape gooseberry	Solanaceae
43	*Physalis angulata* L.	Wild tomato, Camapu	Solanaceae
44	*Solanum pimpinellifolium* L.	Currant tomato	Solanaceae
45	*Solanum linnaeanum* Happer	poison bush, poison weed	Solanaceae
46	*Solanum trilobatum*, L.	Alarkapatramu, Thoothuvalai	Solanaceae
47	*Capsicum annuum* L.	Capsicum, chili.	Solanaceae
48	*Datura matel* L.	Devil's trumpet	Solanaceae
49	*Solanum incanum* L.	Thorn apple, bitter apple	Solanaceae
50	*Solanum melongena* L.	Brinjal, eggplant	Solanaceae
51	*Cyanthillium cinereum (L.) H. Rob.*	West Indian holly, sage rose	Asteraceae

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
52	*Passiflora foediax* L.	Running pop	Passifloraceae
53	*Passiflora edulis Sims*	Passion fruit	Passifloraceae
54	*Leucas aspera* (Willd.) Link	Thambai, Kubo	Lamiaceae
55	*Ocimum tenuiflorum*	Holy basil, Sacred basil, Tulsi	Lamiaceae
56	*Mesosphaerum suaveolens*	Pignut, wild spikenard	Lamiaceae
57	*Ocimum kilimandscharium* Baker ex Gurke	Hoary basil, camphor basil.	Lamiaceae
58	*Merremia disseca* (Jacq.) Hallier f.	Alamo vine	Convolvulaceae
59	*Merremia aegyptia* (L.) Urb.	Hairy morning glory	Convolvulaceae
60	*lpomoea aquatic* Forsk.	Water spinach	Convolvulaceae
61	*Ipomoea quamoclit* L.	Cypress vine	Convolvulaceae
62	*Ipomoea* carnea Jacq.	Pink morning glory	Convolvulaceae
63	*Barleria volkensii* Lindau	Philippine violet	Acanthaceae
64	*Hygrophila auriculata* (K. Schum.)	Marsh barbel	Acanthaceae
65	*Dicliptera paniculata* (Forssk.) I. Darbush	Panicled foldwing,	Acanthaceae
66	*Pedalium mure* L.	Crow thorn	Pedaliaceae
67	*Ludwigia octovalvis* Linn.	Mexican primrose willow	Onagraceae

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
68	*Ludwigia palustris* (L.) Elliott.	Marsh primrose willow	Onagraceae
69	*Heliotropium indicum* L.	Indian heliotrope	Boraginaceae
70	*Stemodia durantifolia* (Linn.)	Whitewoolly twintip	Scrophulariaceae
71	*Schoenoplectus tabernaemontani* (C.C. Gmel.)	Grey club-rush, great bulrush	Cyperaceae
72	*Corchorus trilocularis*, L.	Wild jute	Tiliaceae
73	*Cleoserrata speciosa* (Raf.) Iltis	Showy spider flower	Cleomaceae
74	*Cleome viscosa* L.	Asian spider flower	Capparidaceae
75	*Cleome gynandra* L.	Shona cabbage	Capparidaceae
76	*Commelina communis* L.	Day flower	Commelinaceae
77	*Argemone mexicana* L.	Mexican poppy	Papaveraceae
78	*Impatiens balsamina* L.	Balsam, rose balsam	Geraniaceae
79	*Phyllanthus polygonoides* Nutt.	Smartweed leaf-flower	Euphorbiaceae
80	*Turnera ulmifolia* L	West Indian holly, sage rose	Turneraceae
81	*Hydrocotyle spp.*	Marsh penny, dollar weed	Araliaceae
82	*Jasminum sambac* (L.) Aiton	Arabian jasmine	Oleaceae

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
83	*Ammannia coccinea* Rottb.	Blistering ammannia	Lythraceae
84	*Scleromitrion diffusum* (Willd.) R. J. Wang	Snake needle grass	Rubiaceae.
85	*Oldenlandia umbellate* L.	Chayaver	Rubiaceae
86	*Citrus aurantiifolia* (Christm.) Swingle	Key lime, lime	Rubiaceae
87	*Boehmeria cylindrica* (L.)	False nettle, bog hemp	Urticaceae
88	*Digitaria sanguinalis* (L.)	Hairy crabgrass, finger-grass	Poaceae
89	*Orysa sativa* L.	Rice	Poaceae
90	*Brachiaria brizantha* (Hochst. ex A. Rich.)	Annual beard grass	Poaceae

TABLE 2: OCCURRENCE OF GENERA AND SPECIES WITHIN THE RESPECTIVE PLANT FAMILIES

S.NO.	FAMILIES	NO. OF GENERA	NO. OF SPECIES
01.	Euphorbiaceae	5	7
02.	Verbenaceae	3	4
03.	Leguminosae	3	3
04.	Caesalpiniaceae	1	1
04.	Asclepiadaceae	3	4
05.	Rhamnaceae	1	1
06.	Asteraceae	4	4
07.	Malvaceaae	4	6
08.	Zygophyllaceae	1	1
09.	Cucurbitaceae	7	9
10.	Amaranthaceae	2	2
11.	Sapindaceae	1	1
12.	Solanaceae	4	9
13.	Turneraceae	1	1
14.	Passifloraceae	1	2
15.	Lamiaceae	3	4
16.	Convolvulaceae	2	5

S.NO.	FAMILIES	NO. OF GENERA	NO. OF SPECIES
17	Acanthaceae	3	3
18.	Pedaliaceae	1	1
19.	Onagraceae	1	2
20.	Boraginaceae	1	1
21.	Scrophulariaceae	1	1
22.	Cyperaceae	1	1
23.	Cleomaceae	1	1
24	Capparidaceae	1	2
25.	Commelinaceae	1	1
26.	Papaveraceae	1	1
27.	Geraniaceae	1	1
28.	Turneraceae	1	1
29.	Araliaceae	1	1
30.	Oleaceae	1	1
31.	Lythraceae	1	1
32.	Rubiaceae	3	3
33.	Urticaceae	1	1
34.	Poaceae	3	3

FIGURE 1: DISTRIBUTION OF TAXA IN DIFFERENT CLASSES OF ANGIOSPERM

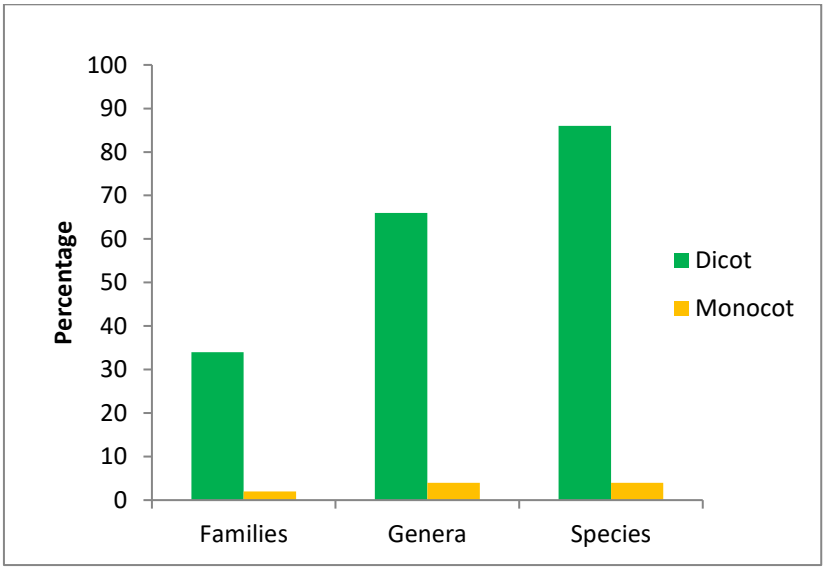


FIGURE 2: PERCENTAGE OF SPECIES OCCURRENCE WITHIN THE IDENTIFIED PLANT HABITS

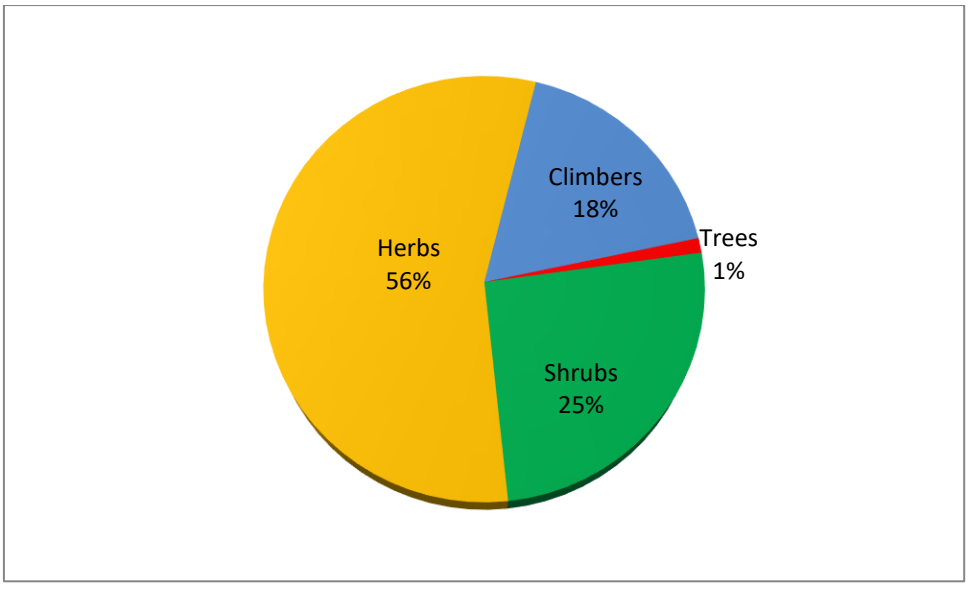


FIGURE 3: GENERIC AND SPECIES DIVERSITY IN THE SELECTED FAMILIES

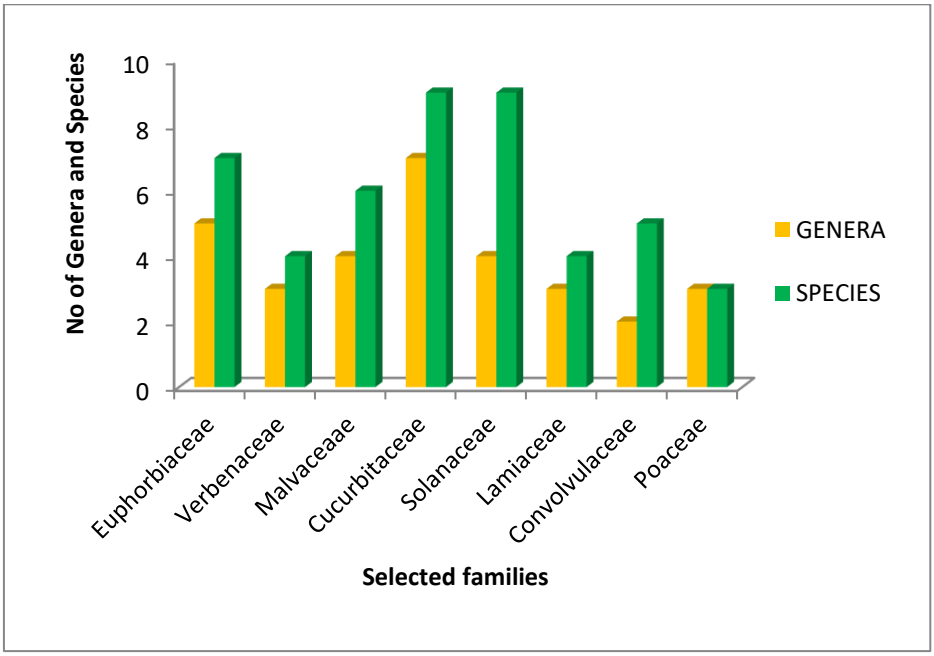


TABLE 3: DIFFERENT LIFE FORMS OF SPECIES IN THE STUDY AREA

S. No.	Botanical Name	Family	Habit
1.	*Ricinus communis* L.	Euphorbiaceae	Shrub
2.	*Jatropha gossypiifolia* L.	Euphorbiaceae	Shrub
3.	*Chrozophora brocchiana* (Vis.) Schweinf.	Euphorbiaceae	Herb
4.	*Euphorbia peplus* L.	Euphorbiaceae	Herb
5.	*Euphorbia serpens* Kunth.	Euphorbiaceae	Herb
6.	*Croton bonplandianus* Baill.	Euphorbiaceae	Herb
7.	*Euphorbia prostrate* Aiton.	Euphorbiaceae	Herb
8.	*Stachytarpheta jamaicens* (L.)	Verbenaceae	Herb
9.	*Lantana camara* L.	Verbenaceae	Shrub
10.	*Lantana aculeate* L.	Verbenaceae	Shrub
11.	*Phyla nodiflora* (L.) Greene	Verbenaceae	Herb
12.	*Acacia tortilis* (Forssk).	Leguminosae	Tree
13.	*Senna occidentalis* (L.) Link	Leguminosae	Shrub
14.	*Galactia spp.*	Leguminosae	Climber
15.	*Cassia angustifolia* Vahl.	Caesalpiniaceae	Shrub
16.	*Pergularia daemia* (Forsskal) Chiov.	Asclepiadaceae	Herb
17.	*Oxystelam esculentum* R. Br.	Asclepiadaceae	Climber
18.	*Calotropis procera* (Aiton) Dryand.	Asclepiadaceae	Shrub

S. No.	Botanical Name	Family	Habit
19.	*Calotropis gigantea* (L.) Dryand.	Asclepiadaceae	Shrub
20	*Ziziphus spina-christi* (L.) Desf.	Rhamnaceae	Shrub
21	*Tridax procumbens* (L.)	Compositae	Herb
22	*Parthenium hysterophorus* L.,	Asteraceae	Herb
23	*Stoebe plumose* L.	Asteraceae	Herb
24	*Sida spinosa* L.	Malvaceae	Herb
25	*Sida cordifolia*, L.	Malvaceae	Shrub
26	*Abelmoschus ficulneus* (L.)	Malvaceae	Shrub
27	*Hibiscus calyphyllus* Cav.	Malvaceae	Shrub
28	*Abutilon indicum*, (L) Sweet.	Malvaceae	Shrub
29	*Tribulus terrestris*, L.	Zygophyllaceae	Herb
30	*Luffa cylindrica (*L.) M. Roem	Cucurbitaceae	Climber
31	*Citrullus lanatus* (Thunb.) Matsum & Nakai	Cucurbitaceae	Climber
32	*Momordica cymbalaria* (Hook & Fenzl)	Cucurbitaceae	Climber
33	*Cucumis melo* L.	Cucurbitaceae	Climber
34	*Momordica charantia* L.	Cucurbitaceae	Climber
35	*Citrullus colocynthis* (L.) Schrad	Cucurbitaceae	Herb
36	*Mukia maderaspatana* W.	Cucurbitaceae	Climber

S. No.	Botanical Name	Family	Habit
37	*Lagenaria siceraria* (Molina) Standley.	Cucurbitaceae	Climber
38	*Cucurbita maxima* Duch.	Cucurbitaceae	Climber
39	*Achyranthes aspera* L.	Amaranthaceae	Herb
40	*Celosia cristata* L.	Amaranthaceae	Herb
41	*Cardiospermum halicacabum* L.	Sapindaceae	Climber
42	*Physalis peruviana* L.	Solanaceae	Herb
43	*Physalis angulata* L.	Solanaceae	Herb
44	*Solanum pimpinellifolium* L.	Solanaceae	Herb
45	*Solanum linnaeanum* Happer	Solanaceae	Shrub
46	*Solanum trilobatum*, L.	Solanaceae	Climber
47	*Capsicum annuum* L.	Solanaceae	Herb
48	*Datura matel* L.	Solanaceae	Shrub
49	*Solanum incanum* L.	Solanaceae	Shrub
50	*Solanum melongena* L.	Solanaceae	Shrub
51	*Cyanthillium cinereum (L.) H. Rob*	Asteraceae	Herb
52	*Passiflora foediax* L.	Passifloraceae	Climber
53	*Passiflora edulis Sims*	Passifloraceae	Climber
54	*Leucas aspera* (Willd.) Link	Lamiaceae	Herb

S. No.	Botanical Name	Family	Habit
55	*Ocimum tenuiflorum*	Lamiaceae	Herb
56	*Mesosphaerum suaveolens*	Lamiaceae	Shrub
57	*Ocimum kilimandscharium* Baker ex Gurke	Lamiaceae	Shrub
58	*Merremia disseca* (Jacq.) Hallier f.	Convolvulaceae	Climber
59	*Merremia aegyptia* (L.) Urb.	Convolvulaceae	Climber
60	*lpomoea aquatic*, Forsk.	Convolvulaceae	Herb
61	*Ipomoea quamoclit* L.	Convolvulaceae	Herb
62	*Ipomoea* carnea Jacq.	Convolvulaceae	Shrub
63	*Barleria volkensii* Lindau	Acanthaceae	Herb
64	*Hygrophila auriculata* (K. Schum.)	Acanthaceae	Herb
65	*Dicliptera paniculata* (Forssk.) I. Darbush	Acanthaceae	Herb
66	*Pedalium mure* L.	Pedaliaceae	Herb
67	*Ludwigia octovalvis* Linn.	Onagraceae	Herb
68	*Ludwigia palustris* (L.) Elliott.	Onagraceae	Herb
69	*Heliotropium indicum* L.	Boraginaceae	Herb
70	*Stemodia durantifolia* (Linn.)	Scrophulariaceae	Shrub
71	*Schoenoplectus tabernaemontani* (C.C. Gmel.)	Cyperaceae	Herb
72	*Corchorus trilocularis*, L.	Tiliaceae	Herb

S. No.	Botanical Name	Family	Habit
73	*Cleoserrata speciosa* (Raf.) Iltis	Cleomaceae	Herb
74	*Cleome viscosa* L.	Capparidaceae	Herb
75	*Cleome gynandra* L.	Capparidaceae	Herb
76	*Commelina communis* L.	Commelinaceae	Herb
77	*Argemone mexicana* L.	Papaveraceae	Herb
78	*Impatiens balsamina* L.	Geraniaceae	Herb
79	*Phyllanthus polygonoides* Nutt.	Euphorbiaceae	Herb
80	*Turnera ulmifolia* L	Turneraceae	Herb
81	*Hydrocotyle spp.*	Araliaceae	Herb
82	*Jasminum sambac* (L.) Aiton	Oleaceae	Shrub
83	*Ammannia coccinea* Rottb.	Lythraceae	Herb
84	*Scleromitrion diffusum* (Willd.) R.J.Wang	Rubiaceae.	Herb
85	*Oldenlandia umbellate* L.	Rubiaceae	Herb
86	*Citrus aurantiifolia* (Christm.) Swingle	Rubiaceae	Shrub
87	*Boehmeria cylindrica* (L.)	Urticaceae	Herb
88	*Brachiaria brizantha* (Hochst. ex A. Rich.)	Poaceae	Herb
89	*Digitaria sanguinalis* (L.)	Poaceae	Herb
90	*Orysa sativa* L.	Poaceae	Herb

INFORMANT CONSENSUS FACTOR (ICF):

To calculate ICF, the reported ailments were first classified into 6 different disease categories on the basis of their use reports. Among 6 major disease categories, dermatological disorders were dominated with fever with 176 user reports, followed by skin disease and cold with 159 use-reports as mentioned in **Table 4.** Consensus analysis has been used as an important tool for analysing of ethnobotanical data, and it also tells the level of prevalence of diseases in the study area. From the finding it is shown that cold is noted with the highest ICF value of 0.98 which may be due to the water used in the area. This is been followed by the fever with the ICF of 0.95. Similar findings have already been reported by **Ayyanar and Ignacimuthu in 2011**. However, **Kadir *et al.* (2013)** and **Singh *et al.* (2012)** described a greater number of species to treat fever compared to dermatological ailments. The ICF value of different disease categories was ranged from 0 (nervous disorder) to 0.39 (GIT diseases). The ICF values indicated the maximal networking of indigenous people in the sharing of their knowledge on medicinal practices; this is usually the case with traditional healers who treat the most frequently encountered diseases in the study area. These high ICF values indicate reasonable reliability of informants on the use of MP species **(Lin *et al.*, 2002).** Pharmaceutical and phytochemical studies should be undertaken to study whether the use of these herbs is valid.

RELATIVE FREQUENCY OF CITATION (RFC) AND USE VALUE (UV):

The RFC and UV indices were applied to select potential plant species for further pharmacological study and recommendation in drug development. The relative frequency citation (RFC) index authenticates the frequency of citation of a medicinal plant species used for various ailments. The RFC of the reported species ranged from 0.1 to 2.41. Highest RCF value of 2.41 was noted for *Ocimum tenuiflorum* which is

Table 4: Informant Consensus Factor (ICF) Values by Category for Treating Various Diseases

S. No	Category	Plant Species Used and Number of Citations	Total number		ICF
			Species	Use citation	
1.	**Skin Disease (SD)**	*Abutilon indicum* (11), *Achyranthes aspera (23), Leucas aspera (17), Ocimum tenuiflorum (27), Solanum trilobatum (14), Ricinus communis (27), Phyla nodiflora (8), Cardiospermum halicacabum (10), Cleome viscosa (5), Cleome gynandra (9), Luffa cylindrica (8)*	11	159	0.94
2.	**Hair Problem**	*Ocimum tenuiflorum (13), Tridax procumbens (7), Ricinus communis (27)., Phyla nodiflora (21), Cassia angustifolia (9), Cardiospermum halicacabum (12),*	6	89	0.93
3.	**Cold (C)**	*Leucas aspera (22), Ocimum tenuiflorum (61), Solanum trilobatum (61), Solanum nigrum (15)*	4	159	0.98
4.	**Head ache (HA)**	*Abutilon indicum (8), Leucas aspera (22), Ocimum tenuiflorum (37), Solanum trilobatum (32), Solanum nigrum (13), Tribulus terrestris (8), Momordica cymbalaria (5), Cleome gynandra (4), Commelina communis (5), Argemone Mexicana (4)*	10	138	0.93
5.	**Fever (F)**	*Abutilon indicum (9), Achyranthes aspera (6), Leucas aspera (21), Ocimum tenuiflorum (60), Solanum trilobatum (48), Solanum nigrum (15), Tribulus terrestris (7), Commelina communis (9), Argemone mexicana (1)*	9	176	0.95

6.	**Menstural issues (MI)**	*Abutilon indicum (4), Ocimum tenuiflorum (10), Solanum nigrum (22), Ricinus communis (8), Citrullus lanatus (13), Momordica cymbalaria (6), Cucumis melo (15), Momordica charantia (12), Lagenaria siceraria (11), Celosia cristata (2)*	10	103	0.91

Table 5: Ailment Type, Used Value, Relative Frequency of Citation, Relative Popularity Level and Rank Order Priority of Selected Medicinal Plants

S. No	Name of the plant	Ailment Type	UV	RFC	RPL	ROP
1.	*Abutilon indicum*	SD, HA, F, MI	0.125	0.372	0.046	46
2.	*Achyranthes aspera*	SD, F	0.039	0.593	0.023	23
3.	*Leucas aspera*	SD, C, HA, F	0.066	0.697	0.046	46
4.	*Ocimum tenuiflorum*	SD, HP, C, HA, F, MI	0.24	2.41	0.058	58
5.	*Solanum trilobatum*	SD, C, HA, F	0.21	1.80	0.047	64
6.	*Solanum nigrum*	C, HA, F, MI	0.061	0.755	0.046	47
7.	*Tribulus terrestris*	HA, F	0.134	0.98	0.026	26
8.	*Tridax procumbens*	HP	0.142	0.081	0.011	11
9.	*Ricinus communis*	SD, HP, MI	0.048	0.720	0.034	34
10.	*Phyla nodiflora*	SD, HP	0.068	0.337	0.023	23
11.	*Cassia angustifolia*	HP	0.111	0.104	0.011	11
12.	*Luffa cylindrica*	SD	0.125	0.093	0.011	11
13.	*Citrullus lanatus*	MI	0.075	0.151	0.011	11

14.	*Momordica cymbalaria*	HA, MI	0.181	0.127	0.023	23
15.	*Cucumis melo*	MI	0.066	0.174	0.011	11
16.	*Momordica charantia*	MI	0.083	0.139	0.011	11
17.	*Lagenaria siceraria*	MI	0.090	0.127	0.011	11
18.	*Celosia cristata*	MI	0.05	0.023	0.011	11
19.	*Cardiospermum halicacabum*	SD, HP	0.090	0.255	0.023	23
20.	*Cleome viscosa*	SD,	0.02	0.058	0.011	11
21.	*Cleome gynandra*	SD, HA	0.158	0.151	0.023	23
22.	*Commelina communis*	HA, F	0.142	0.162	0.023	23
23.	*Argemone mexicana*	HA, F	0.04	0.058	0.023	23

Skin disease - SD, Hair Problem HP, Cold C, Head Ache - HA, Fever - F, Menstrual Issues - MI

Table 6: Fidelity Level Percentage of Medicinal Plants Based on Various Categories of Diseases

S. No	Ailment	Plant	FL%
1.	Skin Disease	*Abutilon indicum*	12
		Achyranthes aspera	26
		Leucas aspera	19
		Ocimum tenuiflorum	31
		Solanum trilobatum	16
		Ricinus communis	31
		Phyla nodiflora	9
		Cardiospermum halicacabum	9
		Cleome viscosa	11
		Cleome gynandra	5
		Luffa cylindrica	9
2.	Hair Problem	*Ocimum tenuiflorum*	15
		Tridax procumbens	8
		Ricinus communis	24
		Phyla nodiflora	31
		Cassia angustifolia	10
		Cardiospermum halicacabum	13
3.	Cold	*Leucas aspera*	25
		Ocimum tenuiflorum	70
		Solanum trilobatum	70
		Solanum nigrum	17
4.	Head ache	*Abutilon indicum*	9
		Leucas aspera	25
		Ocimum tenuiflorum	43
		Solanum trilobatum	37
		Solanum nigrum	15
		Tribulus terrestris	33
		Momordica cymbalaria	5
		Cleome gynandra	4
		Commelina communis	5
		Argemone mexicana	4
5.	Fever	*Abutilon indicum*	10
		Achyranthes aspera	6
		Leucas aspera	24
		Ocimum tenuiflorum	69
		Solanum trilobatum	55
		Solanum nigrum	17
		Tribulus terrestris	8
		Commelina communis	10
		Argemone mexicana	1
6.	Menstrual issues	*Abutilon indicum*	4
		Ocimum tenuiflorum	11
		Solanum nigrum	25

		Ricinus communis	9
		Citrullus lanatus	15
		Momordica cymbalaria	6
		Cucumis melo	17
		Momordica charantia	13
		Lagenaria siceraria	12
		Celosia cristata	2

borne on the same plant (monoecious) in terminal panicle-like inflorescences . The fruit is a spiny, greenish. Castor seeds have a warty appendage called the caruncle.

2. *Jatropha gossypiifolia* L.

Family: Euphorbiaceae

Habit: Shrub

Useful Parts: Leaves, Stem, Roots, Seeds and latex

Uses: Mainly used for biodiesel production. And also used for pesticide, insecticide vermifuge and ornamentation and even it was used in religious rituals.

Description: A small, much-branched shrub. 3-5 lobed, cordate, deep-purplish red at first afterwards green, lobes elliptic-acute, petiole red, covered with glandular hairs, branched from the base and mixed with simple hairs, stipules with glandular hairs, the margins of the leaf ciliate, with simple white hairs and gland tipped hairs. Terminal cymes inflorescence. Flowers are monoecious, Calyx cup-like with 5 lanceolate sepals. Petals 5, dark red, broadly ovate, stamens 6-8, dark-red, broadly ovate, anthers horse-shoe shaped, red, filaments connate in a central column on a glandular disk. Ovary seated on a glandular disk. Styles 3, united below, each ending in bifid stigma. Fruits are capsule 3 furrowed, truncate at both ends, seeds red with a caruncle. Flowering and Fruiting time is August-November.

3. *Chrozophora brocchiana* (Vis.) Schweinf.

Family: Euphorbiaceae,

Habit: Herb

Uses: In Iran, the plant is used to care for warts, emetic, cathartic, and fever whereas root ashes are given to children for cough.

Useful Parts: Leaves, Root, Stem

Description: Herb to undershrub, up to 60 cm high; flowering twigs 2–2.5 mm thick. Indumentum consisting of stellate and (few) simple hairs. Stipules 1.3-2.7 by c. 0.3 mm. Leaves: petiole 0.8–5.5 cm long; blade ovate, not to usually distinctly 3-lobed,

followed by *Solanum trilobatum* and *Tribulus terrestris* with 1.80 and 0.98 RCF value. The positions of these plant species correspond to the fact that they were reported by maximum number of informants, therefore having high frequency of citation (FC). The high values of RFC can be explained by the fact that these plants are the best known and have long been used by the majority of informants, representing a source of reliability. In fact, many biological activity and phytochemical evaluations have been carried out for these plants, and these species are particularly interesting for research in bioactive compounds. The plant species with high RFC should be subjected to pharmacologic, phytochemical, and other biological studies to evaluate and prove their authenticity **(Mukherjee *et al.*, 2012).**

The use value (UV) index demonstrates the relative importance of plant species and families for a population. In the present investigation, the UV of the reported medicinal plant species varied from 0.02 to 0.24 as shown in **Table 5**. The highest UV was observed for *Ocimum tenuiflorum* with 0.24 followed by *Solanum trilobatum* with the value of 0.21. These findings demonstrate the extensive use of above-mentioned species in the treatment of various ailments by local inhabitants/healers and the consciousness of indigenous peoples, which makes such medicinal plants, the first choice to treat a disease **(Jin *et al.*,2011).**

RELATIVE POPULARITY LEVEL (RPL):

Our 86 informants cited 23 plant species for 6 different disease categories. They are given in **Table 5**. *Ocimum tenuiflorum* is considered to be popular plant of that area due to their higher RPL value of 0.058 which is closely followed by *Solanum trilobatum* with the RCF value of 0.047. The high popularity of these plant species might be attributed to their high efficacy and the awareness of indigenous peoples which specifies their use as herbal medicine. This is the first baseline study

borne on the same plant (monoecious) in terminal panicle-like inflorescences . The fruit is a spiny, greenish. Castor seeds have a warty appendage called the caruncle.

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on the indigenous knowledge of local peoples regarding the use of popular plant species for a particular ailment. These findings were in consistent with previous studies on the status of medicinal plants among Bedouins of Negev district (Friedman *et al.*,1986) and medicinal plant survey in Palestinian area **(Ali-Shtayeh *et al.*,2000).**

FIDELITY LEVEL (FL):

The fidelity value is an important means to see for which ailment a particular species is more effective. It indicates the informants' choice for each ailment and the potential of the species related to the ailments. The FL of plant species for specific diseases in the present study area is shown in **Table 6**. For each disease category, different plants showed different FL percentage. For skin disease category, among the 11 species cited, *Ricinus communis* and *Ricinus communis* showed the major FL value of 31% which is followed by *Achyranthes aspera* with the FL of 26%. The least value was reported in *Cleome gynandra* 5%. Out of 6 plants used to treat hair problem the highest FL is noted for *Phyla nodiflora* (31%) and least is noted in *Tridax procumbens* (8%). To treat cold *Ocimum tenuiflorum* and *Solanum trilobatum* are greatly used with the highest FL of 70%. To treat head ache and fever, *Ocimum tenuiflorum, Solanum trilobatum* are greatly used with the highest FL of 43%, 37% and 69%, 55% respectively. The menstrual issues of women can be treated using *Solanum nigrum* and *Cucumis melo* with the FL of 25% and 17% respectively. The medicinal plants that are widely used by the people of the Rif have higher FL values than those that are less popular. On the other hand, more medicinal plants that are known as remedies of a single aliment have a 100% FL than those that are used as remedies for more than one type of aliment **(Verma *et al.*, 2007).**

RANK ORDER PRIORITY (ROP):

The Rank order priority (ROP) index is used to rank appropriately the plant species with different FL values. The ROP values are thus obtained are given in **Table 6.** Of the 23 species, two species attained ROP above 50. This is probably due to decreasing popularity of herbal medicines among the local communities of the study area. Based on ROP value *Ocimum tenuiflorum* and *Solanum trilobatum* are widely used species with the RPL of more than 50%. The ROP values reported for medicinal plants used by Bedouins community in Negev district and in Palestinian area were comparable to present findings **(Ali-Shtayeh *et al.*, 2000).**

DESCRIPTION OF PLANTS IDENTIFIED IN THE STUDY AREA (PLATES 1 – 18)

1. *Ricinus communis* L.

Family: Euphorbiaceae

Habit: Shrub

Usefull Parts: Leaves, Root bark, Seeds.

Uses: Methanolic extracts of the leaves of Ricinus communis were used in antimicrobial testing against eight pathogenic bacteria in rats. Antihistimine and anti-inflammatory properties were also found.

Description: *Ricinus communis* can vary greatly in its growth habit and appearance. The variability has been increased by breeders who have selected a range of cultivars for leaf and flower colours, and for oil production. It is a fast-growing, suckering shrub that can reach the size of a small tree, around 12 metres (39 feet), but it is not cold hardy. The glossy leaves are 15–45 centimetres (6–18 inches) long, long-stalked, alternate and palmate with five to twelve deep lobes with coarsely toothed segments. The fruit capsules of some varieties are showier than the flowers. The flowers lack petals and are unisexual (male and female) where both types are

2.6–9 by 2.6–9 cm, index 1–1.9, pale green, base obtuse to usually emarginate, margin without distinct glands, lower surface with 2 glands near the base, 1 mm in diam., and usually several smaller ones sub marginally, venation impressed above, raised below, nerves c. 4 per side. Inflorescences up to 4 cm long, elongating in fruit to up to 10 cm long. Bracts very inconspicuous to c. 1.6 by 0.3 mm. Staminate flowers 4–6 mm in diam., yellow; calyx white, united part c. 1 mm high, lobes 3.2–4 by c. 1.2 mm; petals 3.7–3.8 by 1.5–1.6 mm; androphore 3.3–3.8 mm long, basal 1.2–1.3 mm without filaments; filaments 0.3–0.8 mm long; anthers 0.9–1.3 by c. 0.7 mm, yellow. Pistillate flowers 3.2–3.3 mm in diam., (greenish to) yellow; pedicel 1.4–2 mm long, elongating in fruit to up to 1.1 cm; calyx lobes only basally united, 1.5–2.2 by 0.5–0.7 mm; petals 1.3–2 by 0.4–0.6 mm; ovary ovoid, 2.7–3 by 2.2–3 mm wide; style 0.5–0.8 mm long, red, stigmas erect, up to 2.3 mm long, apically split for up to 1.8 mm, red. Fruits 8–9 by c. 5 mm; column after dehiscence 3–3.5 mm long. Seeds 3.8–3.9 by 3.2–3.6 by 3–3.2 mm.

4. *Euphorbia peplus* L.

Family: Euphorbiaceae

Habit: Herb

Uses: It is used to treat treating actinic keratoses and nonmelanoma skin cancer.

Useful Parts: Leaves and Root.

Description: Radium Weed is an annual plant growing to between 5-30 cm tall. They are commonly found small in size, growing as weeds of cultivation. Stems are smooth, hairless. Leaves are oval-pointed, 1-3 cm long, with a smooth margin. It has green flowers in three-rayed umbels. The glands, typical of the spurge family, are kidney-shaped with long thin horns. In India it is found in Jammu and Kashmir and Meghalaya.

5. *Euphorbia serpens* Kunth.

Family: Euphorbiaceae

Habit: Herb

Useful Parts: Leaves, Flowers and Seeds.

Uses: It is used to treat breathing disorders.

Description: *Euphorbia serpens* is a species of Euphorbia known by the common name matted sandmat. It is native to South America but it can be found on most continents as an introduced species and often a weed. This is an annual herb forming a mat of prostrate stems which root at nodes where the stem comes in contact with the ground. The oval leaves occur in oppositely arranged pairs, each leaf less than a centimetre long. The inflorescence is a cyathium with scalloped white petal-like appendages surrounding the actual flowers. A red nectar gland is at the base of each appendage, and at the centre of the cyathium are several male flowers around one female flower. The fruit is a lobed, spherical capsule.

6. *Croton bonplandianus* Baill.

Family: Euphorbiaceae

Habit: Herb

Useful Parts: Leaves and Stem

Uses: It is used to treat liver disorders, skin diseases including ring worm infection, to cure the swelling of body, bronchitis and asthma.

Description: This is a wild species of croton. Due to the resemblance of the leaves and flower cymes to that of Tulsi, this plant is often called Ban Tulsi (Jungle Tulsi) It is a small annual herb, growing up to 1-2 ft tall. Alternately arranged leaves, 3-5 cm long, are lance-shaped, with a toothed margin. Small white flowers are borne in 3-8 cm long racemes at the end of branches. Flowers have 5 sepals and 5 petals and numerous long stamens protruding out. Fruit is a 5 mm oblong capsule, with a warty surface. Ban Tulsi is grown abundantly in the rural areas of Malda, West Bengal, and is used as both a fuel and a detergent. First the stems and branches of ban tulsi are used as fuel. Then the ash is collected and kept in a bottle for five or six days. The ash is put in warm water and used as a detergent for cleaning cotton garments. Ban Tulsi is native to S. Bolivia to Uruguay, now widespread in the Indian subcontinent. Flowering: September-November.

7. *Euphorbia prostrate* Aiton.

Family: Euphorbiaceae

Habit: Herb

Useful Parts: Leaves, Seeds and Roots

Uses: It is used to treat piles, bronchitis, asthma, and tumour

Description: Euphorbia prostrata is an annual herb producing slender prostrate stems up to approximately 20 centimetres (7.9 in) long, sometimes purple-tinted in colour. The oval-shaped leaves are up to one centimetre (0.39 in) long with finely toothed edges. The inflorescence is a cyathium less than two millimetres (0.079 in) wide, with white petal-like appendages surrounding the actual flowers. There are four male flowers and a single female flower, the latter developing into a lobed, hairy fruit one to two millimetres (0.039 to 0.079 in) wide.

8. *Stachytarpheta jamaicens* (L.)

Family: Verbenaceae

Habit: Herb

Useful Part: Leaves

Uses: The fresh leaves are consumed in bush tea in cooling tonic and blood cleanser to treat asthma and ulcerated stomachs.

Description: Low, usually spreading annual or perennial herbs, 6-12 dm tall, sometimes somewhat woody toward base, often purplish throughout; stems dichotomously branched, sparsely pubescent or glabrate, nodes usually sparsely pilose. Leaves often bluish or greyish when fresh, alternate or opposite, somewhat fleshy, oblong to elliptic or ovate, 2-9 (-12) cm long, 1.5-4 cm wide, upper surface glabrate, lower surface glabrous to strigose on the veins and margins serrate, the teeth angled forward, petioles 0.3-3.5 cm long. Spikes stout and stiff, 15-50 cm long, glabrous, rachis stout and firm, up to 7 mm in diameter, the furrows conspicuously narrower than the rachis, bracts lanceolate or oblong-lanceolate, 5-8 mm long; calyx completely embedded in rachis furrows, somewhat

compressed, ca. 5 mm long, 2 of the teeth very reduced; corolla usually pale blue, 8-11 mm long.

9. *Lantana camara* L.

Family: Verbenaceae

Habit: Shrub

Useful Part: Leaves

Uses: It has anti-insecticidal, Fungicidal and antimicrobial properties and it is used to cure cancer, skin itches, leprosy, chicken pox, measles, asthma and ulcers.

Description: *Lantana camara* is a perennial, erect sprawling or scandent, shrub which typically grows to around 2 metres. Under the right conditions, it can scramble up into trees and can grow to 6 m (20 ft) tall. The leaves are broadly ovate, opposite, and simple and have a strong odour when crushed. It has small tubular-shaped flowers, which each have four petals and are arranged in clusters in terminal areas stems. Flowers come in many different colours, including red, yellow, white, pink and orange, which differ depending on location in inflorescences, age, and maturity. The flower has a smell with a peppery undertone. After pollination occurs, the colour of the flowers changes; this is believed to be a signal to pollinators that the pre-change colour contains a reward as well as being sexually viable, thus increasing pollination efficiency. In frost-free climates the plant can bloom all year round, especially when the soil is moist.

10. *Lantana aculeate* L.

Family: Verbenaceae

Habit: Shrub

Useful part: Leaves

Uses: It has anti-insecticidal, Fungicidal and antimicrobial properties and it is used to cure cancer, skin itches, leprosy, chicken pox, measles, asthma and ulcers.

Description: The plant has various medicinal applications such as sedative, to relieve urinary tract infections, treatment of chest complaints, to treat snake bite and intoxication. It is used in flavouring in cakes, sweet breads and candy.

11. *Phyla nodiflora* (L.) Greene

Family: Verbenaceae

Habit: Herb

Useful parts: Leaves and Fruits

Uses: Plant decoction is given in uraema. Fresh juice is applied to bleeding gums. Infusion of leaves and tender stalk is given to children in indigestion and to women after delivery.

Description: Appressed pubescent, prostate to ascending or decumbent, perennial herbs, rooting at the nodes, obscurely to definitely 4- angled. Leaves opposite, serrate, base cuneate to attenuate; petioles to 0.5 mm long, often obscured by decurrent blade tissue. Inflorescence a bracteate head, in fruit a spike 8-15 cm long, 5-8 mm in diam., peduncles elongate, usually at alternate nodes and rarely in both axils at a node. Sepals united near base or for ½ their length, shorter than the corolla tube and the subtending bract; corolla zygomorphic, pinkish, lavender or rarely white, salver form, ca. 3 mm long, 5-lobes less than 1 mm long; stamens included, united to the corolla tube near middle at 2 levels. Fruit a schizocarp consisting of 2 mericarps.

12. *Acacia tortilis* (Forssk).

Family: Leguminosae

Habit: Tree

Useful Part: Leaves

Uses: *Acacia tortilis* possesses valuable medicinal property and therapeutic potential so, it is also useful for treatment of various diseases like skin allergy, cough, inflammatory reaction.

Description: *Acacia tortilis* is a small to medium-sized evergreen tree or shrub that grows up to 21 m tall. Leaves glabrous to densely pubescent, glandular, short at 1.25-3.75 cm long; petiole 0.2-0.9 cm long. Inflorescence globose heads; peduncle white, pubescent, 0.4-2.5 cm long, with involucel on the lower half; flowers white or pale yellowish-white, sessile or shortly pedicellate, scented, 0.5-1.1 cm in diameter, on axillary peduncles; calyx 1-2 mm long; corolla 1.5-2.5 mm long. Pods variable, indehiscent, spirally twisted or rarely almost straight, 7-10 cm long, 6-10 (max. 13) mm broad, longitudinally veined, leathery, glabrous to tomentellous or villous, somewhat constricted between the seeds; seeds oblique or parallel to long axis of pod, 4-7 x 3-6 mm, compressed; areole 3-6 x 2-4 mm.

13. *Senna occidentalis* (L.) Link

Family: Leguminosae

Habit: Shrub

Uses: The seed is bitter and has purgative properties. It is also used as a diuretic, liver detoxifier, as a hepato-tonic (balances and strengthens the liver). Further, used in whooping cough and convulsion.

Useful Part: Seed

Description: *Senna occidentalis* is a Fabaceae family weed that grows in damp, disturbed, or junkyards at low altitudes throughout the world. First discovered in tropical South America, the plant is deadly if consumed in excessive numbers; however, all components of the plant are utilised as medicine and food by many people worldwide. The seeds are frequently used as a coffee replacement. It is a 0.5-2.5 m tall, unarmed thin straight shrub. It is a yearly or eternal Ayurvedic plant used in numerous traditional medicines to treat various ailments. This weed contains germ-destroying, antimycotic, antidiabetic, anticancer, antimutagenic, anti-inflammatory, and antihepatotoxic properties.

15. *Cassia angustifolia* Vahl.

Family: Caesalpiniaceae

Habit: Shrub

Useful Parts: Leaves, Pods and Flowers

Uses: It is used to treat stomach pain and constipation**.**

Description: Alexandrian Senna is a shrubby plant that reaches 0.5–1 metre (20" to 40"), rarely two metres (6') in height with a branched, pale-green erect stem and long spreading branches bearing four or five pairs of leaves. These leaves form complex, feathery, mutual pairs. The leaflets vary from 4 to 6 pairs, fully edged, with a sharp top. The midribs are equally divided at the base of the leaflets.

16. *Pergularia daemia* (Forsskal) Chiov

Family: Asclepiadaceae

Habit: Herb

Useful Part: Leaves, Root bark, Stem.

Uses: Used to cure cough, asthma, rheumatism, bronchitis, pies, liver disorder, uterus related problems, digestion related problems, reduces the swelling, inflammation and pain in the joints.

Description: *Pergularia* is a perennial twining herb, foul-smelling when bruised and with much milky juice, stem hairy. Leaves are thin, broadly ovate, heart-shaped or nearly circular, hairless above, velvety beneath. Greenish yellow or dull white, and sweet-scented flowers are borne in lateral cymes which are at first corymb-like, afterwards raceme-like. The five petals are hairy and spreading outwards. Corona outer and inner, outer truncate, inner curved high over the staminal column, spur acute. Fruit is a follicle, with soft spines all over and a long beak. Seeds are densely velvety on both sides. Flowering: August-February.

17. *Oxystelam esculentum* R. Br.

Family: Asclepiadaceae

Habit: Climber

Useful Parts: Leaves and Flower

Uses: This plant has strong phyto-medicinal constituents and used for remedies from cancer, hepatitis, kidney disorders, stress-related disorders and various microbial infections

Description: Twinging subshrubs. Leaves simple, opposite, 4-8 x 0.5-2 cm, oblong to linear, sub coriaceous, base truncate, apex mucronate; petiole ca. 1 cm. Flower(s) usually paired, axillary, solitary or in lax racemes; peduncle 6-15 cm; bracts deciduous; pedicel ca. 1 cm. Calyx copular; lobes equal, ovate, imbricate, 4 mm, chartaceous, glabrous, acute. Corolla white or pink with dark pink stripes within, 2.5 cm across, rotate to angulate; lobes triangular, united at the mid half, valvate, 2 cm, shortly overlapping to right in bud, chartaceous, ciliate, acute, recurved. Pollinia pendulous; pollinial bags oblong, 1.3 mm; caudicle 0.2 mm. Corona double; outer coralline, annular, pubescent at the base of corolla within; inner staminal, basally inflated, 5 mm; staminal column 7 mm. Ovaries globose, 3 mm; placenta bifurcate; style 4 mm. Follicle 5 x 2 cm, inflated, mucronate; seeds ovate, 2.5 x 2 mm, base rounded, apex pointed; coma silky, dull white.

18. *Calotropis procera* (Aiton) Dryand.

Family: Asclepiadaceae

Habit: Shrub

Useful Parts: Roots, Stem, Leaves and flowers

Uses: Arkavaleha', made from this plant, is given to cure irritation of the stomach, nausea, vomiting, diarrhoea etc. Eight patents were found on the medicinal uses mainly for anti-tumour and antidote activity, and bronchial asthma.

Description: Shrub or small tree up to 2.5(–6) m tall, stems erect, simple or branched, woody at base, bark grey or pale brown, fissured, corky, slash yellowish-white, latex copious; young branches densely white hairy, soon almost glabrous. Leaves opposite, decussate, simple and entire, almost sessile; stipules absent; blade oblong-obovate to broadly obovate, 5–30 cm × 2.5–15 cm, apex abruptly and shortly acuminate, base cordate, succulent, densely white short-hairy below when young, pinnately veined with 6–10 pairs of lateral veins. Inflorescence an axillary umbellate cyme up to 10 cm

in diameter. Flowers bisexual, regular, 5-merous; pedicel 1–3 cm long; calyx lobes ovate, 4–7 mm × 3–4 mm; corolla pale whitish-green with large lilac to purple patches on the lobes, campanulate, 2–3 cm in diameter, lobes broadly triangular, 11–20 mm × 9–10 mm, united at base for 6–7 mm; corona with 5 compressed lobes, 6.5–11 mm × 3–4.5 mm, adnate to the staminal column, purple; ovary superior, 2-celled, gynostegium c. 6 mm long, stigma head 5-pointed. Fruit a pair of follicles, each follicle ovoid, fleshy, inflated, 6–10 cm × 3–7 cm, many-seeded. Seeds ovoid, flattened, c. 6 mm long, with 3–4 cm long white coma at one end**.**

19. *Calotropis gigantea* (L.) Dryand

Family: Asclepiadaceae

Habit: Shrub

Useful Part: Flower

Uses: The plant is reported as effective in treating skin, digestive, respiratory, circulatory and neurological disorders and was used to treat fevers, elephantiasis, nausea, vomiting, and diarrhea. The milky juice of Calotropis procera was used against arthritis, cancer, and as an antidote for snake bite

Description: Large, white, not scented, peduncles arising between the petioles. Flower-buds ovoid, angled, Calyx lobes 5, divided to the base, white, ovate; corolla broadly rotate, valvate, lobes 5, deltoid ovate, reflexed, coronate-appendages broad, obtusely 2-auricled below the rounded apex which is lower than the staminal-column. Stamens 5, anthers short with membranous appendages, inflexed over the depressed apex of the pentagonal stigma. Pollinium one in each cell, pendulous caudicle slender. Carpels 2 distinct, styles 2, united to the single pentangular stigma, ovary 2-celled, ovules many. Fruit: A pair of follicles with many, hairy seeds.

20. *Ziziphus spina-christi* (L.) Desf.

Family: Rhamnaceae

Habit: Shrub

Useful Part: Fruits

Uses: It is used for improving muscular strength and weight, for preventing liver diseases and stress ulcers, and as a sedative.

Description: A medium-size tree, with spreading, greyish white branches, glabrous or slightly pubescent. Stipular spines in pairs, one erect, c. 2 cm long, the other recurved 5-8 mm long, sometimes spines absent. Leaves 2-6 x 1-4 cm ovate-elliptic or suborbicular, glabrous or pubescent on nerves beneath, rounded to sub cordate at base, obtuse or shortly acuminate, margin entire or obsoletely crenate, 3-nerved; petiole 3-12 mm long, glabrous. Inflorescence axillary tomentose, pedicel woolly, c. 3-5 mm long. Flowers 4-6 mm across, greenish yellow. Calyx c. 1 mm long, keeled within, pubescent, ovate, ± acute, petals spathulate; 1.25 mm long. Disc prominently 10-lobed, glabrous, grooved.

21. *Tridax procumbens* (L.)

Family: Compositae

Habit: Herb

Useful Part: Whole Plant

Uses: It has been in use in India for wound healing and as an anticoagulant, antifungal, and insect repellent. It is also used as treatment for boils, blisters, and cuts by local healers in parts of India

Description: A decumbent herb. Stem and leaves are covered by hairs. Tap root system. Herbaceous, cylindrical, decumbent and branched. Simple, opposite, exstipulate and margins dentate showing reticulate venation. A terminal heterogamous head and receptacle of the head is convex and surrounded by green involucre. The tubular florets occupy the centre and the ligulate florets are found at the margins.

22. *Parthenium hysterophorus* L.

Family: Asteraceae

Habit: Herb

Useful Part: Leaves

Uses: *Parthenium hysterophorus* confers many health benefits, viz remedy for skin inflammation, rheumatic pain, diarrhoea, urinary tract infections, dysentery, malaria and neuralgia.

Description: This erect ephemeral herb can grow up to 1.5–2 m high and has a deep tap root. It has branching stems that become woody and hairy with age. Leaves are alternate, finely lobed, covered with fine soft hair, 3–20 cm long and 2–10 cm wide. Once stem elongation is initiated, smaller leaves are produced and the plant becomes multi-branched in its extremities. The whole plant has a bluish or greyish-green appearance. Flower heads are small (4 mm across) and numerous in open panicles, creamy-white, with 5 petals. Each flower produces about 5 black achenes which are obovate, 2–2.5 mm long and light weight. The fruit is a cypsela

23. *Stoebe plumose* L.

Family: Asteraceae

Habit: Herb

Useful Part: Underground parts

Uses: The form of *Seriphium plumosum* which was previously known as *Stoebe plumosa* was not used medicinally. However, in the Western Cape, the form previously known as *Stoebe cinerea* is still used as a remedy for heart trouble, whereas another unidentified Stoebe/Seriphium sp**.**

Description: This intricately branched, heath-like shrublet is well known to hikers who use the plant to make soft mattresses when sleeping outdoors. It might be better known to many people as Stoebe plumosa. Despite enormous garden potential and successful use at Kirstenbosch, it is not yet commonly cultivated Preparations of this species are taken orally for gynaecological problems, stomach ache. *Stoebe plumosa* Herb consists of the fresh or dried over ground parts of Stoebe plumose (L.) Thunb.

24. *Sida spinosa* L.

Family: Malvaceae

Habit: Herb

Useful Parts: Leaves and Root

Uses: It is used in the treatment of neurological and uterine disorders, headache, tuberculosis, diabetes, malarial fever, piles, ulcers, wounds, rheumatic and cardiac problems, diarrhoea and dysentery, skin diseases.

Description: The stems are erect to sprawling and branched, growing 50 to 120 centimetres in height, with the lower sections being woody. The dark green, diamond-shaped leaves are arranged alternately along the stem, 4 to 8 centimeters long, with petioles that are less than a third of the length of the leaves. The leaves are paler below, with short, greyish hairs. The apical half of the leaves have toothed or serrated margins while the remainder of the leaves are entire (untoothed). The petioles have small spiny stipules at their bases. The moderately delicate flowers occur singly on flower stalks that arise from the area between the stems and leaf petioles. They consist of five petals that are 4 to 8 millimeters long, creamy to orange-yellow in color, and may be somewhat reddish in the center. Each of the five overlapping petals is asymmetric, having a long lobe on one side. The stamens unite in a short column. The fruit is a ribbed capsule, which breaks up into 8 to 10 segments. The plant blooms throughout the year.

25. *Sida cordifolia* L.

Family: Malvaceae

Habit: Perennial shrub

Useful Part: Leaf and root

Uses: It is used to treat bronchial asthma, cold and flu, chills, lack of perspiration, head ache, nasal congestion, aching joints and bones, cough and wheezing, and oedema.

Description: It is a perennial shrub. The root is tap root system. The stem is woody and round, the bark hoary and woody with soft stellate hairs. The leaves are alternate, broadly ovate, 3-7 cm long and 3-5 cm wide. The leaf margins are dentate, rounded at

the base. The blade is soft to the touch and woolly on the lower surface and along the main nerves with fine stellate hairs. The leafstalk is long, about 4.5 cm. The inflorescence is an axillary or terminal raceme, leafy with many flowers. Calyx 5-7 mm long, accrescent and clasping in fruit; petals cream to pale yellow or orangish yellow, 7-9 mm long. The stamens are fused at the base tube pubescent, but the nets and the anthers are free. Fruit are capsules and each has 8-10 carpels. The fruit is hoary and densely covered with reflexed woody hairs; each carpel bears 2-bristly awns.

26. *Abelmoschus ficulneus* (L.)

Family: Malvaceae

Habit: Shrub

Useful Parts: Stems, Roots, Seeds, Fruit and Leaves

Uses: A good quality fibre is obtained from the stems. The white fibre is long, glossy, fine and strong. It is used for twine and light cordage

Description: Annual herb up to 2 m tall; stem thick, glabrous to densely glandular pubescent. Leaves alternate, simple stellate hairy; stipules linear or filiform, 5–12 mm long, hirsute; petiole 2–21 cm long, hairy; blade orbicular, deeply 3–5-lobed, up to 16 cm × 16 cm, cordate at base, lobes subacute to broadly rounded, margin serrate, scabrous on both sides. Flowers bisexual, regular, solitary in leaf axils or in a terminal raceme; pedicel 0.5–2.0(–2.5) cm long, expanded and cup-shaped apically; epicalyx bracts 5–6, linear to lanceolate, up to 12 mm × 2 mm, rough, caducous before expansion of corolla; calyx 17–23 mm long, 5-toothed, tomentellous; petals 5, obovate, 2–3.5 cm × 1.5–3 cm, uniformly white, turning pink; stamens many, filaments united in a column 1–1.5 cm long, glabrous; ovary superior, 5-celled. Fruit an ellipsoid capsule 3–4 cm × 1.5–2 cm, puberulous to pubescent; valves acute to aristate with up to 3 mm long awns. Seeds globose, 3–4 mm in diameter, black, with concentric lines, glabrous or with stellate or long crisped hairs.

27. *Hibiscus calyphyllus* Cav.

Family: Malvaceae

Habit: Shrub

Useful Part: Leaves

Uses: *Hibiscus calyphyllus* is cultivated throughout the tropics and subtropics as an ornamental. In DR Congo, the leaves are used in a mixture with several other plant species to prepare a cure for ganglions in domestic animals. In Kenya and Tanzania, the leaves are applied to wounds as a dressing.

Description: *Hibiscus calyphyllus* is a dense, rounded shrub; up to 3 m high; the leaves are large, up to 50 mm in diameter, light green, soft and velvety; the flowers are lemon-yellow, large, up to 100 mm in diameter, with a deep red to blackish centre; the fruit is a papery capsule that splits open to reveal hairy to hairless seeds. It is fairly fast growing and will flower repeatedly, the flowers lasting for a reasonable amount of time. The natural habitat of Hibiscus calyphyllus is open bush, thickets and forests, often also found along rivers.

28. *Abutilon indicum* (L) Sweet.

Family: Malvaceae

Habit: Shrub

Useful Parts: Leaves and Root

Uses: It was mainly used to cure Diarrhoea, Gonorrhoea, Antipyretic, cough, piles**.**

Description: Soft-wooded, much-branched shrub up to c. 1.5 m tall. Leaves more or less broadly ovate-cordate, up to 18 × 16 cm, with a long acuminate tip, unlobed, discolours, dark green above, paler grey-green below with prominent veins; margin slightly but distinctly serrate-crenate; petiole up to 18 cm long. Flowers axillary, often on short axillary shoots, yellow, sometimes reddish at the base or with reddish veins, c. 3 cm in diameter. Calyx more or less cup-shaped, 10-18 mm long and 8-10 mm in diameter; lobes 6-12 mm long, ovate-lanceolate to lanceolate-linear, gradually acuminate. Fruit 20-25 mm in diameter, stellate-pubescent with 23-40 mericarps, 2-3-seeded, black when dry.

29. *Tribulus terrestris* L.

Family: Zygophyllaceae

Habit: Herb

Useful Parts: Leaves, Stem, Root

Uses: It is used to cure chest pain, heart problems, dizziness, skin and eye disorders, to expel kidney stones, and as a diuretic and tonic.

Description: It is a trailing perennial, hirsute, procumbent and branched herb. The stems and branches are pilose and young parts are silky-villous. Leaves are stipulate, opposite usually unequal and abruptly pinnate. Leaflets are 5-8 in pairs with length 0.5- 1.3 cm, sub-equal, oblong to linear oblong and mucronate; petioles very short and pilose. Flowers are yellow, solitary, axillary, 8-12 mm in diameter and appear during July-August. Style is short and stout; ovary is bristly 5-10 lobed and with 5-12 celled; fruits are globose and spinous produced during autumn. It consists of 5-12 woody cocci, each with two pairs of hard sharp spines, one pair longer than the other. Each coccus contains several seeds with transverse partition between them. The seeds are obliquely pendulous and have hard seed coat.

30. *Luffa cylindrica (*L.) M. Roem

Family: Cucurbitaceae

Habit: Climber

Useful Parts: Leaves, fruit, seed

Uses: It is used to cure pulmonary troubles, cancer and tumour.

Description: *Luffa* is a large climber with a stout, 5 angled pubescent stem tendrils usually 2–3 branched. Leaves are large, 10–20 cm long, and 9–21 cm wide. Orbicular to reniform orbicular in outline, base cordate, 5–7 lobed, lobes broadly triangular, acute, margins irregularly, shallowly dentate, hispid, scab rid above, finely pubescent-hispid beneath, petioles 2–8 cm long. The male and female flowers deep, bright yellow and male flowers borne in racemes on peduncles 6–17 cm long. It is usually aggregated near apex, stamens usually 15–30 cm long and 6–10 cm in diameter, smooth with 10 dark green longitudinal lines, fibrous within, seeds 1–2 cm long and 0.8 cm wide, including the narrow, smooth, marginal wing, broadly

ellipsoid, longitudinally compressed, rough on the surface The two species of *Luffa* are somewhat similar in appearance. Both are vigorous, climbing, annual vines with several lobed cucumber-like leaves. When crushed, the leaves give off a rank odour. Both male and female flowers occur on the plant with a much greater number of male flowers. The rather large male flowers are bright yellow and occur in clusters. The female flowers are solitary and have the tiny, slender ovary attached. Angled luffa flowers appear later in the day than the smooth type and stay open through the night. Pollination is by entomophilly.

31. *Citrullus lanatus* (Thunb.) Matsum & Nakai

Family: Cucurbitaceae

Habit: Climber

Useful Part: Fruit

Uses: Traditional herbal practitioners employ *C. lanatus* seeds to treat gastrointestinal, respiratory, and urinary diseases in Pakistan and India. However, more investigation is needed to understand the effect of C. lanatus seeds on treating gastrointestinal, respiratory, and urinary disorders.

Description: Watermelon is an annual herbaceous vine with long (up to 10 m) stems lying or creeping on the ground, with curly tendrils. Leaves are 5-20 by 3-19 cm, and hairy, usually deeply palmately lobed with 3-5 lobes. Leaf stalks are 2-19 cm long. Male flowers on 1.2-4.5 cm long pedicels. Flowers 1-2.5 cm long, pale green. Flowers monoecious, solitary, on pedicels up to 4.5 cm long; with 5 shortly united petals, pale green. Fruit of wild plants 1.5-20 cm in diameter, nearly spherical, greenish, mottled with darker green; of cultivated plants up to 30x60 cm, spherical or ellipsoid, green or yellowish, evenly coloured or variously mottled or striped. Fruits vary considerably in morphology. The cultivated forms of the fruit are large oblong.

32. *Momordica cymbalaria* (Hook & Fenzl)

Family: Cucurbitaceae

Habit: Climber

Useful Part: Root, Leaves and Fruit

Uses: Plant parts such as root, leaves and fruit were treated for various ailments.

Description: Little Wild Gourd is a perennial herb with slender, climbing, branched, striped stem. It is a trailing plant with large turnip shaped tuberous rootstock. The leaves are round-kidney-shaped in outline, deeply heart-shaped at the base, bluntly lobed with 5-7 lobes. flowers are white to yellow in color. Flowers are small, unisexual in nature. The male flower flower-cluster-stalk is 5-30 mm long, thread-like, hairy, ebracteate with 2-5 flowers in racemes with a pale-yellow flower and two stamens for each flower. The female flower is solitary on a flower-cluster-stalk of 2.8 cm long. The fruits are 2.0-2.5 cm long, pyriform with 8 sharp ridges, 2.4 x 1.5 cm narrowed at the tip and with the base narrowed into the curved flower-cluster-stalk, which is fleshy, dark green and robbed. Seeds are 4.6 mm long, ovoid shaped, black, smooth and shiny.

33. *Cucumis melo* L.

Family: Cucurbitaceae

Habit: Climbers

Useful Parts: Leaves and Fruit

Uses: It is used for the treatment of kidney stones, flatulence, leprosy, fever, jaundice, diabetes, obesity, cough, bronchitis, ascites, anaemia, constipation and other abdominal disorders.

Description: *C. melo* is an annual trailing vine with pubescent striated stems, lacking stipules, bearing unbranched tendrils at the base of the 4-12cm long petioles. The leaves are simple and alternate, nearly round, basally cordate, and may have 3-7 shallow palmate lobes. The flowers of C. melo can be gynoecium (only female flowers), monoecious (male and female flowers), or perfect. The hypanthium is

broader at the apex and 0.7-0.8cm long. Staminate flowers are solitary or fascicled, bearing 3 free stamens, of which two bear 2-celled anthers and one a 1-celled anther. Pistillate flowers are solitary, bear staminodes, an inferior ovary topped by a short style (1-2mm long) and 3-lobed stigma (2-2.4mm long). The ellipsoid ovary is densely pubescent with white hairs, and is 0.4-1.1cm long (3,9,11,13). The fruit is berry. The seed is endospermous.

34. *Momordica charantia* L.

Family: Cucurbitaceae

Habit: Climber

Useful Parts: Fruit, Flowers and Seeds

Uses: It is used to cure ulcers, breast cancer, diabetes and heart problems**.**

Description: It is annual to perennial herb. The stem is herbaceous, tendril-bearing vine grows to 5 meters. It may be either hairless or slightly hairy. The leaf is Up to 5cm long, with spiral tendrils at opposite sides, Petioles 4-5 cm long, pubescent. The leaves are carried singly along the stems, and each leaf is 4-10 cm long, rounded in outline, and deeply 5-9 lobed. The flowers are Monoecious pale to deep yellow flowers with bract part way on peduncle, solitary in the upper leaf axils on 2-10 cm long stalks with a small leaf-like bract towards the base. Male flowers larger than female flowers and have a slender basal swelling which is continuous with the base of the sepal tube, which ends in five blunt sepals. There are five oval yellow petals 10-20 cm long, and five central stamens. Female flowers are similar to the male flowers but have a distinct warty swelling well below the base of the sepal tube and three stigmas. The fruit is berry. The seed is endospermous.

35. *Citrullus colocynthis* (L.) Schrad

Family: Cucurbitaceae

Habit: Herb

Useful Parts: Fruit, seed and flower

Uses: It is used to cure diabetes, leprosy, common cold, cough, asthma, bronchitis, jaundice, joint pain, cancer, toothache, wound, mastitis, and in gastrointestinal disorders such as indigestion, constipation, dysentery, gastroenteritis

Description: The roots are large, fleshy, and perennial, leading to a high survival rate due to the long tap root. The vine-like stems spread in all directions for a few meters looking for something over which to climb. If present, shrubs and herbs are preferred and climbed by means of auxiliary branching tendrils. The leaves are palmate and angular with three to seven divided lobes. The flowers are yellow and solitary in the axes of leaves and are borne by yellow-greenish peduncles. Each has a sub campanulate five-lobed corolla and a five-parted calyx. They are monoecious, so the male (stamens) and the female reproductive parts (pistils and ovary) are borne in different flowers on the same plant. The male flowers' calyx is shorter than the corolla. They have five stamens, four of which are coupled and one is single with monadelphous anther. The female flowers have three staminoids and a three-carpel ovary. The two sexes are distinguishable by observing the globular and hairy inferior ovary of the female flowers. The fruit is mesocarp. The seeds are endospermous.

36. *Mukia maderaspatana* W.

Family: Cucurbitaceae

Habit: Climber

Useful Parts: Leaves, Root and Fruits

Uses: According to Siddha and Ayurveda medicine, the leaves, root and fruits are considered stomachic, anti-ulcer, anti-inflammatory, antipyretic, diuretic, hepatoprotective, expectorant, carminative, anti-hyperglycaemic, anti-hyperlipidaemic, antimicrobial, antioxidant and anti-rheumatic activities**.**

Description: Prostrate or climbing herbs; tendrils simple. Leaves 4-8 x 3-7 cm, ovate-deltoid, angular or shallowly 3-5-lobed, base cordate, margin denticulate, apex acuminate, mucronate, on both sides; petiole to 6 cm. Male flowers in axillary, sessile clusters. Calyx tube to 2 mm, villous; lobes subulate, erect. Petals 5, c. 3 mm long, ovate-oblong, obtuse, yellow. Stamens 3, free, inserted at base of calyx tube; anthers

oblong, ciliate. Female flowers solitary or in clusters. Ovary villous. Berry c. 1.2 cm across, globose, red. Seeds lenticular, rugose.

37. *Lagenaria siceraria* (Molina) Standley.

Family: Cucurbitaceae

Habit: Annual Climber

Useful Part: Fruit

Uses: It is used as a emetic, purgative, cooling, sedative, anti-bilious, and pectoral. Its pulp, boiled in oil is used to treat Rheumatism**.**

Description: The elongate fruits average 15–20 cm in length and several centimeters in diameter and are eaten prior to maturity, as the mature fruits are bitter. The seeds are covered with sweet, red arils, but the seeds themselves are poisonous. This species is grown mostly in southern Asia.

38. *Cucurbita maxima* Duch.

Family: Cucurbitaceae

Habit: Climber

Useful Parts: Seeds, Fruit and flowers**.**

Uses: The seeds are used to prepare, tonic diuretic and vermifuge. The flowers are used to soothe minor injuries. The fruit pulp is to cure burns, inflammations and ear ache.

Description: *C. maxima* is an annual herb with thick climbing or creeping stems. The root system is well developed and roots are up to 40 cm deep and 5 m long. The stems are branching, covered in soft white pubescence, up to 10 m long, and often produce adventitious roots at nodes. The petioles are densely pubescent, 5-20 cm long, and estipulate. The plant bears tendrils at 90 degrees to the leaf axil; these are lightly pubescent, coiled, and 2-5-branched. The thin leaves are alternate, simple, palmately veined, round to reniform, basally cordate, apically obtuse, unlobed to shallowly 5-7 lobed, 7-30 cm across, broader than long, stiff to soft pubescent, and finely dentate.

The flowers produce nectar and are aromatic. Staminate flowers are 20-25 times more numerous than pistillate flowers, but produce less nectar. The campanulate calyx is covered in white pubescence and bears, 5 free sepals; each sepal is linear-lanceolate and 0.5-2 cm long. The yellow to orange corolla is tubular, at least 5 cm long and broad, 5-parted with reflexed petals that are ovate, apically obtuse, and marginally rugose. The fruits are berries. The seed is endospermous.

39. *Achyranthes aspera* L.

Family: Amaranthaceae

Habit: Herb

Useful Parts: Leaves and Root

Uses: It is used in the treatment of asthma, in facilitating delivery, bleeding, bronchitis, debility, dropsy, cold, colic, cough, dog bite, snake bite, scorpion bite, dysentery, earache, headache, leukoderma, renal complications, pneumonia, and skin diseases

Description: It is a wild, perennial, erect herb. Stem is herbaceous but woody below, erect, branched, cylindrical, solid, angular, hairy, longitudinally striated, nodes and internodes are prominent, green but violet or pink at nodes. Leaves are ramal and cauline, simple, exstipulate, opposite decussate, petiolate, ovate or obovate, entire, acute or acuminate, hairy all over, unicostate reticulate. A spike with reflexed flowers arranged on long peduncle. Flowers are Bracteate, bracteolate, bracteoles two, shorter than perianth, dry, membranous and persistent, sessile, complete, hermaphrodite, actinomorphic, pentamerous, hypogynous, small, spinescent, green. Bracts ovate, persistent. Perianth made up of 5 tepals, polyphyllous, imbricate or quincuncial, green, ovate to oblong, persistent. Androecium made up of 10 stamens, out of which 5 are fertile and 5 are scale-like, fimbriated, sterile staminodes, both alternating with each other, fertile stamens are antiphyllous, monadelphous, filaments slightly fused at the base, dithecous, dorsifixed or versatile, introrse. Gynoecium is bicarpellary, syncarpous, superior, unilocular, ovule one, basal placentation, style single and filiform, stigma capitate. Fruit is oblong utricle. Seeds are endospermic.

41. *Cardiospermum halicacabum* L.

Family: Sapindaceae

Habit: Climber

Useful Parts: Leaves, Fruit, Seed

Uses: This plant is used for the treatment of rheumatism, abdominal pain, orchitis, dropsy, lumbago, skin diseases, cough, nervous disorders, and hyperthermia.

Description: It is a woody annual, many-branched vine with bi-fid (forked) axillary tendrils that are used for climbing. Leaves are alternate and twice ternately compound. Leaflets bear toothed margins, are lanceolate in shape, 2-4cm in length, 1-2cm wide, and faintly pubescent with pinnate venation. Irregular flowers are borne in panicles. Each flower bears four sepals, two large and two small, four whitish petals 4mm long, and eight stamens. Petaloid appendages are at the base of each flower. The 3-celled ovary bears one ovule per cell. Flowering time from July to August. Pollination is by entomophilly. Fruit is capsule. Seed is endospermous.

42. *Physalis peruviana* L.

Family: Solanaceae

Habit: Herb

Useful Parts: Fruits and Leaves

Uses: It is used to cure cancer, malaria, hepatitis, asthma, dermatitis and rheumatism.

Description: The Cape gooseberry is a perennial plant but is commonly grown as an annual in temperate climates. The simple velvety leaves are roughly heart-shaped and usually have entire (non-toothed) margins. The creamy yellow flowers are solitary and somewhat bell-shaped with five fused petals, each with a brown or purple spot at the base. Like the tomatillo, to which it is related, the plant is noted for the inflated bag like calyx (fused sepals) that encloses a fleshy orange berry.

43. *Physalis angulata* L.

Family: Solanaceae

Habit: Herb

Useful Parts: Leaves and Fruit

Uses: The plant extract is used in the treatment of dermatitis, hepatitis, rheumatism, malaria and asthma

Description: The plant is erect. It forms a small bush, abundantly branched, which can reach 90 cm high. The plant has a taproot system. The stem is hollow and polygonal. It is totally glabrous. The leaves are simple and alternate. They are carried by petiole, 3 to 5 cm long. The lamina is oval to elliptical, apiculate at the apex and bottom attenuated in sharp corner. It is 7 to 12 cm long and 3 to 6 cm wide. The margin is sinuous and irregularly serrated, provided some short, white hairs. The leaf blade is marked with 4 to 6 pairs of pinnate venations. Both sides are glabrous, although some short white hairs are present along the veins of the lower face. The flowers are solitary and axillary, located at the intersection of the branches of the plant. The flowers are supported by a glabrous peduncle, 7 to 10 mm long. The calyx, 3mm long, is composed of 5 sepals fused at the section of the base and ending in 5 triangular tines. Campanulate corolla, consisting of 5 fused petals. It is 7 to 8 mm large and creamy white with a purple spot at the base of the petals. 5 stamens are inserted into the corolla tube alternating with petals. The ovary has 2 loculus with many ovules. The fruit is berry. The seeds are flat and lenticular.

44. *Solanum pimpinellifolium* L.

Family: Solanaceae

Habit: Herb

Useful Parts: Fruits and Roots.

Uses: The fruits have medicinal uses as well. It can be used as first aid treatment for burns, scalds, and sunburn. It is also used in the treatment of rheumatism and headaches. Root decoction is ingested to relieve toothache.

Description: Annual or biennial herbs, undergoing secondary growth at the base; branches extremely slender and vining, extending up to 3m from centre. Stem erect initially, later procumbent or decumbent, sparsely pubescent or nearly glabrous.

Leaves are imparipinnately compound. Calyx 0.4-1.0 cm in diameter, pubescent with long and short, simple, uniseriate trichomes; tube less than 0.5 mm; lobes to 5 mm, linear, the apex acute. Corolla 1.6-3 cm in diameter, bright yellow; tube minute, the corolla often divided almost to the base; lobes 0.7-1.2 x 0.2-0.5 cm, four times as long as wide, narrowly lanceolate, strongly reflexed at anthesis. Staminal column 6-8 mm, narrowly cone shaped; filaments 1-2.5 mm; anthers 3.5-5 mm, the sterile tip approximately half the total anther length. Ovary conical, minutely glandular-villous; style 7-10 mm, usually exerted from the staminal column; stigma minute.

45. *Solanum linnaeanum* Happer

Family: Solanaceae

Habit: Shrub

Useful Part: Fruits.

Uses: The leaves are aphrodisiac, ophthalmic. They are used as a treatment for insomnia and for stopping excessive menses. An infusion of the leaves is used as an eye wash for cleaning the eyes.

Description: This perennial plant has attractive flowers each with five purple petals and small yellow stigmas, but it also has vicious thorns. The previous year's fruits can sometimes be seen ripening while the new season's flowers are attracting insect pollinators. The fruits are typically 3cm to 5cm across and look very much like unripe tomatoes; they appear in late summer; turning gradually from green to bright yellow and eventually black. The fruits are deadly poisonous and should never be eaten.

46. *Solanum trilobatum* L.

Family: Solanaceae

Habit: Slender Prickly Shrub

Useful Parts: Leaves, Petioles and Fruit

Uses: The leaves, petioles, and fruit of Solanum Trilobatum are utilised for many reasons in south India, which is economically significant. In some areas of India, it is also consumed to cure gastric complaints.

Description: It is a slender prickly scrambling shrub with prickles being curved, broad-based, yellowish and numerous along the stems, otherwise almost glabrous. Leaves are rounded-ovate in outline, obtusely lobed, 2-7 cm long, slightly stellate, with a few prickles along the petiole and midrib. Inflorescences are extra-axillary, peduncle short, 3-9-flowered; pedicels widely divergent. Calyx 3 mm long, with narrow teeth, sparsely stellate. Corolla is deeply lobed, stellate-pubescent outside, purple, reflexed.

47. *Capsicum annuum* L.

Family: Solanaceae

Habit: A small perennial herb

Useful Parts: Leaves and Fruits

Uses: Capsicum (Capsicum annuum), also known as cayenne pepper, has been used orally for upset stomach, toothache, poor circulation, fever, hyperlipidaemia, and heart disease prevention.

Description: The 'Anaheim Chili' has fruits about 7 inches long, 1½ inches in diameter, slightly tapered, stem end usually without pronounced shoulder but often wrinkled or folded. Flavour is mildly pungent as compared with other chili varieties. Anaheims take about 115 days to green mature and 150 days to red ripe and are also called 'California Chili.' 'College No. 9 Chili,' also called 'New Mexico 9,' has fruits about 5 inches long, 1¾ inches in diameter, tapered and pointed, shoulders sloping and usually smooth. These are less pungent than 'Mexican Chili,' but slightly more pungent than 'Anaheim,' with about the same maturity period as for 'Anaheim.' Mexican, or "native" chili has fruits about 3 inches by 1½ inches, somewhat conical, tapering to a blunt point. Pods generally have a deep shoulder at the stem and are often furrowed and wrinkled. Mexican chilis are the most pungent of the large-fruited

chilis and strains are widely grown in the Southwest, and in central and northern Mexico, where they are preferred for earliness

48. *Datura matel* L.

Family: Solanaceae

Habit: A perennial shrub

Useful Parts: leaves, fruits, flowers, stem or roots

Uses: The bitter narcotic plant relieves pain and encourages the healing process. The seeds of the plant are medicinally the most active. Externally, the plant is used as a poultice in treating fistulas, abscesses wounds and severe neuralgia.

Description: *Datura metel* is a shrub-like annual (zone 5–7) or short-lived, shrubby perennial (zone 8–10), commonly known in Europe as Indian thorn apple, Hindu Datura, or metel and in the United States as devil's trumpet or angel's trumpet. *Datura metel* is naturalized in all the warmer countries of the world.

49. *Solanum incanum* L.

Family: Solanaceae

Habit: An erect prickly shrub

Useful Part: Fruits

Uses: *Solanum incanum* is the traditional medicinal plants widely used to treat various types of ailments like sore throat, stomach-ache, head-ache, painful menstruation, liver pain, malaria, hypertension, stomach problem, asthma, diabetes, common cold and pain caused by onchocerciasis, pneumonia and rheumatism**.**

Description: Erect or spreading shrub up to 3 m tall, occasionally a small tree; stems and leaves with stellate hairs and pale yellow to brown prickles, up to 1 cm long. Leaves alternate, simple; stipules absent; petiole 0.5–8.5 cm long; blade almost round to lanceolate, 1–30 cm × 1–17 cm, base rounded, truncate or cordate, often unequal, apex acute or obtuse, margin entire to pinnately lobed, densely hairy. Inflorescence a 2–15-flowered cyme, inserted above the leaf axil. Flowers bisexual or functionally

male, nodding or pendent, regular, (4–)5–7(–9)-merous; pedicel 0.5–4 cm long; calyx campanulate, lobes up to 1.5 cm long, enlarging and splitting in fruit; corolla campanulate to rotate, 1–4.5 cm in diameter, with ovate or broadly triangular lobes, blue, pink, purple or violet, rarely white; stamens inserted near the base of the corolla tube and alternating with corolla lobes, filaments short, anthers slender; ovary superior, 2(–4)-celled, style up to 15 mm long, densely hairy. Fruit a globose or depressed globose, occasionally ovoid-ellipsoid berry 2.5–3.5 cm × 2–3 cm, yellow, orange or brown when ripe, many-seeded. Seeds lentil-shaped to almost kidney-shaped, up to 3.5 mm × 3 mm, pale yellow to brown. Seedling with epigeal germination.

50. *Solanum melongena* L.

Family: Solanaceae

Habit: Perennial Shrub

Useful Part: Fruit

Uses: Various plant parts are used in decoction, as powder or ash for curing ailments such as diabetes, cholera, bronchitis, dysuria, dysentery, otitis, toothache, skin infections, asthenia and haemorrhoids. Eggplant is also ascribed narcotic, anti-asthmatic and anti-rheumatic properties.

Description: Eggplant is an annual or short-lived perennial plant that is very sensitive to cold temperatures. It grows fastest when the temperature ranges between 70 and 85 degrees. It produces an edible shiny glossy fruit. The plant may grow 2 to 4 feet tall and is multi-branched. The leaves and stems have star-shaped hairs, and the small violet flowers are also star-shaped. Eggplant or Aubergine is a member of the Solanaceae or nightshade family which also includes tomatoes, potatoes, and peppers.

51. *Cyanthillium cinereum (L.) H. Rob.*

Family: Asteraceae

Habit:

Useful Part: Leaves

Uses: The juice of *Cyathillium cinereum* is given to children with urinary incontinence. The leaves are eaten as a potherb. A decoction of it is also given in diarrhea, stomachache and for cough and colic. Leaves have antibiotic properties.

Description: *Cyathillium cinereum* is an erect herb, 20 to 80 cm high, slightly branched and covered with fine grey hairs. The stem is finely striated. The leaves are alternate, simple. They are elliptical, attenuate base in corner and covered with a greyish hair. The flowers are grouped in small purplish heads, assembled in a loose inflorescence. The fruits are carriers of small tufts of white hairs.

52. *Passiflora foediax* L.

Family: Passifloraceae

Habit: Climber

Useful Parts: Fruit and Leaves

Uses: It is used to relieve sleeping problems as well as used in the treatment of itching and coughs.

Description: The stems have an unpleasant odour and vary in hairiness from almost hairless to having a sparse or dense covering of white, yellow or golden-brown sticky hairs. At the base of each leaf stalk there is a tendril and a 1 cm long threadlike appendage covered in sticky glands. The leaves most often have three rounded or pointed lobes, but sometimes they can be entire or five-lobed. These leaves are alternately arranged along the stems and borne on stalks 1-6 cm long. They are hairy on both surfaces, with the hairs along their margins often being sticky. The flowers vary from pinkish to white or purplish in colour and are borne singly in the leaf forks on stalks 2-4.5 cm long. They are surrounded by three deeply-divided bracts that are densely covered in large sticky (i.e. glandular) hairs. Each flower has five sepals and five petals. They also have five stamens, with anthers 4-5 mm long, and an ovary topped with three style tipped with prominent stigmas. Flowering occurs mainly during autumn, winter and spring. The fruit are dry berries partially enclosed by the persistent, deeply-divided, sticky bracts. These fruits are somewhat hairy and turn from green to yellow or orange in colour as they mature**.**

54. *Leucas aspera* (Willd.) Link

Family: Lamiaceae

Habit: annual herb

Uses: The entire plant is also used as an insecticide and indicated in traditional medicine for cough, cold, painful swelling and chronic skin eruption, wound healing.

Useful Parts: Leaves, Root and Stem

Description: *Leucas aspera* is an annual, branched, herb erecting to a height of 15-60 cm with stout and hispid acutely quadrangular stem and branches. Leaves are sub-sessile or shortly petiolate, linear or linearly lanceolate, obtuse, pubescent up to 8.0 cm long and 1.25 cm broad, with entire or crenate margin; petiole 2.5-6 mm long; flowers white, sessile small, in dense terminal or axillary whorls; bracts 6 mm long, linear, acute, bristle-tipped, ciliate with long slender hairs; calyx variable, tubular, 8-13 mm long; tube curved, contracted above the nutlets, the lower half usually glabrous and membranous, the upper half ribbed and hispid.

55. *Ocimum tenuiflorum*

Family: Lamiaceae

Habit: Perennial Herb

Useful Parts: Leaves, Roots and Seeds

Uses: It heals the bacterial borne diseases as well as against Aspergillosis and other fungal borne diseases

Description: Holy basil is an erect, many-branched subshrub, 30–60 cm (12–24 in) tall with hairy stems. Leaves are green or purple; they are simple, petiole, with an ovate blade up to 5 cm (2 in) long, which usually has a slightly toothed margin; they are strongly scented and have a decussate phyllotaxy. The purplish flowers are placed in close whorls on elongated racemes.

56. *Mesosphaerum suaveolens*

Family: Lamiaceae

Habit: Shrub

Useful Parts: Leaves and Flower

Uses: Kuntze is a species widely used traditionally in the treatment of ailments, such as stomach pain, haemorrhoids, cough, ulcer, liver disease, fever, influenza, nasal congestion, and inflammation.

Description: Shrubs, to 1.5 m high; stem obtusely 4-angular, thinly hairy. Leaves ovate, acute, hispid below, glabrate above; petiole to 5 cm long. Flowers in clusters of 1-12; calyx tube 8 mm long, tubular, 10-ribbed, glandular hairy, teeth spinulose, 4 mm long; corolla 5 mm long, lobes short, glabrous inside, blue. Nutlets 4 x 2.5 mm, compressed, with a ridge on dorsal surface, pubescent, deep brown, mucilaginous when wet.

57. *Ocimum kilimandscharium* Baker ex Gurke

Family: Lamiaceae

Habit: Shrub

Useful Parts: Leaves and flowers

Uses: Traditionally, extracts of *Ocimum kilimandscharium* were used to alleviate many ailments in East Africa including treatment of colds, coughs, abdominal pains, measles, diarrhea, insect repellent, particularly against mosquitoes and storage pest control.

Description: *Ocimum kilimandscharium* Guerke (Syn. *Ocimum camphora* Guerke) belongs to family Lamiaceae. It is a native of Kenya and distributed in East Africa, India, Thailand, Uganda and Tanzania. It is extensively grown in the Tropics. In India it is cultivated on a small scale especially in West Bengal, Assam, Tamil Nadu, Karnataka, Kerala and Dehradun. Commonly the plant is called as camphor Basil, African blue basil and in Ayurveda as Karpura Tulasi**.** Morphologically, it is a perennial aromatic evergreen undershrub with pubescent branchlets having pale green leaves which are glandular, ovate or oblong in shape, base is acute, deeply serrated, pubescent on both surfaces, oppositely arranged and about 3-7 cm in length including

petioles which are 4 to 12 mm long, 1 to 2.5 cm wide; indumentum of long white hairs or sometimes glabrous above; petiole 4-10 mm. Stems are brownish green, round-quadrangular, much branched, woody with epidermis sometimes peeling off in strips below, arising from a large woody rootstock, with white glandular hairs, becoming denser in the inflorescence-axis, with sparse sessile glands.

58. *Merremia disseca* (Jacq.) Hallier f.

Family: Convolvulaceae

Habit: Climber

Useful Parts: Leaves and Stem

Uses: An infusion of the leaves is taken as a sedative in the treatment of chest complaints A cold infusion is a remedy for giddiness, snake bites or intoxication. A hot infusion is taken to relieve urinary infection A decoction of the whole plant, used as a wash, is an effective remedy for scabies and itch. A poultice of crushed fresh leaves is applied as a resolutive and sedative for treating inflammations

Description: Vines; the stems herbaceous, sparsely hirsute to glabrous. Leaves alternate, palmately divided almost to the base, the 7-9 lobes sinuate-dentate, usually glabrous, the entire leaf suborbicular in outline. Inflorescence axillary, 1-to few-flowered, cymose, the peduncles 5-10 cm long, hirsute, glabrescent on the upper portion. Pedicels 1.5-2 cm long, thickened toward the apex, glabrous. Sepals oblong, 18-25 mm long, mucronate, glabrous. Corolla 3-4.5 cm long, white with a purple centre, broadly campanulate. Fruits capsular, depressed-globose, 1-2 cm in diameter, subtended and partially surrounded by the accrescent calyx; seeds black, sub rotund, glabrous.

59. *Merremia aegyptia* (L.) Urb.

Family: Convolvulaceae

Habit: Annual climber

Uses: It is used to treat deobstruent, diuretic, rheumatism, neuralgia, cancerous wounds, migraine, purgative, snake bites, ulcer

Useful Part: Stem and Leaves**.**

Description: A robust annual twiner occurring throughout the Region and widespread in the Tropics. Horses will not graze it; other stock in Senegal may or may not take it. The dried leaves are applied in Nigeria as a dressing for burns. The stems are used in Senegal and probably elsewhere as ties.

60. *lpomoea aquatic* Forsk.

Family: Convolvulaceae

Habit: Perennial herb

Useful Parts: Leaves and Flowers

Uses: It is used to treat piles, and nosebleeds, as an anthelmintic, and to treat high blood pressure.

Description: Water Morning Glory is a semi-aquatic tropical plant grown as a leaf vegetable. Its precise natural distribution is unknown due to extensive cultivation, with the species found throughout the tropical and subtropical regions of the world. Water Morning Glory grows in water or on moist soil. Its stems are 2-3 m or more long, hollow, allowing them to float, and these root at the nodes. The leaves vary from sagittate (typical) to lanceolate, 5-15 cm long and 2-8 cm broad. The flowers are trumpet-shaped, 3-5 cm diameter, usually white in colour, with a purple center. It is most commonly grown in East and Southeast Asia. Because it flourishes naturally in waterways and does not require much if any care, it is used extensively in Malay and Chinese cuisine.

61. *Ipomoea quamoclit* L

Family: Convolvulaceae

Habit: A beautiful annual twiner with slender stems.

Useful Parts: Flowers and Stem

Uses: In the Philippines, leaves are used as poultices for bleeding hemorrhoids. Crushed leaves used for carbuncles. Seeds reportedly used as laxative by the Sino-Annamites.

Description: *I. quamoclit* is a herbaceous, twining vine growing up to 3–10 feet (0.91–3.05 m) tall. The leaves are 1–4 inches (25–102 mm) long, deeply lobed (nearly pinnate), with 9-19 lobes on each side of the leaf. The flowers are 1–2 inches (25–51 mm) long and 1 inch (25 mm) in diameter, trumpet-shaped with five points, and can be red, pink or white. *I. quamoclit* is herbaceous, twining vine growing up to 3–10 feet (0.91–3.05 m) tall. The leaves are 1–4 inches (25–102 mm) long, deeply lobed (nearly pinnate), with 9-19 lobes on each side of the leaf. The flowers are 1–2 inches (25–51 mm) long and 1 inch (25 mm) in diameter, trumpet-shaped with five points, and can be red, pink or white.

62. *Ipomoea* carnea Jacq.

Family: Convolvulaceae

Habit: Evergreen Shrub

Useful Parts: Leaves and Flower

Uses: The plant possesses anti-bacterial, anti-fungal, anti-oxidant, anti-cancer, anti-convulsant, immunomodulatory, anti-diabetic, hepatoprotective, anti-inflammatory, anxiolytic, sedative and wound healing activities. However, some toxicological effects have been also reported**.**

Description: *Ipomoea carnea*, the pink morning glory, is a species of morning glory that grows as a bush. This flowering plant has heart-shaped leaves that are a rich green and 6–9 inches (15–23 cm) long. It can be easily grown from seeds. Erect (subsp. *fistulosa*) or climbing (subsp. *carnea*) undershrub to 4 m, often growing in clumps, stems stout, hollow, canescent when young, becoming glabrous. Leaves petiolate, 8 - 20 (- 30) x 3 - 10 (- 12) cm, ovate or elongate-ovate-deltoid, base cordate to sub truncate with rounded auricles, apex acuminate to long-acuminate, both surfaces grey-canescent when young, veins prominent abaxially; petioles 3 - 8 cm. Inflorescence of long-pedunculate axillary, somewhat compact cymes.

63. *Barleria volkensii* Lindau

Family: Acanthaceae

Habit: Herb

Uses*:* Used to reduce inflammation caused by insect bites, snake bites, boils, and rheumatism.

Useful Parts: Leaves and Stem.

Description: It grows as a shrub 60 –100 cm tall. The leaves are dark green on the upper surface and pale green on the lower surface. They are elliptic to narrowly ovate. The flowers are about 5 cm long, funnel-shaped in violet, pink, or white color. The fruits are about 1.5 cm long ellipsoid capsules. They become glabrous and glossy at maturity. It has spikelet inflorescence. In dense spikes, bracteoles prominent, linear. Calyx of 4 unequal sepals white. Corolla blue, funnel-shaped, lobes 5, unequal. Stamens 4, only 2 fertile, disk cup-like. Ovary bicelled, 4 ovuled. The fruit is capsule.

64. *Hygrophila auriculata* (K. Schum.)

Family: Acanthaceae

Habit: A herb growing in wet places

Useful Part: Leaves and Flowers

Uses: Kokilaksha, as it is known in sanskrit, was extensively used in Ayurvedic system of medicine for various ailments like rheumatism, inflammation, jaundice, hepatic obstruction, pain

Description: Marsh Barbel is a stout aquatic perennial herb, 1-2 m high. Erect unbranched stems are hairy near swollen nodes. Densely hairy, lance-like, stalkless leaves, 10-15 cm long, occur in whorls of 6 at each node on the stem. Straight, yellow, 4 cm long spines are present in the axil of each leaf. Flowers occur in 4 pairs at each node. The 3 cm long purple-blue flowers are 2-lipped - the upper lip is 2-lobed and the lower one 3-lobed with lengthwise folds. Flowers open in opposite pairs. Flowering: October

65. *Dicliptera paniculata* (Forssk.) I. Darbush

Family: Acanthaceae

Habit: Erect herb

Useful Parts: Stems and leaves

Uses: It heals the bacterial borne diseases as well as against Aspergillosis and other fungal borne diseases.

Description: Panicled Foldwing is an erect herb, 0.6-1.2 m tall. Young shoots are usually 4-sided; adult shoots 6-sided, white spreading bristle-hairy. Ovate leaves opposite, equal and unequal; leaf-stalk 3-5 mm. Smaller ones 0.8-1.2 x 3-5 mm, larger ones 3-4.5 x 1.5-2 cm, densely hairy and prominently so on veins. Flowers are borne at branch-ends or in leaf-axils, with leaves forming a large lax panicle. Pink flowers, to 1 cm, 2-lipped - lower lip spreading, upper lip erect. Stamens 2; filaments distinct, to 5 mm, white hairy. Panicled Foldwing is found in India, Myanmar and Tropical Africa. It is also found in the Himalayas and Western Ghats, at altitudes of 600-2200 m. Flowering: October-February.

66. *Pedalium mure* L.

Family: Pedaliaceae

Habit: Herb

Useful Parts: Leaves and Fruit

Uses: Based on traditional healers, the plant Pedalium murex L. was used for the dissolution and prevention of kidney stone formation. Further, it is used for the treating ailments like incontinence of urine, gonorrhea, anti-bilious agent, dysuria and control white discharge.

Description: A diffuse annual, much branched, spreading, succulent, glandular, up to 60 cm tall. Roots similar to turmeric in colour. Leaves simple, opposite, ovate or oblong-obovate, 1-4.5 cm long, 0.5-3 cm broad, truncate or obtuse, irregularly and coarsely crenate-serrate, glabrous above, minutely scaly below, petiole-l-4 cm.

Flowers 1.5-2 cm across; pedicel 1-2 mm long, increasing up to 4 mm in fruit. Calyx c.2 mm long; teeth linear, scaly outside, persistent. Petals connate into a broad tube, 1-3 cm long; lobes obtuse. Stamens included, 0.5-1 cm long; filaments dilated, glandular hairy at the base; anthers kidney shaped. Fruit indehiscent, abruptly narrowed at the base and with a patent spreading spine at each basal corner of the broader part, 1-1.8 cm long, 0.5-1 cm broad, spine 2-4 mm long. Seeds 2 or 1 per locule, oblong.

67. *Ludwigia octovalvis* Linn.

Family: Onagraceae

Habit: Herbaceous shrub

Useful Part: Leaves

Uses: It is commonly consumed as a health drink and traditionally used for treating various ailments such as dysentery, diarrhea, diabetes and headache.

Description: A herbaceous shrub, it can grow up to 2m tall and has a branched growth form. Leaves are hirsute, long and narrow with sunken venation and a reddish to pale midrib. The stems grow up to 1cm in diameter and are hirsute and ribbed. The flower is composed of 4 obovate petals (0.2 - 0.4 cm long) arranged in a cross-like pattern. The tip of the petal is sometimes folded down, forming a large notch-like indentation. The petals have distinct sunken venation which creates a rippled surface. The fruit is a capsule about 2 to 4.5cm long with persistent sepals at the apex of the fruit, it splits into 8 linear lobes, releasing wedge-shaped seeds dispersed by water.

68. *Ludwigia palustris* (L.) Elliott.

Family: Onagraceae

Habit: Perennial herb

Useful Parts: Leaves, Roots and Seeds

Uses: Used in the treatment of phthisis (pulmonary tuberculosis), asthma and chronic coughs.

Description: Water Purslane is a mat-forming wet area plant in the primrose family. It is found growing in low water situations along small ponds or streams or in muddy areas. It is also used in aquariums. The stems are succulent and often red in color. The inconspicuous flowers occur mid-summer to fall. This plant may be seed near a pond or grown in rain or water gardens. When growing in mud the plant sprawls along the ground with just the branch tips upright. In the water, branches ascend towards the surface with just the tips out of water. Both in the water and out, roots are continuously forming at the nodes, forming mats. It can be considered weedy but can also be a stabilizing plant on muddy banks, and is used to filter and take in toxins in bioswales and ditches.

69. *Heliotropium indicum* L.

Family: Boraginaceae

Habit: Perennial herb

Uses: The leaf juice is used to treat the stings and boils of scorpions and insect bites. On the other hand, the boiled juice with castor oil is used to treat mad dog bite infections, rheumatism, ulcer, venereal disease, fever, sore throat, and sores in the rectum.

Useful Part: Leaves

Description: Indian heliotrope is an annual, erect, branched plant that can grow to a height of about 15–50 cm (5.9–19.7 in). It has a hairy stem, bearing alternating ovate to oblong-ovate leaves. It has small white or purple flowers with a green calyx; five stamens borne on a corolla tube; a terminal style; and a four-lobed ovary.

70. *Stemodia durantifolia* (Linn.)

Family: Scrophulariaceae

Habit: Shrub

Useful Part: Leaves

Uses: It is used to prepare many medicinal drugs.

Description: Stemodia durantifolia is a perennial herbaceous plant to 100 cm (39 in) tall, with glandular-hairy herbage. The branching habit is both basal and axillary. The leaves are lanceolate and subsessile, and toothed along their edges. The terminal inflorescence is an ascending, spike-like structure. The bracts are equal to or slightly longer in length to the flowers or more or less equal to the length of the sepals. The flowers have a violet corolla. The sepals measure equal to or longer than half of the length of the corolla.

72. *Corchorus trilocularis* L.

Family: Tiliaceae

Habit: Annual herb

Uses: It is used to treat syphilis

Useful Part: Leaves

Description: Wild Jute is a branched annual herb, up to 1 m tall, usually erect, sometimes found prostrate due to browsing by cattle. Young branches are purplish, sparsely hairy. Leaves are oblong to lance shaped, up to 12 × 3.5 cm, hairless or hairy, particularly on the veins. Margins are toothed with a long bristle on the 2 lowermost teeth. Flowers are borne in in 1-3 flowered leaf-opposed clusters. Flowers are yellow, with sepals narrowly lance shaped, as long as the petals. Petals are 4-5, 5-7 mm long, 2-2.5 mm wide, obovate tapering to a short ciliate claw. Stamens are many. Fruit is a slender more or less erect, cylindric, many-seeded capsule, straight or slightly curved, up to 7 cm long, 3-4-angled with a rough surface. The species name trilocularis comes from the three-chambered ovary. Young tender leaves are cooked and eaten.

73. *Cleoserrata speciosa* (Raf.) Iltis

Family: Cleomaceae

Habit: Perennial herb

Useful Part: Leaves

Uses: It was used for many therapeutic potentials and it was used to make many medicines based on its anti-inflammatory properties.

Description: Showy Spider Flower is a herbaceous plant resembling Wild Spider Flower, but with larger, more showy flowers. It was introduced as an ornamental plant, but has escaped from cultivation, and can be seen growing wild. It is an annual herb, growing up to 1.5 m in height, with hairless stem and leaves. Leaves are palmately compound, alternately arranged, elliptic leaflets 2-16 cm long. It can flower any time after maturity. Flowers are many, borne in an erect, showy raceme, at the end of the stem. Each flower is subtended by a small leaf-like bract. Flower has 4 free, inverted-lance shaped, pink to white petals, 2.8-3.8 cm long. Flowers have 6 stamens, on filaments that are 3-6 cm long! Fruit is a linear capsule up to 8 cm long, on a long stalk. The plant is propagated by seed.

74. *Cleome viscosa* L.

Family: Capparidaceae

Habit: Herb

Useful Parts: Stem, Leaves and Flowers

Uses: The leaves are diaphoretic, rubefacient and vesicant. They are used as an external application to wounds and ulcers. The juice of the leaves has been used to relieve earache. The seeds are anthelmintic, carminative, rubefacient and vesicant.

Description: Calyx composed of 4 sepals, polysepalous, valvate, sepals arranged in two whorls of two each. Corolla composed of 4 petals, polypetalous, clawed, imbricate, yellow coloured. Androecium made up of many stamens (12-24), polyandrous, dithecous, basifixed, introrse, filament long and anther lobes curved. Gynoecium bicarpellary, syncarpous, superior, single locule with many ovules, parietal placentation, gynophore is present but very short, style short, stigma sticky and capitate.

76. *Commelina communis* L.

Family: Commelinaceae

Habit: Annual herb

Useful Parts: Leaves, Roots and Flowers.

Uses: It is used to heal swelling, treatment of urinary tract infection and respiratory tract infections, diarrhea, enteritis, and hemorrhoids. The plant has also been used in fever, malaria, insect, bug bites, rheumatoid arthritis, gonorrhea, influenza, and bladder infection.

Description: It is an annual herb. The stem is erect. The leaves are sessile. The flowers are arranged on inflorescences called cincinni, which are also called Scorpioid cymes. This is a form of a monochasial where the lateral branches arise alternately. The cincinni are subtended by a spathe, a modified leaf. There are three fertile stamens, meaning they are on the lower part of the flower, and three infertile stamens, meaning they are on the upper part of the flower. The fruit is a dehiscent, ellipsoid capsule with two locules each containing two seeds. The capsule is glabrous, brown, measures 4.5–8 mm (0.18–0.31 in) long, and dehisces into two valves.

77. *Argemone mexicana* L.

Family: Papaveraceae

Habit: Annual herb

Useful Parts: Leaves, Flowers, Stem and Roots.

Uses: It is used to treat tumours, warts, skin diseases, inflammations, rheumatism, jaundice, leprosy, microbial infections, and malaria**.**

Description: Annual, prickly herb with yellow latex and branched tap root. Stem is erect, branched, woody at the base, solid, cylindrical, spinous, contains yellow latex. Leaves are ramal and cauline, exstipulate, alternate, simple, subsessile, semi - amplexicaul, margin lobed and spinous, apex acute, unicostate reticulate venation, both the surfaces are covered with many spines. It shows Solitary terminal inflorescence. Flowers are ebracteate, pedicellate, complete, actinomorphic, hermaphrodite, hypogynous, yellow in colour. Calyx made up of 2 sepals, polysepalous, spinous, caducous, bear a clear horn like outgrowth at their apex,

twisted or imbricate, inferior and green. Corolla made up of 6 petals, arranged in two whorls of three each (3+3), polypetalous, twisted or imbricate, yellow, each petal is obovate, caduceus, inferior. Androecium made up of numerous stamens, polyandrous, arranged in many whorls, filament long, slender and yellow, anther dithecous, basifixed and extrorse, dehiscence longitudinal. Gynoecium tetra to Hexa carpellary, syncarpous, superior, ovary covered with prickles, unilocular, many anatropous ovules on each placenta, parietal placentation, style reduced with 4 to 6 stigma lobes red. Fruit is prickly capsule.

78. *Impatiens balsamina* L.

Family: Geraniaceae

Habit: Annual herb

Useful Parts: Flowers and stem

Uses: It is used for the treatment for the treatment of rheumatism, isthmus, generalized pain, fractures, inflammation of the nails, scurvy, carbuncles, dysentery, bruises, foot diseases.

Description: Garden Balsam is most common Balsam grown as a garden plant in India. It is an annual plant growing up to 20-75 cm tall, with a thick, but soft stem. The leaves are spirally-arranged, 2.5-9 cm long and 1-2.5 cm broad, with a deeply toothed margin. The flowers are pink, red, mauve, lilac, or white, and 2.5-5 cm diameter. They are pollinated by bees and other insects, and also by nectar-feeding birds. Flower-stalks are up to 1-5 cm long. Lateral sepals are about 2-3 mm long, ovate, sparsely ciliate, lower sepal conical, spur 1-2 cm long, curved inwards. The ripe seed capsules explode when touched, inspiring the genus name Impatiens. Garden Balsam is native to Western Ghats India and SE Asia.

79. *Phyllanthus polygonoides* Nutt.

Family: Euphorbiaceae

Habit: Herb

Useful Parts: Leaves and flower

Uses: In Indian ayurvedic medicine, various herbaceous *Phyllanthus* species are prescribed for jaundice, gonorrhea and diabetes as well as for making poultices for skin problems. Infusions from young shoots are used to treat chronic dysentery.

Description: *Phyllanthus polygonoides*, known as smartweed leaf-flower or knotweed leaf flower, is an herbaceous perennial plant in the family Phyllanthaceae. It grows from 10 to 50 centimeters in height. It is native to the United States northern and central Mexico. Smartweed leaf-flower grows in a variety of habitats throughout its range, including grasslands, shrublands), and glades in forests. It is often associated with limestone and calcareous soils. It is morphologically similar to the closely related species, pinewoods dainties (*Phyllanthus liebmannianus*).

80. *Turnera ulmifolia* L

Family: Turneraceae

Habit: Herb

Useful Parts: leaves and Flower

Description: *Turnera ulmifolia* grows erect, with dark toothed leaves and small, yellow-orange flowers, and is often found as a weed growing on roadsides. These yellow flowers bloom around 6:00 am and wilt around 11:30 AM. Life span for flower is around 6 hours. These plants can survive on minimum water and grow on walls, cement blocks, and rocks. Tawny Coster (*Acraea terpsicore*) butterfly larvae feed on these plants. This plant is commonly misidentified with the closely related *T. diffusa* in horticultural commerce, causing it to be often misrepresented as "Damiana.

82. *Jasminum sambac* (L.) Aiton

Family: Oleaceae

Habit: Shrub

Useful Parts: Leaves

Uses: Traditionally, *Jasminum sambac* has been used to treat dysmenorrhea, amenorrhea, ringworm, leprosy, skin diseases and also as an analgesic, antidepressant, anti-inflammatory, antiseptic, aphrodisiac, sedative, expectorant.

Description: An evergreen vine or shrub, Jasminum sambac can grow to a height of 1.6 to 9.8 feet. Due to auto polyploidy, natural hybridization, and spontaneous mutation, the species is exceedingly varied. The majority of Jasminum sambac plants grown for commercial purposes do not generate seeds; instead, cuttings, layering, marcotting, and other asexual propagation techniques are used to propagate the plant.

83. *Ammannia coccinea* Rottb.

Family: Lythraceae

Habit: Perennial herb

Useful Parts: Leaves and Roots

Uses: It was used to treat scabies, ringworm, parasitic skin infections, common cold, typhoid, strangury, spinal disease, gastroenteropathy and aphrodisiac**.**

Description: It was a perennial herb. Stem is erect and branched. Leaves are opposite, the pairs set at right angles to the ones above and below, ¾ to 4 inches (to 10 cm) long, ¼ to about ½ inch (to 15 mm) wide, pointed to blunt at the tip, toothless, hairless, stalkless, mostly green or tinged red, the lowest leaves more oblong-elliptic becoming lance-linear up the stem. flowers are stalkless or nearly so, the stalks less than .5 mm long. Each flower is ¼ inch across or less, has 4 (rarely 8) pink to lavender, wavy petals 2 to 4 mm long with 4 to 8 light yellow tipped stamens surrounding a stout green style in the center. The calyx cupping the flower is cylindrical, less than ¼ inch (3 to 5 mm) long, the sepal edges fused and broadly triangular at the tip. Fruit is a round capsule 4 to 6 mm (to ¼ inch) in diameter, as long as or slightly longer than the persistent calyx at maturity. Capsules have 4 chambers and turn red as seed ripens.

84. *Scleromitrion diffusum* (Willd.) R. J. Wang

Family: Rubiaceae

Habit: Herb

Useful Parts: Flower and Leaves

Uses: Spreading Diamond Flower is being actively studied for its role in treating cancer.

Description: Annual herbs. Stems erect to semi-protrate. Stipules 1-2.5 mm long. Leaves linear lanceolate, 30-50 x 3-6 mm. Petiole 0-1 mm. Inflorescence an axillary cyme. Corolla tube1-1.5 mm long. Capsule sub globose, 2-2.5 x 2-3 mm. Scleromitrion diffusum - Spreading Diamond Flower. Spreading Diamond Flower is a slender annual herb, rising up to prostrate, up to 50 cm tall; stems slightly flattened to round or young stems sometimes 4-angled, sparsely to densely finely velvet-hairy.

85. *Oldenlandia umbellate* L

Family: Rubiaceae

Habit: Herb

Useful Parts: Leaves

Uses: It is used in combination with other herbs for the treatment of hepatitis, snake bites and tumours of the liver, lung, stomach, and rectum

Description: Small diffuse herbs; stem terete, sparsely pubescent towards nodal regions. Leaves 1-1.5 x c. 1 cm, narrow, linear; stipules sheathing, shortly pectinate on the margins. Flowers axillary, solitary or binate, sessile to shortly pedicelled. Calyx tube sub globose, 1.5-2 cm; lobes triangular, to 0.1 cm long. Corolla white; tube 0.1-0.15 cm long; lobes 0.05 cm long. Style shortly exserted; stigma papillate. Capsule ovoid, 0.2 cm long, laterally compressed, dehiscing loculicidally at tip; seeds minute, angular, reticulate.

87. *Boehmeria cylindrica* (L.)

Family: Urticaceae

Habit: Annual perennial herb

Useful Parts: Whole Plant

Uses: Used to cure cancer

Description: Strongly toothed margins are evident on the 4 to 18 cm long, simple, opposite, rough, lance to oval-shaped leaves of this tall perennial herb (to 1.5 m). It has a single stem with tiny hair***s.***

88. *Digitaria sanguinalis* (L.)

Family: Poaceae

Habit: Annual herb

Useful Part: Leaves

Uses: It is used to treat anti-ulcer, anti-helminitic, anti-inflammatory, anti-diabetic, anti-depressant activity.

Description: Digitaria sanguinalis is a sparse, tufted decumbent annual grass with weak and spreading, hollow stems, to 1 m long or more, usually rooting at the lower nodes, the erect portions up to 60 cm tall. The sheaths are shorter than the internodes and pubescent. The ligule is a thin, truncate membrane, 1-2 mm long, hairless and irregularly dissected. The leaf blades are soft and flat, 5-10 mm wide, pubescent on one or both surfaces to nearly glabrous. The inflorescence consists of 4-9 erect or spreading, digitate racemes at the apex of the stem and sometimes below. The rachis is narrowly winged. Spikelet, 3 mm long, are born in pairs, one stalked and the other stalkless; they are green or purple-tinged. Fruit is glume.

89. *Orysa sativa* L.

Family: Poaceae

Habit: Annual to woody perennial herb

Useful Parts: Stem and Leaves

Uses: rice grain is applied to the skin to treat boils, sores, swellings, and blemishes, and sticky rice is used in remedies for stomach upsets, heartburn, and indigestion. Brown rice extracts have been used as remedies for breast and stomach cancer, as well as indigestion, nausea, and diarrhoea.

Description: Basal sterile florets similar; barren; without significant palea. Lemma of lower sterile floret lanceolate; 2-3 mm long; 0.25 (-0.5) length of spikelet; membranous; without lateral veins; emarginate. Lemma of upper sterile floret lanceolate; 2-3 mm long; 1 length of lower sterile floret; membranous. Fertile lemma elliptic; laterally compressed; 8-11 mm long; coriaceous; keeled; 5 -veined. Lemma midvein ciliate; hairy above.

90. *Brachiaria brizantha (Hochst. Ex A. Rich)*

Family: Poaceae

Description: It is a tufted perennial grass, usually 60-120 cm high (up to 200 cm), with deep roots (down to 2 m) and short rhizomes. It has stout, erect or slightly decumbent culms and bright green leaves. Inflorescence is a panicle consisting of 2-16 racemes, 4-20 cm long. Spikelets are usually on a single row, elliptical, 4-6 mm long with a sub-apical fringe of long purplish hairs.

PLATE 1: LIST OF PLANTS BELONGING TO EUPHORBIACEAE

Ricinus communis

Jatropha gossypiifolia L.

Chrozophora brocchiana (Vis.) Schweinf.

Euphorbia peplus L

Euphorbia serpens Kunth

PLATE 2: LIST OF PLANTS BELONGING TO EUPHORBIACEAE AND VEBENACEAE

Croton bonplandianus Baill.

Euphorbia prostrate Aiton

Stachytarpheta jamaicens (L.)

Lantana camara L.

Lantana aculeate L.

PLATE 3: LIST OF PLANTS BELONGING TO VERBENACEAE, LEGUMINOSAE AND CAESALPINIACEAE

Phyla nodiflora (L.) Greene

Acacia tortilis (Forssk).

Senna occidentalis (L.) Link

Galactia spp.

Cassia angustifolia Vahl.

PLATE 4: LIST OF PLANTS BELONGING TO RUBIACEAE AND ASCLEPIADACEAE

Oldenlandia umbellate, L.

Pergularia daemia (Forsskal) Chiov.

Oxystelam esculentum R. Br.

Calotropis procera (Aiton) Dryand.

Calotropis gigantea (L.) Dryand.

PLATE 5: LIST OF PLANTS BELONGING TO RHAMNACEAE, COMPOSITAE, ASTERACEAE AND MALVACEAE

Ziziphus spina-christi (L.) Desf.

Tridax procumbens (L.)

Parthenium hysterophorus L.

Stoebe plumose L.

Sida spinosa L.

PLATE 6: LIST OF PLANTS BELONGING TO MALVACEAE AND ZYGOPHYLLACEAE

Sida cordifolia, L.

Abelmoschus ficulneus (L.)

Hibiscus calyphyllus Cav.

Abutilon indicum, (L) Sweet.

Tribulus terrestris, L.

PLATE 7: LIST OF PLANTS BELONGING TO CUCURBITACEAE

*Luffa cylindrica (*L.) M. Roem

Citrullus lanatus (Thunb.) Matsum & Nakai

Momordica cymbalaria (Hook & Fenzl)

Cucumis melo L.

Momordica charantia L.

PLATE 8: LIST OF PLANTS BELONGING TO CUCURBITACEAE AND AMARANTHACEAE

Citrullus colocynthis (L.) Schrad

Mukia maderaspatana, W.

Lagenaria siceraria (Molina) Standley.

Cucurbita maxima Duch

Achyranthes aspera L..

PLATE 9: LIST OF PLANTS BELONGING TO AMARANTHACEAE, SAPINDACEAE AND SOLANACEAE

Celosia cristata L

Cardiospermum halicacabum L.

Physalis peruviana L

Physalis angulata L.

Solanum pimpinellifolium L.

PLATE 10: LIST OF PLANTS BELONGING TO SOLANACEAE

Solanum linnaeanum Happer

Solanum trilobatum, L.

Capsicum annuum L.

Datura matel L.

Solanum incanum L.

PLATE 11: LIST OF PLANTS BELONGING TO SOLANACEAE, TURNERACEAE, PASSIFLORACEAE AND LAMIACEAE

Solanum melongena L.

Turnera ulmifolia L.

Passiflora foediax L

Passiflora edulis Sims

Leucas aspera (Willd.) Link

PLATE 12: LIST OF PLANTS BELONGING TO LAMIACEAE AND CONVOLVULACEAE

Ocimum tenuiflorum

Mesosphaerum suaveolens

Ocimum kilimandscharium Baker ex Gurke

Merremia disseca (Jacq.) Hallier f.

Merremia aegyptia (L.) Urb.

PLATE 13: LIST OF PLANTS BELONGING TO CONVOLVULACEAE AND ACANTHACEAE

lpomoea aquatic, Forsk.

Ipomoea quamoclit L.

Ipomoea carnea, Jacq.

Barleria volkensii Lindau

Hygrophila auriculata (K. Schum.)

PLATE 14: LIST OF PLANTS BELONGING TO ACANTHACEAE, PEDALIACEAE, ONAGRACEAE AND BORAGINACEAE

Dicliptera paniculata (Forssk.) I. Darbush

Pedalium mure,, L.

Ludwigia octovalvis Linn.

Ludwigia palustris (L.) Elliott.

Heliotropium indicum L.

PLATE 15: LIST OF PLANTS BELONGING TO SCROPHULARIACEAE, CYPEACEAE, COMPOSITAE, CLEOMACEAE AND CAPPARIDACEAE

Stemodia durantifolia (Linn.)

Schoenoplectus tabernaemontani (C.C. Gmel.)

Corchorus trilocularis, L.

Cleoserrata speciosa (Raf.) Iltis

Cleome viscosa L.

PLATE 16: LIST OF PLANTS BELONGING TO CAPPARIDACEAE, COMMELINACEAE, PAPAVERACEAE, GERANIACEAE AND EUPHOBIACEAE

Cleome gynandra L.

Commelina communis L.

Argemone mexicana L.

Impatiens balsamina L.

Phyllanthus polygonoides Nutt.

PLATE 17: LIST OF PLANTS BELONGING TO COMPOSITAE, ARALIACEAE, OLEACEAE, LYTHRACEAE AND RUBIACEAE

Canthillium cinereum

Hydrocotyle spp.

Jasminum sambac (L.) Aiton

Ammannia coccinea Rottb.

Scleromitrion diffusum (Willd.) R.J.Wang

PLATE 18: LIST OF PLANTS BELONGING TO RUBIACEAE, URTICACEAE AND POACEAE

Brachiaria brizantha (Hochst. ex A. Rich.)

Citrus aurantiifolia (Christm.) Swingle

Boehmeria cylindrica (L.)

Digitaria sanguinalis (L.)

Orysa sativa L.

CONCLUSION

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The present study carried out in Athimarapatti village and has documented 90 plant species belonging to 34 families. The floristic and ethnobotanical survey of the village concludes that rural people of Thoothukudi district possess rich ethnobotanical knowledge about treatment of various diseases but this traditional knowledge is declining due to rapid urbanization and migration of rural people. Thus, it becomes necessary the documentation of ethnobotanical knowledge. This study also suggested that documentation of flora with various uses provides raw material for pharmacological investigation and resulting in discovery of drugs. This documentation of flora and ethnobotanical knowledge provides a catalog of useful plants of the village and will serve as a physical record for both student community those who are studying botany as well as for village people.

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FLORAL DIVERSITY OF ATHIMARAPATTI IN THOOTHUKUDI

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MASTER OF SCIENCE IN BOTANY

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CERTIFICATE

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CONTENT

INTRODUCTION

INTRODUCTION

Biodiversity is referred as the composition of living organisms including plants, animals and microbes inhabiting the terrestrial, aquatic and other habitats of a region or a country. United Nations Environment Programme (UNEP) described biological diversity as the variety and variability of all animals, plants and microorganisms and the ecological complexes of which they are a part. Biodiversity is very indeed to the functioning of various ecosystems. Each species in the world plays a unique role within an ecosystem and every species is dependent on other species for food, shelter and other purposes. Even the loss of a single species can make an impact on ecosystem as well human life.

India is a country rich in a wide variety of Biodiversity. Most of the plants that grow here serve a high medicinal purpose **(Kalisdha *et al.*, 2013).** In India, from the pre-medieval age, holds a possession over natural medicine. Traditional uses of floristic diversity are the foremost vital part of indigenous information system, which is widely practiced by human populations all across the world. This knowledge has been transferred orally from generation to generation.

Floristic studies are nothing but exploring the region by identifying plants and grouping them, data collection of plants present in the region and counting of them. These studies have gifted mankind with the knowledge of plants which are economically important and of high medicinal value. The angiosperms fulfill major needs of human life such as foods, medicines, shelter, cloths and other luxuries **(Kamini Dubey and Sweta Prakash, 2021).** Tamil Nadu is one among the twenty-eight states of India is found with rich floral diversity region. **Irwin *et al.* (2014)**

revealed that there are about 5674 angiospermic species in Tamil Nadu state, which include 212 taxa that are endemic to the state.

Mankind has been utilizing plants for food and medicinal purpose. Therefore, various aspects of plants towards health, economic value, sustainable utility, their conservation, floral assessment and documentation are necessary. India is a rich centre of plants diversity. All types of flora and fauna are elements of biodiversity and influenced by various climatic conditions such as temperature, availability of moisture in the form of humidity and precipitation and variation in physiographical conditions – soil, altitude, slope, etc. **(Suba *et al*., 2014; Sukumaran and Parthiban, 2014).**

The great wealth of biological diversity in tropical regions is due to the myriad environmental conditions existing there. Interest in biodiversity has recently increased in response to the damage caused to ecosystems by anthropogenic activities **(Merigot *et al*., 2007).** It is well known that floristic composition is determined by environmental factors. However, the composition influences biodiversity patterns at regional scales and further reflects both anthropogenic and natural disturbances. Therefore, floristic characteristics and biodiversity patterns are often influenced by environmental factors and anthropogenic disturbances **(Liu *et al*., 2009).**

Conservation of biodiversity is essential for the proper functioning of ecosystems and for the maintenance of the environmental services they provide **(Lopez-del Toro *et al*., 2010).** However, high rates of tropical deforestation and habitat destruction frequently cause the local extinction of plant and animal species. Flowering plants are by far the most numerous, diverse and successful extant plant group containing well over 90%of all land plant species alive today **(Simpson, 2006).**

In India, dicots are represented by 2282 genera and 12750 species whereas, monocots are represented by 702 genera and 4250 species. Dicots accounts for c.75% flowering plants in terms of both genera and species **(Brummit, 1992).**

Documenting basic patterns of biodiversity is fundamental for prioritizing areas for conservation and management action **(Villasenor *et al*., 2007).** Taxonomic inventorying is essential for exploring unexplored genetic resources. Floristic study of smaller areas is more important in comparison with that of larger areas. Smaller areas can be explored thoroughly with critical field observations to find out any additional species which might have been left out from earlier studies. Due to fragmentation of habitats and ecosystems because of various developmental activities many earlier reported taxa might have become extinct, rare and endangered. Study of the existence, size, structure and locality of such taxa deserves more importance.

Floristic explorations and taxonomic studies can provide efficient and convenient information about the nomenclature, distribution, ecology, utility of various plant species, and thus about an ecosystem. Taxonomy is an integrated and perhaps, intuitive science of identifying, naming and classifying plants. This may be considered as the oldest of sciences in the world, as the primitive man had to distinguish the plants that he can eat safely, from those which are poisonous and inedible. Floristic studies help us to understand the basic aspects of biology such as speciation, isolation, endemism and evolution.

A scientific study on medicinal and floristic diversity of the hill group along with traditional knowledge on medicinal plants, which are used by native people in healthcare system, is warranted. Therefore, the importance and scope of the present study is imperative for the conservation and sustainable utilization of plant resources and traditional knowledge for the betterment of future generation without prompting

the use of present generation. The primary goal in the present work is to document the angiosperms associated with Athimarapatti, Thoothukudi District, Tamil Nadu, India that may lead to formulating steps to conservation of natural resources associated with it. The study is also aimed to explore the diversity indices and economic importance of the plants from the study area.

SCOPE AND OBJECTIVE

Natural resources survey like floristic study plays an important role in the economic development of developing country like India. Vegetation is the most precious gift and nature has provided to us as meeting all kinds of essential requirements of the humans in the form of food, fodder, fuel, medicine, timber, resins, and oil, etc. Scientists have been trying to bring to the attention of people and their governments the importance of maintaining biodiversity of planet Earth and of carrying out our daily lives in a fashion that ensures our offspring will inherit a cleaner, greener, more ecologically sustainable world. Floristic studies acquire increasing importance in recent years in response to the need of developing and under developing countries to assess their plant wealth. Keeping all the benefits of floristic studies in mind, the present study is an attempt to realize the target the Mullakadu with the following objectives.

- To make through Floristic survey of flowering plants.
- To provide short description of the dominant families in Dicotyledons (Polypetalae, Gamopetalae, Monochlamydeae) and in monocotyledons
- To provide a list of medicinal plants along with their usage and also to document environmentally, economically useful and ornamental plants.
- To evaluate the present status of the study site and suggest methods for conservation.

REVIEW OF LITERATURE

Vascular plants, an important component of vegetation, must be constantly monitored and managed inorder to direct successional processes towards maintaining species and habitat diversity **(Attua and Pabi 2013; Naidu and Kumar, 2016). Swamy *et al.* (2000)** and **Kothandaraman and Sundarapandian (2017)** explored the flora of low-level evergreen forests of the Western Ghats of Kanyakumari district. **Meena (2007)** listed an exhaustive floristic spectrum of Marundhuvalmalai, a tropical scrub jungle of Western Ghats of Kanyakumari district. Intensive explorations of **Kabeer and Nair (2009)** led to the excellent description of 447 species and 19 intraspecific grass taxa under 136 genera and 19 tribes in Tamilnadu, which form the major collections from Kanyakumari district.

Neelamegam *et al.* (2015) made a baseline survey to evaluate the status, structure, composition, diversity and utilization of home garden plants in two villages one located in a rural area (Arumanalloor) near Nagercoil and the other (Konam) a town municipality of Nagercoil, Kanyakumari district and to understand the impact of socio-economic conditions of households on home garden structure. **Sreekanth *et al.* (2012)** studied the genetic diversity of the teak (*Tectona grandis*) population in Southern Western Ghats. **Karuppusamy and his co-workers (2013a)** enlisted some of the endemic *Caralluma* sp. of Kanyakumari district in their monumentalwork *Caralluma in India*.

Arul *et al.* (2013) reported a new distributional record of *Stylosanthes scabra* from Alamparai hills of Kanyakumari district. While preparing the flora

Plants of the Western Ghats, **Ganeshiah *et al.* (2013)** listed many rare and endemic floristic elements from Western Ghats of Kanyakumari district. Recently, **Karuppusamy (2013b)** and his research team reported the evergreen species *Cleidion nitidum*, so far thought to endemic to Andaman and Nicobar Islands, from Thadagamalai in Kanyakumari district.

Sporadic floristic explorations of Kanyakumari district were made by several researchers recently. **Anderson (2006)** studied the floristic wealth of Adakkadu hills and **Rajeshwari (2009), Anusha (2010), Jani (2011)** and **Vasanthi (2011)** collected the herbaceous plants of Pechiparai, Nagercoil, Marunthuvalmalai and Thovalai, respectively. **Jebaselvi (2010)** surveyed the herbaceous flora of teak and rubber plantations of the district. **Jashbar and Brintha (2011)** botanized the plants of Mothiramalai and Ponmalai hillock, respectively, while the vascular plant diversity of Udayagiri fort of Kanyakumari district was explored by **Sukumaran and Parthiban (2014). Kensa *et al.* (2014)** gave a delightful description of the ornamental flora of an urban environment of Kanyakumari district.

Gajurel *et al.* (2007) studied wild edible plants of Dibang Biosphere Reserve, Arunaehal Pradesh. It has been found that 150 wild plant species used for various purposes of which 80 species are wild edible. **Ghosh and Das (2007)** studied Plants of ethnobotanical significance for the tea Garden Workers in Terai and Duars of Darjeeling. They reported 133 dicotyledonous and 33 monocotyledonous and 4 pteridophytic plants of ethnobotanical significance. Ethnobotanical observations in Pench National Park (Maharashtra) were recorded by **Chaturvedi and Panhi Kumar (2007).**

Sharma *et al.* (2000) reported 694 species which are endemic to Maharashtra. According to annual report of BSI (2001), out of about 17500 species, 5285 species of 140 genera of angiosperm is endemic to India. **Yadav and Sardesai (2002)** compiled 340 endemic plants in the flora of Kolhapur District. **Pawar *et al.* (2006)** studied floristic composition and quantitative assessment of plant resources of Western Ghats from Dhule and Nandurbar District. Quantitative assessment of plant resources from Pamer and Sangamner Tahsils of Ahmednagar District was studied by **Shendage *et al.* (2007).**

Endemic species and their potential value of plant resources are gradually shaping the future of several regions. However, since past hundred years the trends observed in the loss of plant diversity is our biggest concern. Despite all the efforts made to conserve plant diversity, the situation today is still very alarming. **Takhtajan (1986)** stated that India to be cradle for the origin of cultivated plants. On similar lines, **Vavilov (1992) and recently Khoury *et al*. (2016)** stated that India to be prime pocket for the origin of pulses. The rich diversity of edible plants has been valued and consumed consciously by the Indian villagers. According to **Nayar (1996),** 5725 species under 147 genera of angiosperms i.e., 33% are endemic. Documentation on wild flora of Indian region carried out by **Singh and Arora (1978)** showed that around 250 species occur in the western and nearly 300 species occur in the eastern Himalayan ranges. This diversity includes 258species of edible fruits, 121 species of green leafy vegetables, 37 species of roots and tubers, and 20 species of edible flower buds. Amongst the 214 threatened species occurring in the Himalayas, nearly 37 are exploited, being medicinal herbs of commercial value and need priority action for conservation **(Arora and Nayar, 1984).**

Urbanization is one of the major reasons for the destruction of the natural vegetation. This ongoing growth of urban agglomerations leads to far-reaching changes in biodiversity, including the loss of forests and other natural areas **(Kumar *et al*., 2010; Von der Lippe and Kowarik, 2008).** Urbanized areas can also harbour a high number of threatened species **(Sodhi *et al*., 2010).** The destruction of tropical forests and habitats causes global biodiversity degradation **(Singh, 1998).**

Floristic diversity study helps us to evaluate the floristic wealth and its prospects of an area. Floristic inventories help us perceive biological aspects such as endemism, evolution, speciation and isolation **(Elourard *et al*., 1997).** It helps to assess the country's plant wealth, distribution and status **(Ellis, 1987).** There are 15000 species of flowering plants in India belonging to 2250 genera and 315 families, contributing 6% of World's flowering plants **(Nayar, 1977).** The present number of flowering plants in India is found to be 18666 species under 2991 genera and 251 families **(Mao, 2019).**

The total number of angiosperms reported from the Dibrugarh District presents 462 species belonging to 334 genera and 106 families show a similar agreement with other floristic studies previously carried out in different regions of India. An inventory of the native flowering plants in the East Siang district of Arunachal Pradesh presented 508 taxa belonging to 348 genera and 102 families **(Taram *et al.*, 2020).** Higher plant diversity in East Kameng district of Arunachal Prdesh reported 215 species of higher plants belonging to 165 genera and 70 families **(Tag *et al.*, 2012).** Floristic diversity assessment and vegetation analysis of Upper Siang district of Northeast India reported 1003 taxa belonging to 110 families and 529 genera **(Choudhary *et al.*, 2012).** A study of Angiospermic diversity in the Bhadrak

region of Odissa showed 383 species including 262 native and 121 non-native species belonging to 282 genera under 93 families **(Panda *et al.*, 2020).**

Biodiversity reflects variety and variability within and among living organisms, their associations and habitat-oriented ecological complexes. India is one of the 12 "megadiversity" countries in the world. Mankind has been utilizing plants for food and medicinal purpose. Therefore, various aspects of plants towards health, economic value, sustainable utility, their conservation, floral assessment and documentation are necessary. India is a rich centre of plants diversity. All types of flora and fauna are elements of biodiversity and influenced by various climatic conditions such as temperature, availability of moisture in the form of humidity and precipitation, and variation in physiographical conditions – soil, altitude, slope, etc. **(Arul *et al.*,2013; Ghildiyal and Juyal, 2012; Suba *et al.*, 2014; Sukumaran and Parthiban, 2014).**

The great wealth of biological diversity in tropical regions is due to the myriad environmental conditions existing there. Interest in biodiversity has recently increased in response to the damage caused to ecosystems by anthropogenic activities **(Merigot *et al.* 2007).** It is well known that floristic composition is determined by environmental factors; however, the composition influences biodiversity patterns at regional scales and further reflects both anthropogenic and natural disturbances. Therefore, floristic characteristics and biodiversity patterns are often influenced by environmental factors and anthropogenic disturbances **(Liu *et al.*, 2009).**

Conservation of biodiversity is essential for the proper functioning of ecosystems and for the maintenance of the environmental services they provide **(Lopez-del Toro *et al.*, 2010).** However, high rates of tropical deforestation and habitat destruction frequently cause the local extinction of plant and animal species.

Flowering plants are by far the most numerous, diverse and successful extant plant group containing well over 90%of all land plant species alive today **(Simpson, 2006)**. In India dicots are represented by 2282 genera and 12750 species whereas monocots are represented by 702 genera and 4250 species. Dicots accounts for c.75% flowering plants in terms of both genera and species **(Brummit, 1992).**

Natural resources survey like floristic study plays an important role in the economic development of developing country like India. Vegetation is the most precious gift, nature has provided to us as meeting all kinds of essential requirements of the humans in the form of food, fodder, fuel, medicine, timber, resins, and oil etc. **(Gaur, 1999).** Plant communities play a pivotal role in sustainable management by maintaining biodiversity and conserving the environment **(Farooquee and Saxena, 1996).** Floristic diversity refers to the variety and variability of plants in a given region. It refers to the number of types or taxa in a given region or group. It can be measured at any level from overall global diversity to ecosystem, community, species, populations, individuals and even to genes within a single individual.

Floristic study and diversity assessments are necessary to understand the present diversity status and conservation of biodiversity. Floristic study is a necessary prerequisite for much fundamental research in tropical community ecology, such as modeling patterns of species diversity or understanding species distributions **(Phillips *et al.*, 2003).** Floristic studies acquire increasing importance in recent years in response to the need of developing and under developing countries to assess their plant wealth **(Vediya and Kharadi, 2011).** Floristic explorations and taxonomic studies can provide efficient and convenient information about the nomenclature, distribution, ecology, utility of various plant species, and thus about an ecosystem.

Taxonomy is an integrated and perhaps, intuitive science of identifying, naming and classifying plants. This may be considered as the oldest of sciences in the world, as the primitive man had to distinguish the plants that he can eat safely, from those which are poisonous and inedible. The gravity of the situation is so severe due to variety of reasons, the foremost being habitat destruction at an alarming rate leading to loss of biodiversity essential for the sustenance of life on earth. Thus, conservation of biodiversity has gained prime consideration all over the world since the Earth, Summit at Rio de Janeiro in 1992. Floristic studies help us to understand the basic aspects of biology such as speciation, isolation, endemism and evolution. Flora of any area is not fixed up. It changes from time to time. Various ecological factors, mostly biotic, change the floristic components. The total number of species may be changed; dominant species may be replaced with other species; the floristic composition, i.e.; family: genus: species ratio may be changed.

Documentation of existing green spaces of an area is important to determine existing resources and to set target for future improvements. Floras serve as the basic reference of the plant biota of an area; they are critical tools that serve botanists, conservationists, ecologists, foresters, gardeners, agronomists, researchers, and the general public. The botanical exploration of an area and writing a flora to summarize that information was seen as a basic societal need leading to the discovery of economically valuable information. Among the plants inhabiting the earth, the angiosperms or flowering plants are one of the major groups of extant seed plants and arguably the most diverse major extant plant group on the planet with at least 2,60,000 living species classified in 453 families **(Judd *et al.*, 2002).**

In India, the angiosperms comprising a total of 17,817 species constitutes 38.15% of floral diversity of the entire country. According to current estimates, the Indian flora represents nearly 12% of the global floral diversity (excluding viruses). The significance of the Indian flora is further evidenced by the number of species of wild relatives of crop plants in different regions of the country. Since time immemorial man has been using plant resources to meet his daily requirements. The immense diversity which occur in flowering plant provides economic benefits as they are important sources of food, fodder, timber, medicine, fibres, spices, dyes and tannins, beverages, gums and resins etc. **(Singh *et al.*, 2002).**

MATERIALS AND METHODS

PLANT COLLECTION AND IDENTIFICATION

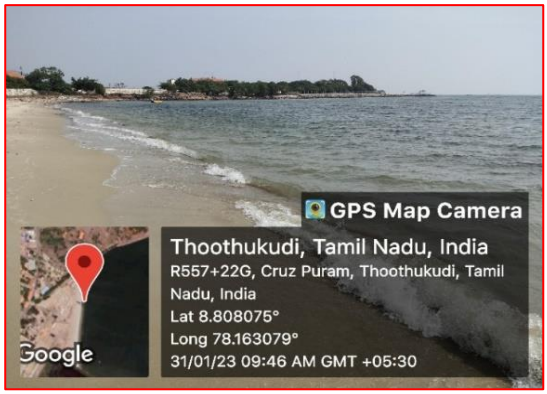
From the month of December, 2022 to March 2023, Athimarapatti village was regularly visited and every plant was photographed and collected in plastic bags for identification. With the aid of Flora of the Presidency of Madras, the plants were identified. In addition to data on taxonomic place (family), vernacular name, common name, life form, and folk medicinal uses, the species entries were supplemented. The life type was graded according to the system suggested by Raunkiær into herbs, shrubs, grasses and trees (annual, biennial or perennial).

ETHNOBOTANICAL SURVEY

The ethnobotanical study of the village includes the testing of knowledge about the usage of common medicinal plants. Based on the literature review, about 14 categories of common ailments like Kidney Disease (KD, Skin disease (SD), Hair problem (HP), Stomach problem (SP), Respiratory disease (RD), Cold (C), Cough (CO), Diuretic (D), Tooth ache (TA), Head ache (HA), Fever (F), Body heat (BH), Insect Bite (IB) and Menstural issues (MI) were selected for the present survey. A questionnaire was prepared using Google forms to collect the ethnobotanical applications of the plants in the village. The link is https://forms.gle/CrHEk1xTXWTosWBH7. The link was then circulated among the residents of the chosen village. Choices were given to each respondent to select the medicinal plants and their uses. There were up to 124 members participated in the survey.

PLATE 1: SAMPLING STATIONS OF THOOTHUKUDI COAST

STATION I: VEMBAR



STATION II: THERESPURAM

STATION III: PUNNAKAYAL

QUANTITATIVE ANALYSIS: (Umair *et al.*, 2017)

The ethnobotanical data was analyzed using different quantitative indices including Informant Consensus Factor (ICF), Use value (UV), Relative frequency citation (RFC), Fidelity level (FL), Relative popularity level (RPL), Rank order priority (ROP). Data were reported in proportions and percentages.

INFORMANT CONSENSUS FACTOR (ICF).

ICF value describes informants' consensus on the medicinal plant consumption species, and evaluates variability in mode of utilization against reported diseases. Before calculating ICF value, ailments are broadly categorized into different categories. The maximum ICF value i.e. close to 1 indicates that well known species are used by a large proportion of local communities due to their authenticity regarding diseases. However, low ICF index close to 0 specifies that the informants use this species randomly to treat reported diseases. The ICF value was calculated using the formula as described earlier.

$$ICF = \frac{N_{ur} - N_t}{N_{ur} - 1}$$

Where,

"Nur " is the total number of use reports for each disease category

"Nt" indicates the number of species used in said category.

USE VALUE (UV)

Use value (UV) determines the relative importance on uses of plant species. It is calculated using the following formula as follows

$$UV = \frac{\sum U_i}{N}$$

Where,

"UV" indicates use value of individual species,

"U" is the number of uses recoded for that species and

"N" represents the number of informants who reported that species.

RELATIVE FREQUENCY OF CITATION (RFC)

Relative frequency of citation (RFC) signifies the local importance of each species in a study area. This index is determined by dividing the number of informants citing a useful species (FC) by total number of informants in the survey (N). RFC is calculated by the formula,

$$RFC = \frac{FC}{N} \quad (0 < RFC < 1)$$

FIDELITY LEVEL (FL)

FL is the percentage of informants who mentioned the uses of certain plant species to treat a particular ailment in a study area. The FL index is calculated using formula,

$$FL\ \% = \frac{NP}{N} \times 100$$

Where,

'Np' is the number of informants that claimed a use of certain plant species for a particular disease

'N' is the total number of informants citing the species for any disease.

The maximum FL indicates the frequency and high use of the plant species for treating a particular ailment by the informants of the study area.

RELATIVE POPULARITY LEVEL (RPL)

RPL is the ratio between number of ailments treated by a particular plant species and the total number of informants for any disease. However, plant species with comparable FL may vary in their healing potential. A correction scale is therefore introduced, in which all the encountered plant species are divided into popular and unpopular groups. The relative popularity level (RPL) assumes a value 0 and 1.0, with '1' being complete popularity of a plant for major ailments and '0' no ailments treated by a plant species. When all plant species are frequently used to treat some major ailments, popularity index would be maximum (1.0); then decrease towards zero as the relative popularity of the species diverge away from popular side. For popular plant species, the RPL value is rationally selected to equal unity (i.e. equal to 1), while RPL value is less than 1 for unpopular plant species. The relative popularity level (RPL) of the plant species is calculated and designated as popular or unpopular. The RPL value may be determined for each specific plant in accordance with its exact position on graph.

RANK ORDER PRIORITY (ROP)

ROP is a correction factor, used for appropriate ranking of the plant species with different FL and RPL values. The ROP is derived from FL; by multiplying RPL and FL values as follows.

ROP = FL x RPL

RESULTS AND DISCUSSION

FLORISTIC ANALYSIS

Flowering plants are the most numerous, diverse and successful extant plant group containing well over 90% of all land plant species alive today **(Simpson, 2006).** In India, dicots are represented by 2282 genera and 12750 species whereas monocots are represented by 702 genera and 4250 species. Dicots accounts for c.75% flowering plants in terms of both genera and species **(Brummit, 1992).** Species richness is measured on samples carefully chosen in a particular area. Such data are important for prioritizing conservation strategies since they allow identification of geographic regions of the world wide exceptional or with very poor diversity **(Krishnamoorthy 2003).**

Floristic studies help us to understand the basic aspects of biology such as speciation, isolation, endemism and evolution. Flora of any area is not fixed up. It changes from time to time. Various ecological factors, mostly biotic, change the floristic components. The total number of species may be changed; dominant species may be replaced with other species; the floristic composition, i.e. family: genus: species ratio may be changed. The present study revealed that the study area is housing more taxa, however, identified only 90 taxa **(Table 1).** These 90 species were found to be dispersed in 32 families in Dicot and 2 families in Monocot, 66 genera in Dicot and 4 genera in Monocot, 86 species in Dicot and 4 species in Monocot **(Fig 1).**

GENERIC AND SPECIES DIVERSITY

Figure 3 and Table 2 show generic and species diversity of 34 families studied. Out of 34 families, Cucurbitaceae and Euphorbiaceae have more generic

diversity compared to other families. 5 genera in Euphorbiaceae, 7 genera in Cucurbitaceae, 4 genera in 3 families such as Asteraceae, Malvaceae and Solanaceae, 3 genera in 7 families, 2 genera in 2 Families and 20 families are monogeneric. Maximum species diversity occurs in the family Solanaceae (9 species), Cucurbitaceae (9 species), Euphorbiaceae (7 species), Malvaceae (6 species) and Convolvulaceae (5 species).

DIVERSITY OF HABIT FORMS

Different life forms of species are shown in **Table 3**. In general, the vegetation is dominated by herbs (56%) followed by Shrubs (25%), Climbers (18%) and Tree (1%). The percentage distribution of habit forms in different class is shown in **Fig.2.**

USE REPORT AND USE CATEGORIES:

Based on the literature review, about 6 categories of common ailments like Skin disease (SD), Hair problem (HP), Cold (C), Head ache (HA), Fever (F), and Menstrual issues (MI) were selected for the present survey and about 23 plant species were given in the survey to note down their usage. Among which, about 11 species were surveyed to cure skin diseases which is followed by head ache and menstrual issues reported by 10 as shown in **Table 4**. Many researchers previously documented the ethnobotanical surveys also stated that, the skin disorder reports higher number of plants **(Khajoei and Khosravi, 2014).** Due to poor dietary conditions and unsafe drinking water, this ailment category is one of the most common problems in the study area and in fact in other parts of the world. Dermatological infections such as scabies, small pox, lesion odour and burns are other prevalent ailments in the study area which are treated with medicinal plants by Malayali ethnic people. Similarly, **Morvin *et al.*, (2014)** reported that medicinal plants for the treatment of dermatological infections had a high prevalence in Kerala, India.

INFORMANT CONSENSUS FACTOR (ICF):

To calculate ICF, the reported ailments were first classified into 6 different disease categories on the basis of their use reports. Among 6 major disease categories, dermatological disorders were dominated with fever with 176 user reports, followed by skin disease and cold with 159 use-reports as mentioned in **Table 4.** Consensus analysis has been used as an important tool for analysing of ethnobotanical data, and it also tells the level of prevalence of diseases in the study area. From the finding it is shown that cold is noted with the highest ICF value of 0.98 which may be due to the water used in the area. This is been followed by the fever with the ICF of 0.95. Similar findings have already been reported by **Ayyanar and Ignacimuthu in 2011**. However, **Kadir *et al.* (2013)** and **Singh *et al.* (2012)** described a greater number of species to treat fever compared to dermatological ailments. The ICF value of different disease categories was ranged from 0 (nervous disorder) to 0.39 (GIT diseases). The ICF values indicated the maximal networking of indigenous people in the sharing of their knowledge on medicinal practices; this is usually the case with traditional healers who treat the most frequently encountered diseases in the study area. These high ICF values indicate reasonable reliability of informants on the use of MP species **(Lin *et al.*, 2002).** Pharmaceutical and phytochemical studies should be undertaken to study whether the use of these herbs is valid.

RELATIVE FREQUENCY OF CITATION (RFC) AND USE VALUE (UV):

The RFC and UV indices were applied to select potential plant species for further pharmacological study and recommendation in drug development. The relative frequency citation (RFC) index authenticates the frequency of citation of a medicinal plant species used for various ailments. The RFC of the reported species ranged from 0.1 to 2.41. Highest RCF value of 2.41 was noted for *Ocimum tenuiflorum* which is

followed by *Solanum trilobatum* and *Tribulus terrestris* with 1.80 and 0.98 RCF value. The positions of these plant species correspond to the fact that they were reported by maximum number of informants, therefore having high frequency of citation (FC). The high values of RFC can be explained by the fact that these plants are the best known and have long been used by the majority of informants, representing a source of reliability. In fact, many biological activity and phytochemical evaluations have been carried out for these plants, and these species are particularly interesting for research in bioactive compounds. The plant species with high RFC should be subjected to pharmacologic, phytochemical, and other biological studies to evaluate and prove their authenticity **(Mukherjee *et al.*, 2012).**

The use value (UV) index demonstrates the relative importance of plant species and families for a population. In the present investigation, the UV of the reported medicinal plant species varied from 0.02 to 0.24 as shown in **Table 5**. The highest UV was observed for *Ocimum tenuiflorum* with 0.24 followed by *Solanum trilobatum* with the value of 0.21. These findings demonstrate the extensive use of above-mentioned species in the treatment of various ailments by local inhabitants/healers and the consciousness of indigenous peoples, which makes such medicinal plants, the first choice to treat a disease **(Jin *et al.*,2011).**

RELATIVE POPULARITY LEVEL (RPL):

Our 86 informants cited 23 plant species for 6 different disease categories. They are given in **Table 5**. *Ocimum tenuiflorum* is considered to be popular plant of that area due to their higher RPL value of 0.058 which is closely followed by *Solanum trilobatum* with the RCF value of 0.047. The high popularity of these plant species might be attributed to their high efficacy and the awareness of indigenous peoples which specifies their use as herbal medicine. This is the first baseline study

on the indigenous knowledge of local peoples regarding the use of popular plant species for a particular ailment. These findings were in consistent with previous studies on the status of medicinal plants among Bedouins of Negev district (Friedman *et al.*,1986) and medicinal plant survey in Palestinian area **(Ali-Shtayeh *et al.*,2000).**

FIDELITY LEVEL (FL):

The fidelity value is an important means to see for which ailment a particular species is more effective. It indicates the informants' choice for each ailment and the potential of the species related to the ailments. The FL of plant species for specific diseases in the present study area is shown in **Table 6**. For each disease category, different plants showed different FL percentage. For skin disease category, among the 11 species cited, *Ricinus communis* and *Ricinus communis* showed the major FL value of 31% which is followed by *Achyranthes aspera* with the FL of 26%. The least value was reported in *Cleome gynandra* 5%. Out of 6 plants used to treat hair problem the highest FL is noted for *Phyla nodiflora* (31%) and least is noted in *Tridax procumbens* (8%). To treat cold *Ocimum tenuiflorum* and *Solanum trilobatum* are greatly used with the highest FL of 70%. To treat head ache and fever, *Ocimum tenuiflorum, Solanum trilobatum* are greatly used with the highest FL of 43%, 37% and 69%, 55% respectively. The menstrual issues of women can be treated using *Solanum nigrum* and *Cucumis melo* with the FL of 25% and 17% respectively. The medicinal plants that are widely used by the people of the Rif have higher FL values than those that are less popular. On the other hand, more medicinal plants that are known as remedies of a single aliment have a 100% FL than those that are used as remedies for more than one type of aliment **(Verma *et al.*, 2007).**

RANK ORDER PRIORITY (ROP):

The Rank order priority (ROP) index is used to rank appropriately the plant species with different FL values. The ROP values are thus obtained are given in **Table 6.** Of the 23 species, two species attained ROP above 50. This is probably due to decreasing popularity of herbal medicines among the local communities of the study area. Based on ROP value *Ocimum tenuiflorum* and *Solanum trilobatum* are widely used species with the RPL of more than 50%. The ROP values reported for medicinal plants used by Bedouins community in Negev district and in Palestinian area were comparable to present findings **(Ali-Shtayeh *et al.*, 2000).**

DESCRIPTION OF PLANTS IDENTIFIED IN THE STUDY AREA (PLATES 1 – 18)

1. *Ricinus communis* L.

Family: Euphorbiaceae

Habit: Shrub

Usefull Parts: Leaves, Root bark, Seeds.

Uses: Methanolic extracts of the leaves of Ricinus communis were used in antimicrobial testing against eight pathogenic bacteria in rats. Antihistimine and anti-inflammatory properties were also found.

Description: *Ricinus communis* can vary greatly in its growth habit and appearance. The variability has been increased by breeders who have selected a range of cultivars for leaf and flower colours, and for oil production. It is a fast-growing, suckering shrub that can reach the size of a small tree, around 12 metres (39 feet), but it is not cold hardy. The glossy leaves are 15–45 centimetres (6–18 inches) long, long-stalked, alternate and palmate with five to twelve deep lobes with coarsely toothed segments. The fruit capsules of some varieties are showier than the flowers. The flowers lack petals and are unisexual (male and female) where both types are

TABLE 1: CHECK LIST OF ANGIOSPERM DIVERSITY IN THE STUDY AREA

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
1.	*Ricinus communis* L.	Castor bean	Euphorbiaceae
2.	*Jatropha gossypiifolia* L.	Bellyache bush	Euphorbiaceae
3.	*Chrozophora brocchiana* (Vis.) Schweinf.	Poison bush	Euphorbiaceae
4.	*Euphorbia peplus* L.	Petty spurge, milk weed	Euphorbiaceae
5.	*Euphorbia serpens* Kunth.	Matted sandmat	Euphorbiaceae
6.	*Croton bonplandianus* Baill.	Aathuppoondu	Euphorbiaceae
7.	*Euphorbia prostrate* Aiton.	Prostrate sandmat	Euphorbiaceae
8.	*Stachytarpheta jamaicens* (L.)	Blue porterweed	Verbenaceae
9.	*Lantana camara* L.	Arch man	Verbenaceae
10.	*Lantana aculeate* L.	Largeleaf lantana	Verbenaceae
11.	*Phyla nodiflora* (L.) Greene	Turkey tangle, Frog fruit	Verbenaceae
12.	*Acacia tortilis* (Forssk).	Umbrella thorn	Leguminosae
13.	*Senna occidentalis* (L.) Link	Antbush, Coffee senna	Leguminosae
14.	*Galactia spp.*	Milk peas	Leguminosae
15.	*Cassia angustifolia* Vahl.	Senna, Indian senna	Caesalpiniaceae
16.	*Pergularia daemia* (Forsskal) Chiov.	Veliparutthi	Asclepiadaceae
17.	*Oxystelam esculentum* R. Br.	Rosy milkweed vine	Asclepiadaceae

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
18.	*Calotropis procera* (Aiton) Dryand.	Apple of sodom	Asclepiadaceae
19.	*Calotropis gigantea* (L.) Dryand.	Bleaching powder	Asclepiadaceae
20.	*Ziziphus spina-christi* (L.) Desf.	Christ's thorn	Rhamnaceae
21.	*Tridax procumbens* (L.)	Coatbuttons, kinarruppacan	Compositae
22.	*Parthenium hysterophorus* L.	Santa Maria feverfew	Asteraceae
23	*Stoebe plumose* L.	Silver stoebe	Asteraceae
24.	*Sida spinosa* L.	Arrow leaf sida	Malvaceae
25	*Sida cordifolia* L.	Heart leaf sida	Malvaceae
26	*Abelmoschus ficulneus* (L.)	White wild musk mallow	Malvaceae
27	*Hibiscus calyphyllus* Cav.	Hibiscus	Malvaceae
28	*Abutilon indicum* (L) Sweet.	Monkey Bush	Malvaceae
29	*Tribulus terrestris* L.	Puncture Vine	Zygophyllaceae
30	*Luffa cylindrica (*L.) M. Roem	Galka, Bath sponge gourd	Cucurbitaceae
31	*Citrullus lanatus* (Thunb.) Matsum & Nakai	Water melon	Cucurbitaceae
32	*Momordica cymbalaria* (Hook & Fenzl)	Little wild gourd	Cucurbitaceae
33	*Cucumis melo* L.	Muskmelon, sweet melon	Cucurbitaceae
34	*Momordica charantia* L.	Balsam pear; Bitter gourd	Cucurbitaceae

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
35	*Citrullus colocynthis* (L.) Schrad	Vine of sodom	Cucurbitaceae
36	*Mukia maderaspatana* W.	Madras pea pumpkin.	Cucurbitaceae
37	*Lagenaria siceraria* (Molina) Standley.	Bottle gourd,	Cucurbitaceae
38	*Cucurbita maxima* Duch.	Winter squash	Cucurbitaceae
39	*Achyranthes aspera* L.	Prickly chaff flower	Amaranthaceae
40	*Celosia cristata* L.	Cockscomb	Amaranthaceae
41	*Cardiospermum halicacabum* L.	Balloon vine	Sapindaceae
42	*Physalis peruviana* L.	Cape gooseberry	Solanaceae
43	*Physalis angulata* L.	Wild tomato, Camapu	Solanaceae
44	*Solanum pimpinellifolium* L.	Currant tomato	Solanaceae
45	*Solanum linnaeanum* Happer	poison bush, poison weed	Solanaceae
46	*Solanum trilobatum*, L.	Alarkapatramu, Thoothuvalai	Solanaceae
47	*Capsicum annuum* L.	Capsicum, chili.	Solanaceae
48	*Datura matel* L.	Devil's trumpet	Solanaceae
49	*Solanum incanum* L.	Thorn apple, bitter apple	Solanaceae
50	*Solanum melongena* L.	Brinjal, eggplant	Solanaceae
51	*Cyanthillium cinereum (L.) H. Rob.*	West Indian holly, sage rose	Asteraceae

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
52	*Passiflora foediax* L.	Running pop	Passifloraceae
53	*Passiflora edulis Sims*	Passion fruit	Passifloraceae
54	*Leucas aspera* (Willd.) Link	Thambai, Kubo	Lamiaceae
55	*Ocimum tenuiflorum*	Holy basil, Sacred basil, Tulsi	Lamiaceae
56	*Mesosphaerum suaveolens*	Pignut, wild spikenard	Lamiaceae
57	*Ocimum kilimandscharium* Baker ex Gurke	Hoary basil, camphor basil.	Lamiaceae
58	*Merremia disseca* (Jacq.) Hallier f.	Alamo vine	Convolvulaceae
59	*Merremia aegyptia* (L.) Urb.	Hairy morning glory	Convolvulaceae
60	*lpomoea aquatic* Forsk.	Water spinach	Convolvulaceae
61	*Ipomoea quamoclit* L.	Cypress vine	Convolvulaceae
62	*Ipomoea* carnea Jacq.	Pink morning glory	Convolvulaceae
63	*Barleria volkensii* Lindau	Philippine violet	Acanthaceae
64	*Hygrophila auriculata* (K. Schum.)	Marsh barbel	Acanthaceae
65	*Dicliptera paniculata* (Forssk.) I. Darbush	Panicled foldwing,	Acanthaceae
66	*Pedalium mure* L.	Crow thorn	Pedaliaceae
67	*Ludwigia octovalvis* Linn.	Mexican primrose willow	Onagraceae

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
68	*Ludwigia palustris* (L.) Elliott.	Marsh primrose willow	Onagraceae
69	*Heliotropium indicum* L.	Indian heliotrope	Boraginaceae
70	*Stemodia durantifolia* (Linn.)	Whitewoolly twintip	Scrophulariaceae
71	*Schoenoplectus tabernaemontani* (C.C. Gmel.)	Grey club-rush, great bulrush	Cyperaceae
72	*Corchorus trilocularis*, L.	Wild jute	Tiliaceae
73	*Cleoserrata speciosa* (Raf.) Iltis	Showy spider flower	Cleomaceae
74	*Cleome viscosa* L.	Asian spider flower	Capparidaceae
75	*Cleome gynandra* L.	Shona cabbage	Capparidaceae
76	*Commelina communis* L.	Day flower	Commelinaceae
77	*Argemone mexicana* L.	Mexican poppy	Papaveraceae
78	*Impatiens balsamina* L.	Balsam, rose balsam	Geraniaceae
79	*Phyllanthus polygonoides* Nutt.	Smartweed leaf-flower	Euphorbiaceae
80	*Turnera ulmifolia* L	West Indian holly, sage rose	Turneraceae
81	*Hydrocotyle spp.*	Marsh penny, dollar weed	Araliaceae
82	*Jasminum sambac* (L.) Aiton	Arabian jasmine	Oleaceae

S.No.	BOTANICAL NAME	COMMON NAME	FAMILY
83	*Ammannia coccinea* Rottb.	Blistering ammannia	Lythraceae
84	*Scleromitrion diffusum* (Willd.) R. J. Wang	Snake needle grass	Rubiaceae.
85	*Oldenlandia umbellate* L.	Chayaver	Rubiaceae
86	*Citrus aurantiifolia* (Christm.) Swingle	Key lime, lime	Rubiaceae
87	*Boehmeria cylindrica* (L.)	False nettle, bog hemp	Urticaceae
88	*Digitaria sanguinalis* (L.)	Hairy crabgrass, finger-grass	Poaceae
89	*Orysa sativa* L.	Rice	Poaceae
90	*Brachiaria brizantha* (Hochst. ex A. Rich.)	Annual beard grass	Poaceae

TABLE 2: OCCURRENCE OF GENERA AND SPECIES WITHIN THE RESPECTIVE PLANT FAMILIES

S.NO.	FAMILIES	NO. OF GENERA	NO. OF SPECIES
01.	Euphorbiaceae	5	7
02.	Verbenaceae	3	4
03.	Leguminosae	3	3
04.	Caesalpiniaceae	1	1
04.	Asclepiadaceae	3	4
05.	Rhamnaceae	1	1
06.	Asteraceae	4	4
07.	Malvaceaae	4	6
08.	Zygophyllaceae	1	1
09.	Cucurbitaceae	7	9
10.	Amaranthaceae	2	2
11.	Sapindaceae	1	1
12.	Solanaceae	4	9
13.	Turneraceae	1	1
14.	Passifloraceae	1	2
15.	Lamiaceae	3	4
16.	Convolvulaceae	2	5

S.NO.	FAMILIES	NO. OF GENERA	NO. OF SPECIES
17	Acanthaceae	3	3
18.	Pedaliaceae	1	1
19.	Onagraceae	1	2
20.	Boraginaceae	1	1
21.	Scrophulariaceae	1	1
22.	Cyperaceae	1	1
23.	Cleomaceae	1	1
24	Capparidaceae	1	2
25.	Commelinaceae	1	1
26.	Papaveraceae	1	1
27.	Geraniaceae	1	1
28.	Turneraceae	1	1
29.	Araliaceae	1	1
30.	Oleaceae	1	1
31.	Lythraceae	1	1
32.	Rubiaceae	3	3
33.	Urticaceae	1	1
34.	Poaceae	3	3

FIGURE 1: DISTRIBUTION OF TAXA IN DIFFERENT CLASSES OF ANGIOSPERM

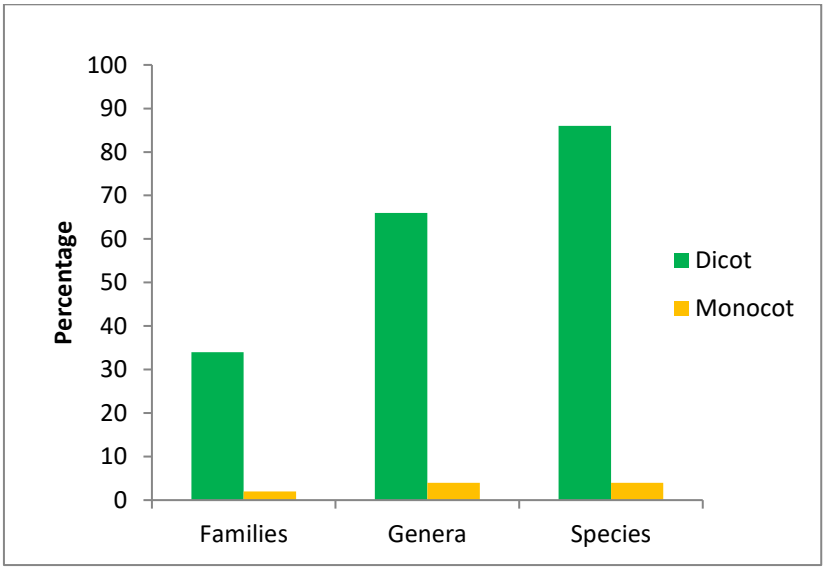


FIGURE 2: PERCENTAGE OF SPECIES OCCURRENCE WITHIN THE IDENTIFIED PLANT HABITS

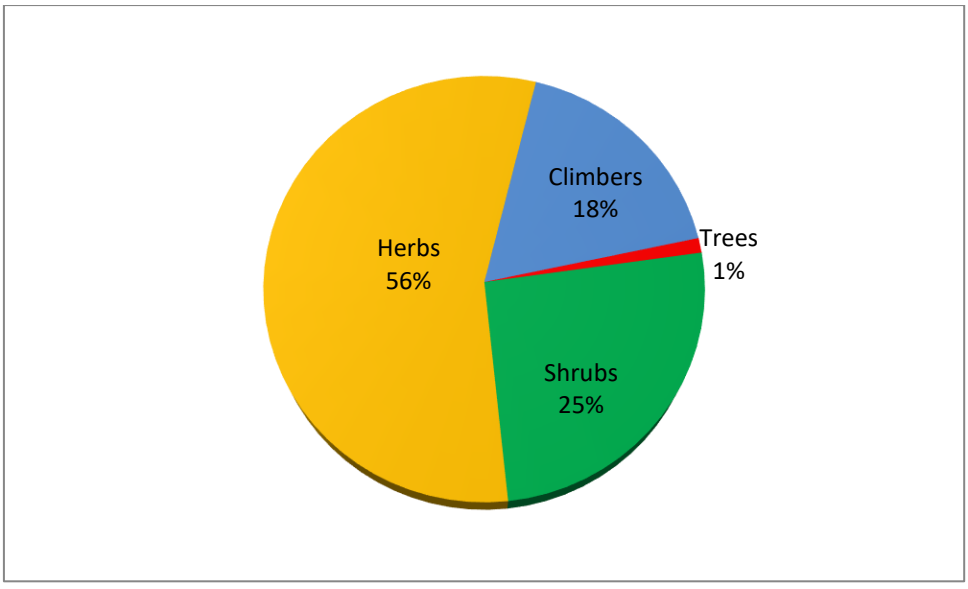


FIGURE 3: GENERIC AND SPECIES DIVERSITY IN THE SELECTED FAMILIES

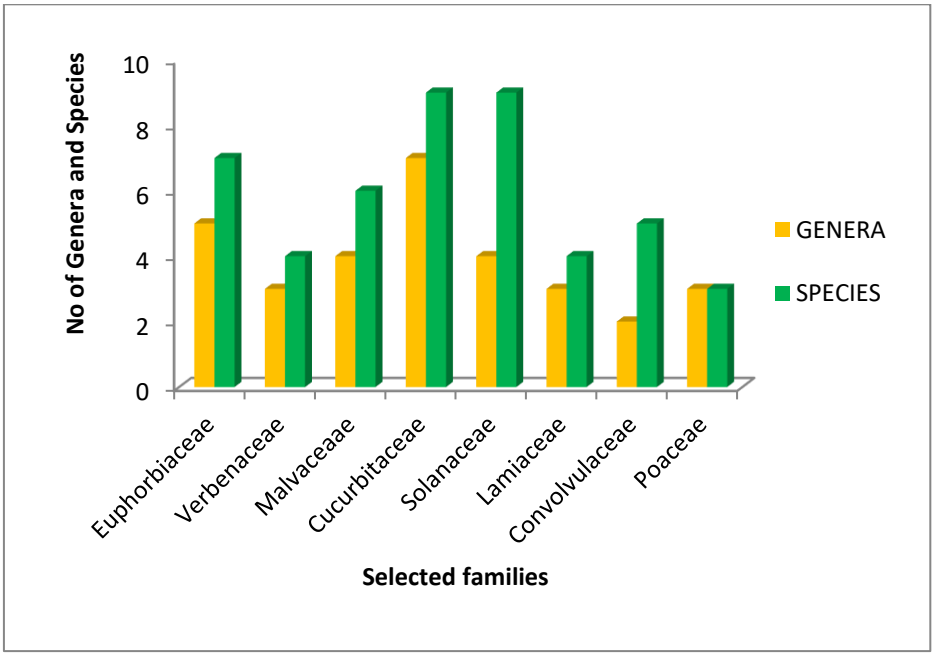


TABLE 3: DIFFERENT LIFE FORMS OF SPECIES IN THE STUDY AREA

S. No.	Botanical Name	Family	Habit
1.	*Ricinus communis* L.	Euphorbiaceae	Shrub
2.	*Jatropha gossypiifolia* L.	Euphorbiaceae	Shrub
3.	*Chrozophora brocchiana* (Vis.) Schweinf.	Euphorbiaceae	Herb
4.	*Euphorbia peplus* L.	Euphorbiaceae	Herb
5.	*Euphorbia serpens* Kunth.	Euphorbiaceae	Herb
6.	*Croton bonplandianus* Baill.	Euphorbiaceae	Herb
7.	*Euphorbia prostrate* Aiton.	Euphorbiaceae	Herb
8.	*Stachytarpheta jamaicens* (L.)	Verbenaceae	Herb
9.	*Lantana camara* L.	Verbenaceae	Shrub
10.	*Lantana aculeate* L.	Verbenaceae	Shrub
11.	*Phyla nodiflora* (L.) Greene	Verbenaceae	Herb
12.	*Acacia tortilis* (Forssk).	Leguminosae	Tree
13.	*Senna occidentalis* (L.) Link	Leguminosae	Shrub
14.	*Galactia spp.*	Leguminosae	Climber
15.	*Cassia angustifolia* Vahl.	Caesalpiniaceae	Shrub
16.	*Pergularia daemia* (Forsskal) Chiov.	Asclepiadaceae	Herb
17.	*Oxystelam esculentum* R. Br.	Asclepiadaceae	Climber
18.	*Calotropis procera* (Aiton) Dryand.	Asclepiadaceae	Shrub

S. No.	Botanical Name	Family	Habit
19.	*Calotropis gigantea* (L.) Dryand.	Asclepiadaceae	Shrub
20	*Ziziphus spina-christi* (L.) Desf.	Rhamnaceae	Shrub
21	*Tridax procumbens* (L.)	Compositae	Herb
22	*Parthenium hysterophorus* L.,	Asteraceae	Herb
23	*Stoebe plumose* L.	Asteraceae	Herb
24	*Sida spinosa* L.	Malvaceae	Herb
25	*Sida cordifolia*, L.	Malvaceae	Shrub
26	*Abelmoschus ficulneus* (L.)	Malvaceae	Shrub
27	*Hibiscus calyphyllus* Cav.	Malvaceae	Shrub
28	*Abutilon indicum*, (L) Sweet.	Malvaceae	Shrub
29	*Tribulus terrestris*, L.	Zygophyllaceae	Herb
30	*Luffa cylindrica (*L.) M. Roem	Cucurbitaceae	Climber
31	*Citrullus lanatus* (Thunb.) Matsum & Nakai	Cucurbitaceae	Climber
32	*Momordica cymbalaria* (Hook & Fenzl)	Cucurbitaceae	Climber
33	*Cucumis melo* L.	Cucurbitaceae	Climber
34	*Momordica charantia* L.	Cucurbitaceae	Climber
35	*Citrullus colocynthis* (L.) Schrad	Cucurbitaceae	Herb
36	*Mukia maderaspatana* W.	Cucurbitaceae	Climber

S. No.	Botanical Name	Family	Habit
37	*Lagenaria siceraria* (Molina) Standley.	Cucurbitaceae	Climber
38	*Cucurbita maxima* Duch.	Cucurbitaceae	Climber
39	*Achyranthes aspera* L.	Amaranthaceae	Herb
40	*Celosia cristata* L.	Amaranthaceae	Herb
41	*Cardiospermum halicacabum* L.	Sapindaceae	Climber
42	*Physalis peruviana* L.	Solanaceae	Herb
43	*Physalis angulata* L.	Solanaceae	Herb
44	*Solanum pimpinellifolium* L.	Solanaceae	Herb
45	*Solanum linnaeanum* Happer	Solanaceae	Shrub
46	*Solanum trilobatum*, L.	Solanaceae	Climber
47	*Capsicum annuum* L.	Solanaceae	Herb
48	*Datura matel* L.	Solanaceae	Shrub
49	*Solanum incanum* L.	Solanaceae	Shrub
50	*Solanum melongena* L.	Solanaceae	Shrub
51	*Cyanthillium cinereum (L.) H. Rob*	Asteraceae	Herb
52	*Passiflora foediax* L.	Passifloraceae	Climber
53	*Passiflora edulis Sims*	Passifloraceae	Climber
54	*Leucas aspera* (Willd.) Link	Lamiaceae	Herb

S. No.	Botanical Name	Family	Habit
55	*Ocimum tenuiflorum*	Lamiaceae	Herb
56	*Mesosphaerum suaveolens*	Lamiaceae	Shrub
57	*Ocimum kilimandscharium* Baker ex Gurke	Lamiaceae	Shrub
58	*Merremia disseca* (Jacq.) Hallier f.	Convolvulaceae	Climber
59	*Merremia aegyptia* (L.) Urb.	Convolvulaceae	Climber
60	*lpomoea aquatic*, Forsk.	Convolvulaceae	Herb
61	*Ipomoea quamoclit* L.	Convolvulaceae	Herb
62	*Ipomoea* carnea Jacq.	Convolvulaceae	Shrub
63	*Barleria volkensii* Lindau	Acanthaceae	Herb
64	*Hygrophila auriculata* (K. Schum.)	Acanthaceae	Herb
65	*Dicliptera paniculata* (Forssk.) I. Darbush	Acanthaceae	Herb
66	*Pedalium mure* L.	Pedaliaceae	Herb
67	*Ludwigia octovalvis* Linn.	Onagraceae	Herb
68	*Ludwigia palustris* (L.) Elliott.	Onagraceae	Herb
69	*Heliotropium indicum* L.	Boraginaceae	Herb
70	*Stemodia durantifolia* (Linn.)	Scrophulariaceae	Shrub
71	*Schoenoplectus tabernaemontani* (C.C. Gmel.)	Cyperaceae	Herb
72	*Corchorus trilocularis*, L.	Tiliaceae	Herb

S. No.	Botanical Name	Family	Habit
73	*Cleoserrata speciosa* (Raf.) Iltis	Cleomaceae	Herb
74	*Cleome viscosa* L.	Capparidaceae	Herb
75	*Cleome gynandra* L.	Capparidaceae	Herb
76	*Commelina communis* L.	Commelinaceae	Herb
77	*Argemone mexicana* L.	Papaveraceae	Herb
78	*Impatiens balsamina* L.	Geraniaceae	Herb
79	*Phyllanthus polygonoides* Nutt.	Euphorbiaceae	Herb
80	*Turnera ulmifolia* L	Turneraceae	Herb
81	*Hydrocotyle spp.*	Araliaceae	Herb
82	*Jasminum sambac* (L.) Aiton	Oleaceae	Shrub
83	*Ammannia coccinea* Rottb.	Lythraceae	Herb
84	*Scleromitrion diffusum* (Willd.) R.J.Wang	Rubiaceae.	Herb
85	*Oldenlandia umbellate* L.	Rubiaceae	Herb
86	*Citrus aurantiifolia* (Christm.) Swingle	Rubiaceae	Shrub
87	*Boehmeria cylindrica* (L.)	Urticaceae	Herb
88	*Brachiaria brizantha* (Hochst. ex A. Rich.)	Poaceae	Herb
89	*Digitaria sanguinalis* (L.)	Poaceae	Herb
90	*Orysa sativa* L.	Poaceae	Herb

borne on the same plant (monoecious) in terminal panicle-like inflorescences . The fruit is a spiny, greenish. Castor seeds have a warty appendage called the caruncle.

2. *Jatropha gossypiifolia* L.

Family: Euphorbiaceae

Habit: Shrub

Useful Parts: Leaves, Stem, Roots, Seeds and latex

Uses: Mainly used for biodiesel production. And also used for pesticide, insecticide vermifuge and ornamentation and even it was used in religious rituals.

Description: A small, much-branched shrub. 3-5 lobed, cordate, deep-purplish red at first afterwards green, lobes elliptic-acute, petiole red, covered with glandular hairs, branched from the base and mixed with simple hairs, stipules with glandular hairs, the margins of the leaf ciliate, with simple white hairs and gland tipped hairs. Terminal cymes inflorescence. Flowers are monoecious, Calyx cup-like with 5 lanceolate sepals. Petals 5, dark red, broadly ovate, stamens 6-8, dark-red, broadly ovate, anthers horse-shoe shaped, red, filaments connate in a central column on a glandular disk. Ovary seated on a glandular disk. Styles 3, united below, each ending in bifid stigma. Fruits are capsule 3 furrowed, truncate at both ends, seeds red with a caruncle. Flowering and Fruiting time is August-November.

3. *Chrozophora brocchiana* (Vis.) Schweinf.

Family: Euphorbiaceae,

Habit: Herb

Uses: In Iran, the plant is used to care for warts, emetic, cathartic, and fever whereas root ashes are given to children for cough.

Useful Parts: Leaves, Root, Stem

Description: Herb to undershrub, up to 60 cm high; flowering twigs 2–2.5 mm thick. Indumentum consisting of stellate and (few) simple hairs. Stipules 1.3-2.7 by c. 0.3 mm. Leaves: petiole 0.8–5.5 cm long; blade ovate, not to usually distinctly 3-lobed,

2.6–9 by 2.6–9 cm, index 1–1.9, pale green, base obtuse to usually emarginate, margin without distinct glands, lower surface with 2 glands near the base, 1 mm in diam., and usually several smaller ones sub marginally, venation impressed above, raised below, nerves c. 4 per side. Inflorescences up to 4 cm long, elongating in fruit to up to 10 cm long. Bracts very inconspicuous to c. 1.6 by 0.3 mm. Staminate flowers 4–6 mm in diam., yellow; calyx white, united part c. 1 mm high, lobes 3.2–4 by c. 1.2 mm; petals 3.7–3.8 by 1.5–1.6 mm; androphore 3.3–3.8 mm long, basal 1.2–1.3 mm without filaments; filaments 0.3–0.8 mm long; anthers 0.9–1.3 by c. 0.7 mm, yellow. Pistillate flowers 3.2–3.3 mm in diam., (greenish to) yellow; pedicel 1.4–2 mm long, elongating in fruit to up to 1.1 cm; calyx lobes only basally united, 1.5–2.2 by 0.5–0.7 mm; petals 1.3–2 by 0.4–0.6 mm; ovary ovoid, 2.7–3 by 2.2–3 mm wide; style 0.5–0.8 mm long, red, stigmas erect, up to 2.3 mm long, apically split for up to 1.8 mm, red. Fruits 8–9 by c. 5 mm; column after dehiscence 3–3.5 mm long. Seeds 3.8–3.9 by 3.2–3.6 by 3–3.2 mm.

4. *Euphorbia peplus* L.

Family: Euphorbiaceae

Habit: Herb

Uses: It is used to treat treating actinic keratoses and nonmelanoma skin cancer.

Useful Parts: Leaves and Root.

Description: Radium Weed is an annual plant growing to between 5-30 cm tall. They are commonly found small in size, growing as weeds of cultivation. Stems are smooth, hairless. Leaves are oval-pointed, 1-3 cm long, with a smooth margin. It has green flowers in three-rayed umbels. The glands, typical of the spurge family, are kidney-shaped with long thin horns. In India it is found in Jammu and Kashmir and Meghalaya.

5. *Euphorbia serpens* Kunth.

Family: Euphorbiaceae

Habit: Herb

Useful Parts: Leaves, Flowers and Seeds.

Uses: It is used to treat breathing disorders.

Description: *Euphorbia serpens* is a species of Euphorbia known by the common name matted sandmat. It is native to South America but it can be found on most continents as an introduced species and often a weed. This is an annual herb forming a mat of prostrate stems which root at nodes where the stem comes in contact with the ground. The oval leaves occur in oppositely arranged pairs, each leaf less than a centimetre long. The inflorescence is a cyathium with scalloped white petal-like appendages surrounding the actual flowers. A red nectar gland is at the base of each appendage, and at the centre of the cyathium are several male flowers around one female flower. The fruit is a lobed, spherical capsule.

6. *Croton bonplandianus* Baill.

Family: Euphorbiaceae

Habit: Herb

Useful Parts: Leaves and Stem

Uses: It is used to treat liver disorders, skin diseases including ring worm infection, to cure the swelling of body, bronchitis and asthma.

Description: This is a wild species of croton. Due to the resemblance of the leaves and flower cymes to that of Tulsi, this plant is often called Ban Tulsi (Jungle Tulsi) It is a small annual herb, growing up to 1-2 ft tall. Alternately arranged leaves, 3-5 cm long, are lance-shaped, with a toothed margin. Small white flowers are borne in 3-8 cm long racemes at the end of branches. Flowers have 5 sepals and 5 petals and numerous long stamens protruding out. Fruit is a 5 mm oblong capsule, with a warty surface. Ban Tulsi is grown abundantly in the rural areas of Malda, West Bengal, and is used as both a fuel and a detergent. First the stems and branches of ban tulsi are used as fuel. Then the ash is collected and kept in a bottle for five or six days. The ash is put in warm water and used as a detergent for cleaning cotton garments. Ban Tulsi is native to S. Bolivia to Uruguay, now widespread in the Indian subcontinent. Flowering: September-November.

7. *Euphorbia prostrate* Aiton.

Family: Euphorbiaceae

Habit: Herb

Useful Parts: Leaves, Seeds and Roots

Uses: It is used to treat piles, bronchitis, asthma, and tumour

Description: Euphorbia prostrata is an annual herb producing slender prostrate stems up to approximately 20 centimetres (7.9 in) long, sometimes purple-tinted in colour. The oval-shaped leaves are up to one centimetre (0.39 in) long with finely toothed edges. The inflorescence is a cyathium less than two millimetres (0.079 in) wide, with white petal-like appendages surrounding the actual flowers. There are four male flowers and a single female flower, the latter developing into a lobed, hairy fruit one to two millimetres (0.039 to 0.079 in) wide.

8. *Stachytarpheta jamaicens* (L.)

Family: Verbenaceae

Habit: Herb

Useful Part: Leaves

Uses: The fresh leaves are consumed in bush tea in cooling tonic and blood cleanser to treat asthma and ulcerated stomachs.

Description: Low, usually spreading annual or perennial herbs, 6-12 dm tall, sometimes somewhat woody toward base, often purplish throughout; stems dichotomously branched, sparsely pubescent or glabrate, nodes usually sparsely pilose. Leaves often bluish or greyish when fresh, alternate or opposite, somewhat fleshy, oblong to elliptic or ovate, 2-9 (-12) cm long, 1.5-4 cm wide, upper surface glabrate, lower surface glabrous to strigose on the veins and margins serrate, the teeth angled forward, petioles 0.3-3.5 cm long. Spikes stout and stiff, 15-50 cm long, glabrous, rachis stout and firm, up to 7 mm in diameter, the furrows conspicuously narrower than the rachis, bracts lanceolate or oblong-lanceolate, 5-8 mm long; calyx completely embedded in rachis furrows, somewhat

compressed, ca. 5 mm long, 2 of the teeth very reduced; corolla usually pale blue, 8-11 mm long.

9. *Lantana camara* L.

Family: Verbenaceae

Habit: Shrub

Useful Part: Leaves

Uses: It has anti-insecticidal, Fungicidal and antimicrobial properties and it is used to cure cancer, skin itches, leprosy, chicken pox, measles, asthma and ulcers.

Description: *Lantana camara* is a perennial, erect sprawling or scandent, shrub which typically grows to around 2 metres. Under the right conditions, it can scramble up into trees and can grow to 6 m (20 ft) tall. The leaves are broadly ovate, opposite, and simple and have a strong odour when crushed. It has small tubular-shaped flowers, which each have four petals and are arranged in clusters in terminal areas stems. Flowers come in many different colours, including red, yellow, white, pink and orange, which differ depending on location in inflorescences, age, and maturity. The flower has a smell with a peppery undertone. After pollination occurs, the colour of the flowers changes; this is believed to be a signal to pollinators that the pre-change colour contains a reward as well as being sexually viable, thus increasing pollination efficiency. In frost-free climates the plant can bloom all year round, especially when the soil is moist.

10. *Lantana aculeate* L.

Family: Verbenaceae

Habit: Shrub

Useful part: Leaves

Uses: It has anti-insecticidal, Fungicidal and antimicrobial properties and it is used to cure cancer, skin itches, leprosy, chicken pox, measles, asthma and ulcers.

Description: The plant has various medicinal applications such as sedative, to relieve urinary tract infections, treatment of chest complaints, to treat snake bite and intoxication. It is used in flavouring in cakes, sweet breads and candy.

11. *Phyla nodiflora* (L.) Greene

Family: Verbenaceae

Habit: Herb

Useful parts: Leaves and Fruits

Uses: Plant decoction is given in uraema. Fresh juice is applied to bleeding gums. Infusion of leaves and tender stalk is given to children in indigestion and to women after delivery.

Description: Appressed pubescent, prostate to ascending or decumbent, perennial herbs, rooting at the nodes, obscurely to definitely 4- angled. Leaves opposite, serrate, base cuneate to attenuate; petioles to 0.5 mm long, often obscured by decurrent blade tissue. Inflorescence a bracteate head, in fruit a spike 8-15 cm long, 5-8 mm in diam., peduncles elongate, usually at alternate nodes and rarely in both axils at a node. Sepals united near base or for ½ their length, shorter than the corolla tube and the subtending bract; corolla zygomorphic, pinkish, lavender or rarely white, salver form, ca. 3 mm long, 5-lobes less than 1 mm long; stamens included, united to the corolla tube near middle at 2 levels. Fruit a schizocarp consisting of 2 mericarps.

12. *Acacia tortilis* (Forssk).

Family: Leguminosae

Habit: Tree

Useful Part: Leaves

Uses: *Acacia tortilis* possesses valuable medicinal property and therapeutic potential so, it is also useful for treatment of various diseases like skin allergy, cough, inflammatory reaction.

Description: *Acacia tortilis* is a small to medium-sized evergreen tree or shrub that grows up to 21 m tall. Leaves glabrous to densely pubescent, glandular, short at 1.25-3.75 cm long; petiole 0.2-0.9 cm long. Inflorescence globose heads; peduncle white, pubescent, 0.4-2.5 cm long, with involucel on the lower half; flowers white or pale yellowish-white, sessile or shortly pedicellate, scented, 0.5-1.1 cm in diameter, on axillary peduncles; calyx 1-2 mm long; corolla 1.5-2.5 mm long. Pods variable, indehiscent, spirally twisted or rarely almost straight, 7-10 cm long, 6-10 (max. 13) mm broad, longitudinally veined, leathery, glabrous to tomentellous or villous, somewhat constricted between the seeds; seeds oblique or parallel to long axis of pod, 4-7 x 3-6 mm, compressed; areole 3-6 x 2-4 mm.

13. *Senna occidentalis* (L.) Link

Family: Leguminosae

Habit: Shrub

Uses: The seed is bitter and has purgative properties. It is also used as a diuretic, liver detoxifier, as a hepato-tonic (balances and strengthens the liver). Further, used in whooping cough and convulsion.

Useful Part: Seed

Description: *Senna occidentalis* is a Fabaceae family weed that grows in damp, disturbed, or junkyards at low altitudes throughout the world. First discovered in tropical South America, the plant is deadly if consumed in excessive numbers; however, all components of the plant are utilised as medicine and food by many people worldwide. The seeds are frequently used as a coffee replacement. It is a 0.5-2.5 m tall, unarmed thin straight shrub. It is a yearly or eternal Ayurvedic plant used in numerous traditional medicines to treat various ailments. This weed contains germ-destroying, antimycotic, antidiabetic, anticancer, antimutagenic, anti-inflammatory, and antihepatotoxic properties.

15. *Cassia angustifolia* Vahl.

Family: Caesalpiniaceae

Habit: Shrub

Useful Parts: Leaves, Pods and Flowers

Uses: It is used to treat stomach pain and constipation**.**

Description: Alexandrian Senna is a shrubby plant that reaches 0.5–1 metre (20" to 40"), rarely two metres (6') in height with a branched, pale-green erect stem and long spreading branches bearing four or five pairs of leaves. These leaves form complex, feathery, mutual pairs. The leaflets vary from 4 to 6 pairs, fully edged, with a sharp top. The midribs are equally divided at the base of the leaflets.

16. *Pergularia daemia* (Forsskal) Chiov

Family: Asclepiadaceae

Habit: Herb

Useful Part: Leaves, Root bark, Stem.

Uses: Used to cure cough, asthma, rheumatism, bronchitis, pies, liver disorder, uterus related problems, digestion related problems, reduces the swelling, inflammation and pain in the joints.

Description: *Pergularia* is a perennial twining herb, foul-smelling when bruised and with much milky juice, stem hairy. Leaves are thin, broadly ovate, heart-shaped or nearly circular, hairless above, velvety beneath. Greenish yellow or dull white, and sweet-scented flowers are borne in lateral cymes which are at first corymb-like, afterwards raceme-like. The five petals are hairy and spreading outwards. Corona outer and inner, outer truncate, inner curved high over the staminal column, spur acute. Fruit is a follicle, with soft spines all over and a long beak. Seeds are densely velvety on both sides. Flowering: August-February.

17. *Oxystelam esculentum* R. Br.

Family: Asclepiadaceae

Habit: Climber

Useful Parts: Leaves and Flower

Uses: This plant has strong phyto-medicinal constituents and used for remedies from cancer, hepatitis, kidney disorders, stress-related disorders and various microbial infections

Description: Twinging subshrubs. Leaves simple, opposite, 4-8 x 0.5-2 cm, oblong to linear, sub coriaceous, base truncate, apex mucronate; petiole ca. 1 cm. Flower(s) usually paired, axillary, solitary or in lax racemes; peduncle 6-15 cm; bracts deciduous; pedicel ca. 1 cm. Calyx copular; lobes equal, ovate, imbricate, 4 mm, chartaceous, glabrous, acute. Corolla white or pink with dark pink stripes within, 2.5 cm across, rotate to angulate; lobes triangular, united at the mid half, valvate, 2 cm, shortly overlapping to right in bud, chartaceous, ciliate, acute, recurved. Pollinia pendulous; pollinial bags oblong, 1.3 mm; caudicle 0.2 mm. Corona double; outer coralline, annular, pubescent at the base of corolla within; inner staminal, basally inflated, 5 mm; staminal column 7 mm. Ovaries globose, 3 mm; placenta bifurcate; style 4 mm. Follicle 5 x 2 cm, inflated, mucronate; seeds ovate, 2.5 x 2 mm, base rounded, apex pointed; coma silky, dull white.

18. *Calotropis procera* (Aiton) Dryand.

Family: Asclepiadaceae

Habit: Shrub

Useful Parts: Roots, Stem, Leaves and flowers

Uses: Arkavaleha', made from this plant, is given to cure irritation of the stomach, nausea, vomiting, diarrhoea etc. Eight patents were found on the medicinal uses mainly for anti-tumour and antidote activity, and bronchial asthma.

Description: Shrub or small tree up to 2.5(–6) m tall, stems erect, simple or branched, woody at base, bark grey or pale brown, fissured, corky, slash yellowish-white, latex copious; young branches densely white hairy, soon almost glabrous. Leaves opposite, decussate, simple and entire, almost sessile; stipules absent; blade oblong-obovate to broadly obovate, 5–30 cm × 2.5–15 cm, apex abruptly and shortly acuminate, base cordate, succulent, densely white short-hairy below when young, pinnately veined with 6–10 pairs of lateral veins. Inflorescence an axillary umbellate cyme up to 10 cm

in diameter. Flowers bisexual, regular, 5-merous; pedicel 1–3 cm long; calyx lobes ovate, 4–7 mm × 3–4 mm; corolla pale whitish-green with large lilac to purple patches on the lobes, campanulate, 2–3 cm in diameter, lobes broadly triangular, 11–20 mm × 9–10 mm, united at base for 6–7 mm; corona with 5 compressed lobes, 6.5–11 mm × 3–4.5 mm, adnate to the staminal column, purple; ovary superior, 2-celled, gynostegium c. 6 mm long, stigma head 5-pointed. Fruit a pair of follicles, each follicle ovoid, fleshy, inflated, 6–10 cm × 3–7 cm, many-seeded. Seeds ovoid, flattened, c. 6 mm long, with 3–4 cm long white coma at one end**.**

19. *Calotropis gigantea* (L.) Dryand

Family: Asclepiadaceae

Habit: Shrub

Useful Part: Flower

Uses: The plant is reported as effective in treating skin, digestive, respiratory, circulatory and neurological disorders and was used to treat fevers, elephantiasis, nausea, vomiting, and diarrhea. The milky juice of Calotropis procera was used against arthritis, cancer, and as an antidote for snake bite

Description: Large, white, not scented, peduncles arising between the petioles. Flower-buds ovoid, angled, Calyx lobes 5, divided to the base, white, ovate; corolla broadly rotate, valvate, lobes 5, deltoid ovate, reflexed, coronate-appendages broad, obtusely 2-auricled below the rounded apex which is lower than the staminal-column. Stamens 5, anthers short with membranous appendages, inflexed over the depressed apex of the pentagonal stigma. Pollinium one in each cell, pendulous caudicle slender. Carpels 2 distinct, styles 2, united to the single pentangular stigma, ovary 2-celled, ovules many. Fruit: A pair of follicles with many, hairy seeds.

20. *Ziziphus spina-christi* (L.) Desf.

Family: Rhamnaceae

Habit: Shrub

Useful Part: Fruits

Uses: It is used for improving muscular strength and weight, for preventing liver diseases and stress ulcers, and as a sedative.

Description: A medium-size tree, with spreading, greyish white branches, glabrous or slightly pubescent. Stipular spines in pairs, one erect, c. 2 cm long, the other recurved 5-8 mm long, sometimes spines absent. Leaves 2-6 x 1-4 cm ovate-elliptic or suborbicular, glabrous or pubescent on nerves beneath, rounded to sub cordate at base, obtuse or shortly acuminate, margin entire or obsoletely crenate, 3-nerved; petiole 3-12 mm long, glabrous. Inflorescence axillary tomentose, pedicel woolly, c. 3-5 mm long. Flowers 4-6 mm across, greenish yellow. Calyx c. 1 mm long, keeled within, pubescent, ovate, ± acute, petals spathulate; 1.25 mm long. Disc prominently 10-lobed, glabrous, grooved.

21. *Tridax procumbens* (L.)

Family: Compositae

Habit: Herb

Useful Part: Whole Plant

Uses: It has been in use in India for wound healing and as an anticoagulant, antifungal, and insect repellent. It is also used as treatment for boils, blisters, and cuts by local healers in parts of India

Description: A decumbent herb. Stem and leaves are covered by hairs. Tap root system. Herbaceous, cylindrical, decumbent and branched. Simple, opposite, exstipulate and margins dentate showing reticulate venation. A terminal heterogamous head and receptacle of the head is convex and surrounded by green involucre. The tubular florets occupy the centre and the ligulate florets are found at the margins.

22. *Parthenium hysterophorus* L.

Family: Asteraceae

Habit: Herb

Useful Part: Leaves

Uses: *Parthenium hysterophorus* confers many health benefits, viz remedy for skin inflammation, rheumatic pain, diarrhoea, urinary tract infections, dysentery, malaria and neuralgia.

Description: This erect ephemeral herb can grow up to 1.5–2 m high and has a deep tap root. It has branching stems that become woody and hairy with age. Leaves are alternate, finely lobed, covered with fine soft hair, 3–20 cm long and 2–10 cm wide. Once stem elongation is initiated, smaller leaves are produced and the plant becomes multi-branched in its extremities. The whole plant has a bluish or greyish-green appearance. Flower heads are small (4 mm across) and numerous in open panicles, creamy-white, with 5 petals. Each flower produces about 5 black achenes which are obovate, 2–2.5 mm long and light weight. The fruit is a cypsela

23. *Stoebe plumose* L.

Family: Asteraceae

Habit: Herb

Useful Part: Underground parts

Uses: The form of *Seriphium plumosum* which was previously known as *Stoebe plumosa* was not used medicinally. However, in the Western Cape, the form previously known as *Stoebe cinerea* is still used as a remedy for heart trouble, whereas another unidentified Stoebe/Seriphium sp**.**

Description: This intricately branched, heath-like shrublet is well known to hikers who use the plant to make soft mattresses when sleeping outdoors. It might be better known to many people as Stoebe plumosa. Despite enormous garden potential and successful use at Kirstenbosch, it is not yet commonly cultivated Preparations of this species are taken orally for gynaecological problems, stomach ache. *Stoebe plumosa* Herb consists of the fresh or dried over ground parts of Stoebe plumose (L.) Thunb.

24. *Sida spinosa* L.

Family: Malvaceae

Habit: Herb

Useful Parts: Leaves and Root

Uses: It is used in the treatment of neurological and uterine disorders, headache, tuberculosis, diabetes, malarial fever, piles, ulcers, wounds, rheumatic and cardiac problems, diarrhoea and dysentery, skin diseases.

Description: The stems are erect to sprawling and branched, growing 50 to 120 centimetres in height, with the lower sections being woody. The dark green, diamond-shaped leaves are arranged alternately along the stem, 4 to 8 centimeters long, with petioles that are less than a third of the length of the leaves. The leaves are paler below, with short, greyish hairs. The apical half of the leaves have toothed or serrated margins while the remainder of the leaves are entire (untoothed). The petioles have small spiny stipules at their bases. The moderately delicate flowers occur singly on flower stalks that arise from the area between the stems and leaf petioles. They consist of five petals that are 4 to 8 millimeters long, creamy to orange-yellow in color, and may be somewhat reddish in the center. Each of the five overlapping petals is asymmetric, having a long lobe on one side. The stamens unite in a short column. The fruit is a ribbed capsule, which breaks up into 8 to 10 segments. The plant blooms throughout the year.

25. *Sida cordifolia* L.

Family: Malvaceae

Habit: Perennial shrub

Useful Part: Leaf and root

Uses: It is used to treat bronchial asthma, cold and flu, chills, lack of perspiration, head ache, nasal congestion, aching joints and bones, cough and wheezing, and oedema.

Description: It is a perennial shrub. The root is tap root system. The stem is woody and round, the bark hoary and woody with soft stellate hairs. The leaves are alternate, broadly ovate, 3-7 cm long and 3-5 cm wide. The leaf margins are dentate, rounded at

the base. The blade is soft to the touch and woolly on the lower surface and along the main nerves with fine stellate hairs. The leafstalk is long, about 4.5 cm. The inflorescence is an axillary or terminal raceme, leafy with many flowers. Calyx 5-7 mm long, accrescent and clasping in fruit; petals cream to pale yellow or orangish yellow, 7-9 mm long. The stamens are fused at the base tube pubescent, but the nets and the anthers are free. Fruit are capsules and each has 8-10 carpels. The fruit is hoary and densely covered with reflexed woody hairs; each carpel bears 2-bristly awns.

26. *Abelmoschus ficulneus* (L.)

Family: Malvaceae

Habit: Shrub

Useful Parts: Stems, Roots, Seeds, Fruit and Leaves

Uses: A good quality fibre is obtained from the stems. The white fibre is long, glossy, fine and strong. It is used for twine and light cordage

Description: Annual herb up to 2 m tall; stem thick, glabrous to densely glandular pubescent. Leaves alternate, simple stellate hairy; stipules linear or filiform, 5–12 mm long, hirsute; petiole 2–21 cm long, hairy; blade orbicular, deeply 3–5-lobed, up to 16 cm × 16 cm, cordate at base, lobes subacute to broadly rounded, margin serrate, scabrous on both sides. Flowers bisexual, regular, solitary in leaf axils or in a terminal raceme; pedicel 0.5–2.0(–2.5) cm long, expanded and cup-shaped apically; epicalyx bracts 5–6, linear to lanceolate, up to 12 mm × 2 mm, rough, caducous before expansion of corolla; calyx 17–23 mm long, 5-toothed, tomentellous; petals 5, obovate, 2–3.5 cm × 1.5–3 cm, uniformly white, turning pink; stamens many, filaments united in a column 1–1.5 cm long, glabrous; ovary superior, 5-celled. Fruit an ellipsoid capsule 3–4 cm × 1.5–2 cm, puberulous to pubescent; valves acute to aristate with up to 3 mm long awns. Seeds globose, 3–4 mm in diameter, black, with concentric lines, glabrous or with stellate or long crisped hairs.

27. *Hibiscus calyphyllus* Cav.

Family: Malvaceae

Habit: Shrub

Useful Part: Leaves

Uses: *Hibiscus calyphyllus* is cultivated throughout the tropics and subtropics as an ornamental. In DR Congo, the leaves are used in a mixture with several other plant species to prepare a cure for ganglions in domestic animals. In Kenya and Tanzania, the leaves are applied to wounds as a dressing.

Description: *Hibiscus calyphyllus* is a dense, rounded shrub; up to 3 m high; the leaves are large, up to 50 mm in diameter, light green, soft and velvety; the flowers are lemon-yellow, large, up to 100 mm in diameter, with a deep red to blackish centre; the fruit is a papery capsule that splits open to reveal hairy to hairless seeds. It is fairly fast growing and will flower repeatedly, the flowers lasting for a reasonable amount of time. The natural habitat of Hibiscus calyphyllus is open bush, thickets and forests, often also found along rivers.

28. *Abutilon indicum* (L) Sweet.

Family: Malvaceae

Habit: Shrub

Useful Parts: Leaves and Root

Uses: It was mainly used to cure Diarrhoea, Gonorrhoea, Antipyretic, cough, piles**.**

Description: Soft-wooded, much-branched shrub up to c. 1.5 m tall. Leaves more or less broadly ovate-cordate, up to 18 × 16 cm, with a long acuminate tip, unlobed, discolours, dark green above, paler grey-green below with prominent veins; margin slightly but distinctly serrate-crenate; petiole up to 18 cm long. Flowers axillary, often on short axillary shoots, yellow, sometimes reddish at the base or with reddish veins, c. 3 cm in diameter. Calyx more or less cup-shaped, 10-18 mm long and 8-10 mm in diameter; lobes 6-12 mm long, ovate-lanceolate to lanceolate-linear, gradually acuminate. Fruit 20-25 mm in diameter, stellate-pubescent with 23-40 mericarps, 2-3-seeded, black when dry.

29. *Tribulus terrestris* L.

Family: Zygophyllaceae

Habit: Herb

Useful Parts: Leaves, Stem, Root

Uses: It is used to cure chest pain, heart problems, dizziness, skin and eye disorders, to expel kidney stones, and as a diuretic and tonic.

Description: It is a trailing perennial, hirsute, procumbent and branched herb. The stems and branches are pilose and young parts are silky-villous. Leaves are stipulate, opposite usually unequal and abruptly pinnate. Leaflets are 5-8 in pairs with length 0.5- 1.3 cm, sub-equal, oblong to linear oblong and mucronate; petioles very short and pilose. Flowers are yellow, solitary, axillary, 8-12 mm in diameter and appear during July-August. Style is short and stout; ovary is bristly 5-10 lobed and with 5-12 celled; fruits are globose and spinous produced during autumn. It consists of 5-12 woody cocci, each with two pairs of hard sharp spines, one pair longer than the other. Each coccus contains several seeds with transverse partition between them. The seeds are obliquely pendulous and have hard seed coat.

30. *Luffa cylindrica (*L.) M. Roem

Family: Cucurbitaceae

Habit: Climber

Useful Parts: Leaves, fruit, seed

Uses: It is used to cure pulmonary troubles, cancer and tumour.

Description: *Luffa* is a large climber with a stout, 5 angled pubescent stem tendrils usually 2–3 branched. Leaves are large, 10–20 cm long, and 9–21 cm wide. Orbicular to reniform orbicular in outline, base cordate, 5–7 lobed, lobes broadly triangular, acute, margins irregularly, shallowly dentate, hispid, scab rid above, finely pubescent-hispid beneath, petioles 2–8 cm long. The male and female flowers deep, bright yellow and male flowers borne in racemes on peduncles 6–17 cm long. It is usually aggregated near apex, stamens usually 15–30 cm long and 6–10 cm in diameter, smooth with 10 dark green longitudinal lines, fibrous within, seeds 1–2 cm long and 0.8 cm wide, including the narrow, smooth, marginal wing, broadly

ellipsoid, longitudinally compressed, rough on the surface The two species of *Luffa* are somewhat similar in appearance. Both are vigorous, climbing, annual vines with several lobed cucumber-like leaves. When crushed, the leaves give off a rank odour. Both male and female flowers occur on the plant with a much greater number of male flowers. The rather large male flowers are bright yellow and occur in clusters. The female flowers are solitary and have the tiny, slender ovary attached. Angled luffa flowers appear later in the day than the smooth type and stay open through the night. Pollination is by entomophilly.

31. *Citrullus lanatus* (Thunb.) Matsum & Nakai

Family: Cucurbitaceae

Habit: Climber

Useful Part: Fruit

Uses: Traditional herbal practitioners employ *C. lanatus* seeds to treat gastrointestinal, respiratory, and urinary diseases in Pakistan and India. However, more investigation is needed to understand the effect of C. lanatus seeds on treating gastrointestinal, respiratory, and urinary disorders.

Description: Watermelon is an annual herbaceous vine with long (up to 10 m) stems lying or creeping on the ground, with curly tendrils. Leaves are 5-20 by 3-19 cm, and hairy, usually deeply palmately lobed with 3-5 lobes. Leaf stalks are 2-19 cm long. Male flowers on 1.2-4.5 cm long pedicels. Flowers 1-2.5 cm long, pale green. Flowers monoecious, solitary, on pedicels up to 4.5 cm long; with 5 shortly united petals, pale green. Fruit of wild plants 1.5-20 cm in diameter, nearly spherical, greenish, mottled with darker green; of cultivated plants up to 30x60 cm, spherical or ellipsoid, green or yellowish, evenly coloured or variously mottled or striped. Fruits vary considerably in morphology. The cultivated forms of the fruit are large oblong.

32. *Momordica cymbalaria* (Hook & Fenzl)

Family: Cucurbitaceae

Habit: Climber

Useful Part: Root, Leaves and Fruit

Uses: Plant parts such as root, leaves and fruit were treated for various ailments.

Description: Little Wild Gourd is a perennial herb with slender, climbing, branched, striped stem. It is a trailing plant with large turnip shaped tuberous rootstock. The leaves are round-kidney-shaped in outline, deeply heart-shaped at the base, bluntly lobed with 5-7 lobes. flowers are white to yellow in color. Flowers are small, unisexual in nature. The male flower flower-cluster-stalk is 5-30 mm long, thread-like, hairy, ebracteate with 2-5 flowers in racemes with a pale-yellow flower and two stamens for each flower. The female flower is solitary on a flower-cluster-stalk of 2.8 cm long. The fruits are 2.0-2.5 cm long, pyriform with 8 sharp ridges, 2.4 x 1.5 cm narrowed at the tip and with the base narrowed into the curved flower-cluster-stalk, which is fleshy, dark green and robbed. Seeds are 4.6 mm long, ovoid shaped, black, smooth and shiny.

33. *Cucumis melo* L.

Family: Cucurbitaceae

Habit: Climbers

Useful Parts: Leaves and Fruit

Uses: It is used for the treatment of kidney stones, flatulence, leprosy, fever, jaundice, diabetes, obesity, cough, bronchitis, ascites, anaemia, constipation and other abdominal disorders.

Description: *C. melo* is an annual trailing vine with pubescent striated stems, lacking stipules, bearing unbranched tendrils at the base of the 4-12cm long petioles. The leaves are simple and alternate, nearly round, basally cordate, and may have 3-7 shallow palmate lobes. The flowers of C. melo can be gynoecium (only female flowers), monoecious (male and female flowers), or perfect. The hypanthium is

broader at the apex and 0.7-0.8cm long. Staminate flowers are solitary or fascicled, bearing 3 free stamens, of which two bear 2-celled anthers and one a 1-celled anther. Pistillate flowers are solitary, bear staminodes, an inferior ovary topped by a short style (1-2mm long) and 3-lobed stigma (2-2.4mm long). The ellipsoid ovary is densely pubescent with white hairs, and is 0.4-1.1cm long (3,9,11,13). The fruit is berry. The seed is endospermous.

34. *Momordica charantia* L.

Family: Cucurbitaceae

Habit: Climber

Useful Parts: Fruit, Flowers and Seeds

Uses: It is used to cure ulcers, breast cancer, diabetes and heart problems**.**

Description: It is annual to perennial herb. The stem is herbaceous, tendril-bearing vine grows to 5 meters. It may be either hairless or slightly hairy. The leaf is Up to 5cm long, with spiral tendrils at opposite sides, Petioles 4-5 cm long, pubescent. The leaves are carried singly along the stems, and each leaf is 4-10 cm long, rounded in outline, and deeply 5-9 lobed. The flowers are Monoecious pale to deep yellow flowers with bract part way on peduncle, solitary in the upper leaf axils on 2-10 cm long stalks with a small leaf-like bract towards the base. Male flowers larger than female flowers and have a slender basal swelling which is continuous with the base of the sepal tube, which ends in five blunt sepals. There are five oval yellow petals 10-20 cm long, and five central stamens. Female flowers are similar to the male flowers but have a distinct warty swelling well below the base of the sepal tube and three stigmas. The fruit is berry. The seed is endospermous.

35. *Citrullus colocynthis* (L.) Schrad

Family: Cucurbitaceae

Habit: Herb

Useful Parts: Fruit, seed and flower

Uses: It is used to cure diabetes, leprosy, common cold, cough, asthma, bronchitis, jaundice, joint pain, cancer, toothache, wound, mastitis, and in gastrointestinal disorders such as indigestion, constipation, dysentery, gastroenteritis

Description: The roots are large, fleshy, and perennial, leading to a high survival rate due to the long tap root. The vine-like stems spread in all directions for a few meters looking for something over which to climb. If present, shrubs and herbs are preferred and climbed by means of auxiliary branching tendrils. The leaves are palmate and angular with three to seven divided lobes. The flowers are yellow and solitary in the axes of leaves and are borne by yellow-greenish peduncles. Each has a sub campanulate five-lobed corolla and a five-parted calyx. They are monoecious, so the male (stamens) and the female reproductive parts (pistils and ovary) are borne in different flowers on the same plant. The male flowers' calyx is shorter than the corolla. They have five stamens, four of which are coupled and one is single with monadelphous anther. The female flowers have three staminoids and a three-carpel ovary. The two sexes are distinguishable by observing the globular and hairy inferior ovary of the female flowers. The fruit is mesocarp. The seeds are endospermous.

36. *Mukia maderaspatana* W.

Family: Cucurbitaceae

Habit: Climber

Useful Parts: Leaves, Root and Fruits

Uses: According to Siddha and Ayurveda medicine, the leaves, root and fruits are considered stomachic, anti-ulcer, anti-inflammatory, antipyretic, diuretic, hepatoprotective, expectorant, carminative, anti-hyperglycaemic, anti-hyperlipidaemic, antimicrobial, antioxidant and anti-rheumatic activities**.**

Description: Prostrate or climbing herbs; tendrils simple. Leaves 4-8 x 3-7 cm, ovate-deltoid, angular or shallowly 3-5-lobed, base cordate, margin denticulate, apex acuminate, mucronate, on both sides; petiole to 6 cm. Male flowers in axillary, sessile clusters. Calyx tube to 2 mm, villous; lobes subulate, erect. Petals 5, c. 3 mm long, ovate-oblong, obtuse, yellow. Stamens 3, free, inserted at base of calyx tube; anthers

oblong, ciliate. Female flowers solitary or in clusters. Ovary villous. Berry c. 1.2 cm across, globose, red. Seeds lenticular, rugose.

37. *Lagenaria siceraria* (Molina) Standley.

Family: Cucurbitaceae

Habit: Annual Climber

Useful Part: Fruit

Uses: It is used as a emetic, purgative, cooling, sedative, anti-bilious, and pectoral. Its pulp, boiled in oil is used to treat Rheumatism**.**

Description: The elongate fruits average 15–20 cm in length and several centimeters in diameter and are eaten prior to maturity, as the mature fruits are bitter. The seeds are covered with sweet, red arils, but the seeds themselves are poisonous. This species is grown mostly in southern Asia.

38. *Cucurbita maxima* Duch.

Family: Cucurbitaceae

Habit: Climber

Useful Parts: Seeds, Fruit and flowers**.**

Uses: The seeds are used to prepare, tonic diuretic and vermifuge. The flowers are used to soothe minor injuries. The fruit pulp is to cure burns, inflammations and ear ache.

Description: *C. maxima* is an annual herb with thick climbing or creeping stems. The root system is well developed and roots are up to 40 cm deep and 5 m long. The stems are branching, covered in soft white pubescence, up to 10 m long, and often produce adventitious roots at nodes. The petioles are densely pubescent, 5-20 cm long, and estipulate. The plant bears tendrils at 90 degrees to the leaf axil; these are lightly pubescent, coiled, and 2-5-branched. The thin leaves are alternate, simple, palmately veined, round to reniform, basally cordate, apically obtuse, unlobed to shallowly 5-7 lobed, 7-30 cm across, broader than long, stiff to soft pubescent, and finely dentate.

The flowers produce nectar and are aromatic. Staminate flowers are 20-25 times more numerous than pistillate flowers, but produce less nectar. The campanulate calyx is covered in white pubescence and bears, 5 free sepals; each sepal is linear-lanceolate and 0.5-2 cm long. The yellow to orange corolla is tubular, at least 5 cm long and broad, 5-parted with reflexed petals that are ovate, apically obtuse, and marginally rugose. The fruits are berries. The seed is endospermous.

39. *Achyranthes aspera* L.

Family: Amaranthaceae

Habit: Herb

Useful Parts: Leaves and Root

Uses: It is used in the treatment of asthma, in facilitating delivery, bleeding, bronchitis, debility, dropsy, cold, colic, cough, dog bite, snake bite, scorpion bite, dysentery, earache, headache, leukoderma, renal complications, pneumonia, and skin diseases

Description: It is a wild, perennial, erect herb. Stem is herbaceous but woody below, erect, branched, cylindrical, solid, angular, hairy, longitudinally striated, nodes and internodes are prominent, green but violet or pink at nodes. Leaves are ramal and cauline, simple, exstipulate, opposite decussate, petiolate, ovate or obovate, entire, acute or acuminate, hairy all over, unicostate reticulate. A spike with reflexed flowers arranged on long peduncle. Flowers are Bracteate, bracteolate, bracteoles two, shorter than perianth, dry, membranous and persistent, sessile, complete, hermaphrodite, actinomorphic, pentamerous, hypogynous, small, spinescent, green. Bracts ovate, persistent. Perianth made up of 5 tepals, polyphyllous, imbricate or quincuncial, green, ovate to oblong, persistent. Androecium made up of 10 stamens, out of which 5 are fertile and 5 are scale-like, fimbriated, sterile staminodes, both alternating with each other, fertile stamens are antiphyllous, monadelphous, filaments slightly fused at the base, dithecous, dorsifixed or versatile, introrse. Gynoecium is bicarpellary, syncarpous, superior, unilocular, ovule one, basal placentation, style single and filiform, stigma capitate. Fruit is oblong utricle. Seeds are endospermic.

41. *Cardiospermum halicacabum* L.

Family: Sapindaceae

Habit: Climber

Useful Parts: Leaves, Fruit, Seed

Uses: This plant is used for the treatment of rheumatism, abdominal pain, orchitis, dropsy, lumbago, skin diseases, cough, nervous disorders, and hyperthermia.

Description: It is a woody annual, many-branched vine with bi-fid (forked) axillary tendrils that are used for climbing. Leaves are alternate and twice ternately compound. Leaflets bear toothed margins, are lanceolate in shape, 2-4cm in length, 1-2cm wide, and faintly pubescent with pinnate venation. Irregular flowers are borne in panicles. Each flower bears four sepals, two large and two small, four whitish petals 4mm long, and eight stamens. Petaloid appendages are at the base of each flower. The 3-celled ovary bears one ovule per cell. Flowering time from July to August. Pollination is by entomophilly. Fruit is capsule. Seed is endospermous.

42. *Physalis peruviana* L.

Family: Solanaceae

Habit: Herb

Useful Parts: Fruits and Leaves

Uses: It is used to cure cancer, malaria, hepatitis, asthma, dermatitis and rheumatism.

Description: The Cape gooseberry is a perennial plant but is commonly grown as an annual in temperate climates. The simple velvety leaves are roughly heart-shaped and usually have entire (non-toothed) margins. The creamy yellow flowers are solitary and somewhat bell-shaped with five fused petals, each with a brown or purple spot at the base. Like the tomatillo, to which it is related, the plant is noted for the inflated bag like calyx (fused sepals) that encloses a fleshy orange berry.

43. *Physalis angulata* L.

Family: Solanaceae

Habit: Herb

Useful Parts: Leaves and Fruit

Uses: The plant extract is used in the treatment of dermatitis, hepatitis, rheumatism, malaria and asthma

Description: The plant is erect. It forms a small bush, abundantly branched, which can reach 90 cm high. The plant has a taproot system. The stem is hollow and polygonal. It is totally glabrous. The leaves are simple and alternate. They are carried by petiole, 3 to 5 cm long. The lamina is oval to elliptical, apiculate at the apex and bottom attenuated in sharp corner. It is 7 to 12 cm long and 3 to 6 cm wide. The margin is sinuous and irregularly serrated, provided some short, white hairs. The leaf blade is marked with 4 to 6 pairs of pinnate venations. Both sides are glabrous, although some short white hairs are present along the veins of the lower face. The flowers are solitary and axillary, located at the intersection of the branches of the plant. The flowers are supported by a glabrous peduncle, 7 to 10 mm long. The calyx, 3mm long, is composed of 5 sepals fused at the section of the base and ending in 5 triangular tines. Campanulate corolla, consisting of 5 fused petals. It is 7 to 8 mm large and creamy white with a purple spot at the base of the petals. 5 stamens are inserted into the corolla tube alternating with petals. The ovary has 2 loculus with many ovules. The fruit is berry. The seeds are flat and lenticular.

44. *Solanum pimpinellifolium* L.

Family: Solanaceae

Habit: Herb

Useful Parts: Fruits and Roots.

Uses: The fruits have medicinal uses as well. It can be used as first aid treatment for burns, scalds, and sunburn. It is also used in the treatment of rheumatism and headaches. Root decoction is ingested to relieve toothache.

Description: Annual or biennial herbs, undergoing secondary growth at the base; branches extremely slender and vining, extending up to 3m from centre. Stem erect initially, later procumbent or decumbent, sparsely pubescent or nearly glabrous.

Leaves are imparipinnately compound. Calyx 0.4-1.0 cm in diameter, pubescent with long and short, simple, uniseriate trichomes; tube less than 0.5 mm; lobes to 5 mm, linear, the apex acute. Corolla 1.6-3 cm in diameter, bright yellow; tube minute, the corolla often divided almost to the base; lobes 0.7-1.2 x 0.2-0.5 cm, four times as long as wide, narrowly lanceolate, strongly reflexed at anthesis. Staminal column 6-8 mm, narrowly cone shaped; filaments 1-2.5 mm; anthers 3.5-5 mm, the sterile tip approximately half the total anther length. Ovary conical, minutely glandular-villous; style 7-10 mm, usually exerted from the staminal column; stigma minute.

45. *Solanum linnaeanum* Happer

Family: Solanaceae

Habit: Shrub

Useful Part: Fruits.

Uses: The leaves are aphrodisiac, ophthalmic. They are used as a treatment for insomnia and for stopping excessive menses. An infusion of the leaves is used as an eye wash for cleaning the eyes.

Description: This perennial plant has attractive flowers each with five purple petals and small yellow stigmas, but it also has vicious thorns. The previous year's fruits can sometimes be seen ripening while the new season's flowers are attracting insect pollinators. The fruits are typically 3cm to 5cm across and look very much like unripe tomatoes; they appear in late summer; turning gradually from green to bright yellow and eventually black. The fruits are deadly poisonous and should never be eaten.

46. *Solanum trilobatum* L.

Family: Solanaceae

Habit: Slender Prickly Shrub

Useful Parts: Leaves, Petioles and Fruit

Uses: The leaves, petioles, and fruit of Solanum Trilobatum are utilised for many reasons in south India, which is economically significant. In some areas of India, it is also consumed to cure gastric complaints.

Description: It is a slender prickly scrambling shrub with prickles being curved, broad-based, yellowish and numerous along the stems, otherwise almost glabrous. Leaves are rounded-ovate in outline, obtusely lobed, 2-7 cm long, slightly stellate, with a few prickles along the petiole and midrib. Inflorescences are extra-axillary, peduncle short, 3-9-flowered; pedicels widely divergent. Calyx 3 mm long, with narrow teeth, sparsely stellate. Corolla is deeply lobed, stellate-pubescent outside, purple, reflexed.

47. *Capsicum annuum* L.

Family: Solanaceae

Habit: A small perennial herb

Useful Parts: Leaves and Fruits

Uses: Capsicum (Capsicum annuum), also known as cayenne pepper, has been used orally for upset stomach, toothache, poor circulation, fever, hyperlipidaemia, and heart disease prevention.

Description: The 'Anaheim Chili' has fruits about 7 inches long, 1½ inches in diameter, slightly tapered, stem end usually without pronounced shoulder but often wrinkled or folded. Flavour is mildly pungent as compared with other chili varieties. Anaheims take about 115 days to green mature and 150 days to red ripe and are also called 'California Chili.' 'College No. 9 Chili,' also called 'New Mexico 9,' has fruits about 5 inches long, 1¾ inches in diameter, tapered and pointed, shoulders sloping and usually smooth. These are less pungent than 'Mexican Chili,' but slightly more pungent than 'Anaheim,' with about the same maturity period as for 'Anaheim.' Mexican, or "native" chili has fruits about 3 inches by 1½ inches, somewhat conical, tapering to a blunt point. Pods generally have a deep shoulder at the stem and are often furrowed and wrinkled. Mexican chilis are the most pungent of the large-fruited

chilis and strains are widely grown in the Southwest, and in central and northern Mexico, where they are preferred for earliness

48. *Datura matel* L.

Family: Solanaceae

Habit: A perennial shrub

Useful Parts: leaves, fruits, flowers, stem or roots

Uses: The bitter narcotic plant relieves pain and encourages the healing process. The seeds of the plant are medicinally the most active. Externally, the plant is used as a poultice in treating fistulas, abscesses wounds and severe neuralgia.

Description: *Datura metel* is a shrub-like annual (zone 5–7) or short-lived, shrubby perennial (zone 8–10), commonly known in Europe as Indian thorn apple, Hindu Datura, or metel and in the United States as devil's trumpet or angel's trumpet. *Datura metel* is naturalized in all the warmer countries of the world.

49. *Solanum incanum* L.

Family: Solanaceae

Habit: An erect prickly shrub

Useful Part: Fruits

Uses: *Solanum incanum* is the traditional medicinal plants widely used to treat various types of ailments like sore throat, stomach-ache, head-ache, painful menstruation, liver pain, malaria, hypertension, stomach problem, asthma, diabetes, common cold and pain caused by onchocerciasis, pneumonia and rheumatism**.**

Description: Erect or spreading shrub up to 3 m tall, occasionally a small tree; stems and leaves with stellate hairs and pale yellow to brown prickles, up to 1 cm long. Leaves alternate, simple; stipules absent; petiole 0.5–8.5 cm long; blade almost round to lanceolate, 1–30 cm × 1–17 cm, base rounded, truncate or cordate, often unequal, apex acute or obtuse, margin entire to pinnately lobed, densely hairy. Inflorescence a 2–15-flowered cyme, inserted above the leaf axil. Flowers bisexual or functionally

male, nodding or pendent, regular, (4–)5–7(–9)-merous; pedicel 0.5–4 cm long; calyx campanulate, lobes up to 1.5 cm long, enlarging and splitting in fruit; corolla campanulate to rotate, 1–4.5 cm in diameter, with ovate or broadly triangular lobes, blue, pink, purple or violet, rarely white; stamens inserted near the base of the corolla tube and alternating with corolla lobes, filaments short, anthers slender; ovary superior, 2(–4)-celled, style up to 15 mm long, densely hairy. Fruit a globose or depressed globose, occasionally ovoid-ellipsoid berry 2.5–3.5 cm × 2–3 cm, yellow, orange or brown when ripe, many-seeded. Seeds lentil-shaped to almost kidney-shaped, up to 3.5 mm × 3 mm, pale yellow to brown. Seedling with epigeal germination.

50. *Solanum melongena* L.

Family: Solanaceae

Habit: Perennial Shrub

Useful Part: Fruit

Uses: Various plant parts are used in decoction, as powder or ash for curing ailments such as diabetes, cholera, bronchitis, dysuria, dysentery, otitis, toothache, skin infections, asthenia and haemorrhoids. Eggplant is also ascribed narcotic, anti-asthmatic and anti-rheumatic properties.

Description: Eggplant is an annual or short-lived perennial plant that is very sensitive to cold temperatures. It grows fastest when the temperature ranges between 70 and 85 degrees. It produces an edible shiny glossy fruit. The plant may grow 2 to 4 feet tall and is multi-branched. The leaves and stems have star-shaped hairs, and the small violet flowers are also star-shaped. Eggplant or Aubergine is a member of the Solanaceae or nightshade family which also includes tomatoes, potatoes, and peppers.

51. *Cyanthillium cinereum (L.) H. Rob.*

Family: Asteraceae

Habit:

Useful Part: Leaves

Uses: The juice of *Cyathillium cinereum* is given to children with urinary incontinence. The leaves are eaten as a potherb. A decoction of it is also given in diarrhea, stomachache and for cough and colic. Leaves have antibiotic properties.

Description: *Cyathillium cinereum* is an erect herb, 20 to 80 cm high, slightly branched and covered with fine grey hairs. The stem is finely striated. The leaves are alternate, simple. They are elliptical, attenuate base in corner and covered with a greyish hair. The flowers are grouped in small purplish heads, assembled in a loose inflorescence. The fruits are carriers of small tufts of white hairs.

52. *Passiflora foediax* L.

Family: Passifloraceae

Habit: Climber

Useful Parts: Fruit and Leaves

Uses: It is used to relieve sleeping problems as well as used in the treatment of itching and coughs.

Description: The stems have an unpleasant odour and vary in hairiness from almost hairless to having a sparse or dense covering of white, yellow or golden-brown sticky hairs. At the base of each leaf stalk there is a tendril and a 1 cm long threadlike appendage covered in sticky glands. The leaves most often have three rounded or pointed lobes, but sometimes they can be entire or five-lobed. These leaves are alternately arranged along the stems and borne on stalks 1-6 cm long. They are hairy on both surfaces, with the hairs along their margins often being sticky. The flowers vary from pinkish to white or purplish in colour and are borne singly in the leaf forks on stalks 2-4.5 cm long. They are surrounded by three deeply-divided bracts that are densely covered in large sticky (i.e. glandular) hairs. Each flower has five sepals and five petals. They also have five stamens, with anthers 4-5 mm long, and an ovary topped with three style tipped with prominent stigmas. Flowering occurs mainly during autumn, winter and spring. The fruit are dry berries partially enclosed by the persistent, deeply-divided, sticky bracts. These fruits are somewhat hairy and turn from green to yellow or orange in colour as they mature**.**

54. *Leucas aspera* (Willd.) Link

Family: Lamiaceae

Habit: annual herb

Uses: The entire plant is also used as an insecticide and indicated in traditional medicine for cough, cold, painful swelling and chronic skin eruption, wound healing.

Useful Parts: Leaves, Root and Stem

Description: *Leucas aspera* is an annual, branched, herb erecting to a height of 15-60 cm with stout and hispid acutely quadrangular stem and branches. Leaves are sub-sessile or shortly petiolate, linear or linearly lanceolate, obtuse, pubescent up to 8.0 cm long and 1.25 cm broad, with entire or crenate margin; petiole 2.5-6 mm long; flowers white, sessile small, in dense terminal or axillary whorls; bracts 6 mm long, linear, acute, bristle-tipped, ciliate with long slender hairs; calyx variable, tubular, 8-13 mm long; tube curved, contracted above the nutlets, the lower half usually glabrous and membranous, the upper half ribbed and hispid.

55. *Ocimum tenuiflorum*

Family: Lamiaceae

Habit: Perennial Herb

Useful Parts: Leaves, Roots and Seeds

Uses: It heals the bacterial borne diseases as well as against Aspergillosis and other fungal borne diseases

Description: Holy basil is an erect, many-branched subshrub, 30–60 cm (12–24 in) tall with hairy stems. Leaves are green or purple; they are simple, petiole, with an ovate blade up to 5 cm (2 in) long, which usually has a slightly toothed margin; they are strongly scented and have a decussate phyllotaxy. The purplish flowers are placed in close whorls on elongated racemes.

56. *Mesosphaerum suaveolens*

Family: Lamiaceae

Habit: Shrub

Useful Parts: Leaves and Flower

Uses: Kuntze is a species widely used traditionally in the treatment of ailments, such as stomach pain, haemorrhoids, cough, ulcer, liver disease, fever, influenza, nasal congestion, and inflammation.

Description: Shrubs, to 1.5 m high; stem obtusely 4-angular, thinly hairy. Leaves ovate, acute, hispid below, glabrate above; petiole to 5 cm long. Flowers in clusters of 1-12; calyx tube 8 mm long, tubular, 10-ribbed, glandular hairy, teeth spinulose, 4 mm long; corolla 5 mm long, lobes short, glabrous inside, blue. Nutlets 4 x 2.5 mm, compressed, with a ridge on dorsal surface, pubescent, deep brown, mucilaginous when wet.

57. *Ocimum kilimandscharium* Baker ex Gurke

Family: Lamiaceae

Habit: Shrub

Useful Parts: Leaves and flowers

Uses: Traditionally, extracts of *Ocimum kilimandscharium* were used to alleviate many ailments in East Africa including treatment of colds, coughs, abdominal pains, measles, diarrhea, insect repellent, particularly against mosquitoes and storage pest control.

Description: *Ocimum kilimandscharium* Guerke (Syn. *Ocimum camphora* Guerke) belongs to family Lamiaceae. It is a native of Kenya and distributed in East Africa, India, Thailand, Uganda and Tanzania. It is extensively grown in the Tropics. In India it is cultivated on a small scale especially in West Bengal, Assam, Tamil Nadu, Karnataka, Kerala and Dehradun. Commonly the plant is called as camphor Basil, African blue basil and in Ayurveda as Karpura Tulasi**.** Morphologically, it is a perennial aromatic evergreen undershrub with pubescent branchlets having pale green leaves which are glandular, ovate or oblong in shape, base is acute, deeply serrated, pubescent on both surfaces, oppositely arranged and about 3-7 cm in length including

petioles which are 4 to 12 mm long, 1 to 2.5 cm wide; indumentum of long white hairs or sometimes glabrous above; petiole 4-10 mm. Stems are brownish green, round-quadrangular, much branched, woody with epidermis sometimes peeling off in strips below, arising from a large woody rootstock, with white glandular hairs, becoming denser in the inflorescence-axis, with sparse sessile glands.

58. *Merremia disseca* (Jacq.) Hallier f.

Family: Convolvulaceae

Habit: Climber

Useful Parts: Leaves and Stem

Uses: An infusion of the leaves is taken as a sedative in the treatment of chest complaints A cold infusion is a remedy for giddiness, snake bites or intoxication. A hot infusion is taken to relieve urinary infection A decoction of the whole plant, used as a wash, is an effective remedy for scabies and itch. A poultice of crushed fresh leaves is applied as a resolutive and sedative for treating inflammations

Description: Vines; the stems herbaceous, sparsely hirsute to glabrous. Leaves alternate, palmately divided almost to the base, the 7-9 lobes sinuate-dentate, usually glabrous, the entire leaf suborbicular in outline. Inflorescence axillary, 1-to few-flowered, cymose, the peduncles 5-10 cm long, hirsute, glabrescent on the upper portion. Pedicels 1.5-2 cm long, thickened toward the apex, glabrous. Sepals oblong, 18-25 mm long, mucronate, glabrous. Corolla 3-4.5 cm long, white with a purple centre, broadly campanulate. Fruits capsular, depressed-globose, 1-2 cm in diameter, subtended and partially surrounded by the accrescent calyx; seeds black, sub rotund, glabrous.

59. *Merremia aegyptia* (L.) Urb.

Family: Convolvulaceae

Habit: Annual climber

Uses: It is used to treat deobstruent, diuretic, rheumatism, neuralgia, cancerous wounds, migraine, purgative, snake bites, ulcer

Useful Part: Stem and Leaves**.**

Description: A robust annual twiner occurring throughout the Region and widespread in the Tropics. Horses will not graze it; other stock in Senegal may or may not take it. The dried leaves are applied in Nigeria as a dressing for burns. The stems are used in Senegal and probably elsewhere as ties.

60. *lpomoea aquatic* Forsk.

Family: Convolvulaceae

Habit: Perennial herb

Useful Parts: Leaves and Flowers

Uses: It is used to treat piles, and nosebleeds, as an anthelmintic, and to treat high blood pressure.

Description: Water Morning Glory is a semi-aquatic tropical plant grown as a leaf vegetable. Its precise natural distribution is unknown due to extensive cultivation, with the species found throughout the tropical and subtropical regions of the world. Water Morning Glory grows in water or on moist soil. Its stems are 2-3 m or more long, hollow, allowing them to float, and these root at the nodes. The leaves vary from sagittate (typical) to lanceolate, 5-15 cm long and 2-8 cm broad. The flowers are trumpet-shaped, 3-5 cm diameter, usually white in colour, with a purple center. It is most commonly grown in East and Southeast Asia. Because it flourishes naturally in waterways and does not require much if any care, it is used extensively in Malay and Chinese cuisine.

61. *Ipomoea quamoclit* L

Family: Convolvulaceae

Habit: A beautiful annual twiner with slender stems.

Useful Parts: Flowers and Stem

Uses: In the Philippines, leaves are used as poultices for bleeding hemorrhoids. Crushed leaves used for carbuncles. Seeds reportedly used as laxative by the Sino-Annamites.

Description: *I. quamoclit* is a herbaceous, twining vine growing up to 3–10 feet (0.91–3.05 m) tall. The leaves are 1–4 inches (25–102 mm) long, deeply lobed (nearly pinnate), with 9-19 lobes on each side of the leaf. The flowers are 1–2 inches (25–51 mm) long and 1 inch (25 mm) in diameter, trumpet-shaped with five points, and can be red, pink or white. *I. quamoclit* is herbaceous, twining vine growing up to 3–10 feet (0.91–3.05 m) tall. The leaves are 1–4 inches (25–102 mm) long, deeply lobed (nearly pinnate), with 9-19 lobes on each side of the leaf. The flowers are 1–2 inches (25–51 mm) long and 1 inch (25 mm) in diameter, trumpet-shaped with five points, and can be red, pink or white.

62. *Ipomoea* carnea Jacq.

Family: Convolvulaceae

Habit: Evergreen Shrub

Useful Parts: Leaves and Flower

Uses: The plant possesses anti-bacterial, anti-fungal, anti-oxidant, anti-cancer, anti-convulsant, immunomodulatory, anti-diabetic, hepatoprotective, anti-inflammatory, anxiolytic, sedative and wound healing activities. However, some toxicological effects have been also reported**.**

Description: *Ipomoea carnea*, the pink morning glory, is a species of morning glory that grows as a bush. This flowering plant has heart-shaped leaves that are a rich green and 6–9 inches (15–23 cm) long. It can be easily grown from seeds. Erect (subsp. *fistulosa*) or climbing (subsp. *carnea*) undershrub to 4 m, often growing in clumps, stems stout, hollow, canescent when young, becoming glabrous. Leaves petiolate, 8 - 20 (- 30) x 3 - 10 (- 12) cm, ovate or elongate-ovate-deltoid, base cordate to sub truncate with rounded auricles, apex acuminate to long-acuminate, both surfaces grey-canescent when young, veins prominent abaxially; petioles 3 - 8 cm. Inflorescence of long-pedunculate axillary, somewhat compact cymes.

63. *Barleria volkensii* Lindau

Family: Acanthaceae

Habit: Herb

Uses*:* Used to reduce inflammation caused by insect bites, snake bites, boils, and rheumatism.

Useful Parts: Leaves and Stem.

Description: It grows as a shrub 60 –100 cm tall. The leaves are dark green on the upper surface and pale green on the lower surface. They are elliptic to narrowly ovate. The flowers are about 5 cm long, funnel-shaped in violet, pink, or white color. The fruits are about 1.5 cm long ellipsoid capsules. They become glabrous and glossy at maturity. It has spikelet inflorescence. In dense spikes, bracteoles prominent, linear. Calyx of 4 unequal sepals white. Corolla blue, funnel-shaped, lobes 5, unequal. Stamens 4, only 2 fertile, disk cup-like. Ovary bicelled, 4 ovuled. The fruit is capsule.

64. *Hygrophila auriculata* (K. Schum.)

Family: Acanthaceae

Habit: A herb growing in wet places

Useful Part: Leaves and Flowers

Uses: Kokilaksha, as it is known in sanskrit, was extensively used in Ayurvedic system of medicine for various ailments like rheumatism, inflammation, jaundice, hepatic obstruction, pain

Description: Marsh Barbel is a stout aquatic perennial herb, 1-2 m high. Erect unbranched stems are hairy near swollen nodes. Densely hairy, lance-like, stalkless leaves, 10-15 cm long, occur in whorls of 6 at each node on the stem. Straight, yellow, 4 cm long spines are present in the axil of each leaf. Flowers occur in 4 pairs at each node. The 3 cm long purple-blue flowers are 2-lipped - the upper lip is 2-lobed and the lower one 3-lobed with lengthwise folds. Flowers open in opposite pairs. Flowering: October

65. *Dicliptera paniculata* (Forssk.) I. Darbush

Family: Acanthaceae

Habit: Erect herb

Useful Parts: Stems and leaves

Uses: It heals the bacterial borne diseases as well as against Aspergillosis and other fungal borne diseases.

Description: Panicled Foldwing is an erect herb, 0.6-1.2 m tall. Young shoots are usually 4-sided; adult shoots 6-sided, white spreading bristle-hairy. Ovate leaves opposite, equal and unequal; leaf-stalk 3-5 mm. Smaller ones 0.8-1.2 x 3-5 mm, larger ones 3-4.5 x 1.5-2 cm, densely hairy and prominently so on veins. Flowers are borne at branch-ends or in leaf-axils, with leaves forming a large lax panicle. Pink flowers, to 1 cm, 2-lipped - lower lip spreading, upper lip erect. Stamens 2; filaments distinct, to 5 mm, white hairy. Panicled Foldwing is found in India, Myanmar and Tropical Africa. It is also found in the Himalayas and Western Ghats, at altitudes of 600-2200 m. Flowering: October-February.

66. *Pedalium mure* L.

Family: Pedaliaceae

Habit: Herb

Useful Parts: Leaves and Fruit

Uses: Based on traditional healers, the plant Pedalium murex L. was used for the dissolution and prevention of kidney stone formation. Further, it is used for the treating ailments like incontinence of urine, gonorrhea, anti-bilious agent, dysuria and control white discharge.

Description: A diffuse annual, much branched, spreading, succulent, glandular, up to 60 cm tall. Roots similar to turmeric in colour. Leaves simple, opposite, ovate or oblong-obovate, 1-4.5 cm long, 0.5-3 cm broad, truncate or obtuse, irregularly and coarsely crenate-serrate, glabrous above, minutely scaly below, petiole-l-4 cm.

Flowers 1.5-2 cm across; pedicel 1-2 mm long, increasing up to 4 mm in fruit. Calyx c.2 mm long; teeth linear, scaly outside, persistent. Petals connate into a broad tube, 1-3 cm long; lobes obtuse. Stamens included, 0.5-1 cm long; filaments dilated, glandular hairy at the base; anthers kidney shaped. Fruit indehiscent, abruptly narrowed at the base and with a patent spreading spine at each basal corner of the broader part, 1-1.8 cm long, 0.5-1 cm broad, spine 2-4 mm long. Seeds 2 or 1 per locule, oblong.

67. *Ludwigia octovalvis* Linn.

Family: Onagraceae

Habit: Herbaceous shrub

Useful Part: Leaves

Uses: It is commonly consumed as a health drink and traditionally used for treating various ailments such as dysentery, diarrhea, diabetes and headache.

Description: A herbaceous shrub, it can grow up to 2m tall and has a branched growth form. Leaves are hirsute, long and narrow with sunken venation and a reddish to pale midrib. The stems grow up to 1cm in diameter and are hirsute and ribbed. The flower is composed of 4 obovate petals (0.2 - 0.4 cm long) arranged in a cross-like pattern. The tip of the petal is sometimes folded down, forming a large notch-like indentation. The petals have distinct sunken venation which creates a rippled surface. The fruit is a capsule about 2 to 4.5cm long with persistent sepals at the apex of the fruit, it splits into 8 linear lobes, releasing wedge-shaped seeds dispersed by water.

68. *Ludwigia palustris* (L.) Elliott.

Family: Onagraceae

Habit: Perennial herb

Useful Parts: Leaves, Roots and Seeds

Uses: Used in the treatment of phthisis (pulmonary tuberculosis), asthma and chronic coughs.

Description: Water Purslane is a mat-forming wet area plant in the primrose family. It is found growing in low water situations along small ponds or streams or in muddy areas. It is also used in aquariums. The stems are succulent and often red in color. The inconspicuous flowers occur mid-summer to fall. This plant may be seed near a pond or grown in rain or water gardens. When growing in mud the plant sprawls along the ground with just the branch tips upright. In the water, branches ascend towards the surface with just the tips out of water. Both in the water and out, roots are continuously forming at the nodes, forming mats. It can be considered weedy but can also be a stabilizing plant on muddy banks, and is used to filter and take in toxins in bioswales and ditches.

69. *Heliotropium indicum* L.

Family: Boraginaceae

Habit: Perennial herb

Uses: The leaf juice is used to treat the stings and boils of scorpions and insect bites. On the other hand, the boiled juice with castor oil is used to treat mad dog bite infections, rheumatism, ulcer, venereal disease, fever, sore throat, and sores in the rectum.

Useful Part: Leaves

Description: Indian heliotrope is an annual, erect, branched plant that can grow to a height of about 15–50 cm (5.9–19.7 in). It has a hairy stem, bearing alternating ovate to oblong-ovate leaves. It has small white or purple flowers with a green calyx; five stamens borne on a corolla tube; a terminal style; and a four-lobed ovary.

70. *Stemodia durantifolia* (Linn.)

Family: Scrophulariaceae

Habit: Shrub

Useful Part: Leaves

Uses: It is used to prepare many medicinal drugs.

Description: Stemodia durantifolia is a perennial herbaceous plant to 100 cm (39 in) tall, with glandular-hairy herbage. The branching habit is both basal and axillary. The leaves are lanceolate and subsessile, and toothed along their edges. The terminal inflorescence is an ascending, spike-like structure. The bracts are equal to or slightly longer in length to the flowers or more or less equal to the length of the sepals. The flowers have a violet corolla. The sepals measure equal to or longer than half of the length of the corolla.

72. *Corchorus trilocularis* L.

Family: Tiliaceae

Habit: Annual herb

Uses: It is used to treat syphilis

Useful Part: Leaves

Description: Wild Jute is a branched annual herb, up to 1 m tall, usually erect, sometimes found prostrate due to browsing by cattle. Young branches are purplish, sparsely hairy. Leaves are oblong to lance shaped, up to 12 × 3.5 cm, hairless or hairy, particularly on the veins. Margins are toothed with a long bristle on the 2 lowermost teeth. Flowers are borne in in 1-3 flowered leaf-opposed clusters. Flowers are yellow, with sepals narrowly lance shaped, as long as the petals. Petals are 4-5, 5-7 mm long, 2-2.5 mm wide, obovate tapering to a short ciliate claw. Stamens are many. Fruit is a slender more or less erect, cylindric, many-seeded capsule, straight or slightly curved, up to 7 cm long, 3-4-angled with a rough surface. The species name trilocularis comes from the three-chambered ovary. Young tender leaves are cooked and eaten.

73. *Cleoserrata speciosa* (Raf.) Iltis

Family: Cleomaceae

Habit: Perennial herb

Useful Part: Leaves

Uses: It was used for many therapeutic potentials and it was used to make many medicines based on its anti-inflammatory properties.

Description: Showy Spider Flower is a herbaceous plant resembling Wild Spider Flower, but with larger, more showy flowers. It was introduced as an ornamental plant, but has escaped from cultivation, and can be seen growing wild. It is an annual herb, growing up to 1.5 m in height, with hairless stem and leaves. Leaves are palmately compound, alternately arranged, elliptic leaflets 2-16 cm long. It can flower any time after maturity. Flowers are many, borne in an erect, showy raceme, at the end of the stem. Each flower is subtended by a small leaf-like bract. Flower has 4 free, inverted-lance shaped, pink to white petals, 2.8-3.8 cm long. Flowers have 6 stamens, on filaments that are 3-6 cm long! Fruit is a linear capsule up to 8 cm long, on a long stalk. The plant is propagated by seed.

74. *Cleome viscosa* L.

Family: Capparidaceae

Habit: Herb

Useful Parts: Stem, Leaves and Flowers

Uses: The leaves are diaphoretic, rubefacient and vesicant. They are used as an external application to wounds and ulcers. The juice of the leaves has been used to relieve earache. The seeds are anthelmintic, carminative, rubefacient and vesicant.

Description: Calyx composed of 4 sepals, polysepalous, valvate, sepals arranged in two whorls of two each. Corolla composed of 4 petals, polypetalous, clawed, imbricate, yellow coloured. Androecium made up of many stamens (12-24), polyandrous, dithecous, basifixed, introrse, filament long and anther lobes curved. Gynoecium bicarpellary, syncarpous, superior, single locule with many ovules, parietal placentation, gynophore is present but very short, style short, stigma sticky and capitate.

76. *Commelina communis* L.

Family: Commelinaceae

Habit: Annual herb

Useful Parts: Leaves, Roots and Flowers.

Uses: It is used to heal swelling, treatment of urinary tract infection and respiratory tract infections, diarrhea, enteritis, and hemorrhoids. The plant has also been used in fever, malaria, insect, bug bites, rheumatoid arthritis, gonorrhea, influenza, and bladder infection.

Description: It is an annual herb. The stem is erect. The leaves are sessile. The flowers are arranged on inflorescences called cincinni, which are also called Scorpioid cymes. This is a form of a monochasial where the lateral branches arise alternately. The cincinni are subtended by a spathe, a modified leaf. There are three fertile stamens, meaning they are on the lower part of the flower, and three infertile stamens, meaning they are on the upper part of the flower. The fruit is a dehiscent, ellipsoid capsule with two locules each containing two seeds. The capsule is glabrous, brown, measures 4.5–8 mm (0.18–0.31 in) long, and dehisces into two valves.

77. *Argemone mexicana* L.

Family: Papaveraceae

Habit: Annual herb

Useful Parts: Leaves, Flowers, Stem and Roots.

Uses: It is used to treat tumours, warts, skin diseases, inflammations, rheumatism, jaundice, leprosy, microbial infections, and malaria**.**

Description: Annual, prickly herb with yellow latex and branched tap root. Stem is erect, branched, woody at the base, solid, cylindrical, spinous, contains yellow latex. Leaves are ramal and cauline, exstipulate, alternate, simple, subsessile, semi - amplexicaul, margin lobed and spinous, apex acute, unicostate reticulate venation, both the surfaces are covered with many spines. It shows Solitary terminal inflorescence. Flowers are ebracteate, pedicellate, complete, actinomorphic, hermaphrodite, hypogynous, yellow in colour. Calyx made up of 2 sepals, polysepalous, spinous, caducous, bear a clear horn like outgrowth at their apex,

twisted or imbricate, inferior and green. Corolla made up of 6 petals, arranged in two whorls of three each (3+3), polypetalous, twisted or imbricate, yellow, each petal is obovate, caduceus, inferior. Androecium made up of numerous stamens, polyandrous, arranged in many whorls, filament long, slender and yellow, anther dithecous, basifixed and extrorse, dehiscence longitudinal. Gynoecium tetra to Hexa carpellary, syncarpous, superior, ovary covered with prickles, unilocular, many anatropous ovules on each placenta, parietal placentation, style reduced with 4 to 6 stigma lobes red. Fruit is prickly capsule.

78. *Impatiens balsamina* L.

Family: Geraniaceae

Habit: Annual herb

Useful Parts: Flowers and stem

Uses: It is used for the treatment for the treatment of rheumatism, isthmus, generalized pain, fractures, inflammation of the nails, scurvy, carbuncles, dysentery, bruises, foot diseases.

Description: Garden Balsam is most common Balsam grown as a garden plant in India. It is an annual plant growing up to 20-75 cm tall, with a thick, but soft stem. The leaves are spirally-arranged, 2.5-9 cm long and 1-2.5 cm broad, with a deeply toothed margin. The flowers are pink, red, mauve, lilac, or white, and 2.5-5 cm diameter. They are pollinated by bees and other insects, and also by nectar-feeding birds. Flower-stalks are up to 1-5 cm long. Lateral sepals are about 2-3 mm long, ovate, sparsely ciliate, lower sepal conical, spur 1-2 cm long, curved inwards. The ripe seed capsules explode when touched, inspiring the genus name Impatiens. Garden Balsam is native to Western Ghats India and SE Asia.

79. *Phyllanthus polygonoides* Nutt.

Family: Euphorbiaceae

Habit: Herb

Useful Parts: Leaves and flower

Uses: In Indian ayurvedic medicine, various herbaceous *Phyllanthus* species are prescribed for jaundice, gonorrhea and diabetes as well as for making poultices for skin problems. Infusions from young shoots are used to treat chronic dysentery.

Description: *Phyllanthus polygonoides*, known as smartweed leaf-flower or knotweed leaf flower, is an herbaceous perennial plant in the family Phyllanthaceae. It grows from 10 to 50 centimeters in height. It is native to the United States northern and central Mexico. Smartweed leaf-flower grows in a variety of habitats throughout its range, including grasslands, shrublands), and glades in forests. It is often associated with limestone and calcareous soils. It is morphologically similar to the closely related species, pinewoods dainties (*Phyllanthus liebmannianus*).

80. *Turnera ulmifolia* L

Family: Turneraceae

Habit: Herb

Useful Parts: leaves and Flower

Description: *Turnera ulmifolia* grows erect, with dark toothed leaves and small, yellow-orange flowers, and is often found as a weed growing on roadsides. These yellow flowers bloom around 6:00 am and wilt around 11:30 AM. Life span for flower is around 6 hours. These plants can survive on minimum water and grow on walls, cement blocks, and rocks. Tawny Coster (*Acraea terpsicore*) butterfly larvae feed on these plants. This plant is commonly misidentified with the closely related *T. diffusa* in horticultural commerce, causing it to be often misrepresented as "Damiana.

82. *Jasminum sambac* (L.) Aiton

Family: Oleaceae

Habit: Shrub

Useful Parts: Leaves

Uses: Traditionally, *Jasminum sambac* has been used to treat dysmenorrhea, amenorrhea, ringworm, leprosy, skin diseases and also as an analgesic, antidepressant, anti-inflammatory, antiseptic, aphrodisiac, sedative, expectorant.

Description: An evergreen vine or shrub, Jasminum sambac can grow to a height of 1.6 to 9.8 feet. Due to auto polyploidy, natural hybridization, and spontaneous mutation, the species is exceedingly varied. The majority of Jasminum sambac plants grown for commercial purposes do not generate seeds; instead, cuttings, layering, marcotting, and other asexual propagation techniques are used to propagate the plant.

83. *Ammannia coccinea* Rottb.

Family: Lythraceae

Habit: Perennial herb

Useful Parts: Leaves and Roots

Uses: It was used to treat scabies, ringworm, parasitic skin infections, common cold, typhoid, strangury, spinal disease, gastroenteropathy and aphrodisiac**.**

Description: It was a perennial herb. Stem is erect and branched. Leaves are opposite, the pairs set at right angles to the ones above and below, ¾ to 4 inches (to 10 cm) long, ¼ to about ½ inch (to 15 mm) wide, pointed to blunt at the tip, toothless, hairless, stalkless, mostly green or tinged red, the lowest leaves more oblong-elliptic becoming lance-linear up the stem. flowers are stalkless or nearly so, the stalks less than .5 mm long. Each flower is ¼ inch across or less, has 4 (rarely 8) pink to lavender, wavy petals 2 to 4 mm long with 4 to 8 light yellow tipped stamens surrounding a stout green style in the center. The calyx cupping the flower is cylindrical, less than ¼ inch (3 to 5 mm) long, the sepal edges fused and broadly triangular at the tip. Fruit is a round capsule 4 to 6 mm (to ¼ inch) in diameter, as long as or slightly longer than the persistent calyx at maturity. Capsules have 4 chambers and turn red as seed ripens.

84. *Scleromitrion diffusum* (Willd.) R. J. Wang

Family: Rubiaceae

Habit: Herb

Useful Parts: Flower and Leaves

Uses: Spreading Diamond Flower is being actively studied for its role in treating cancer.

Description: Annual herbs. Stems erect to semi-protrate. Stipules 1-2.5 mm long. Leaves linear lanceolate, 30-50 x 3-6 mm. Petiole 0-1 mm. Inflorescence an axillary cyme. Corolla tube1-1.5 mm long. Capsule sub globose, 2-2.5 x 2-3 mm. Scleromitrion diffusum - Spreading Diamond Flower. Spreading Diamond Flower is a slender annual herb, rising up to prostrate, up to 50 cm tall; stems slightly flattened to round or young stems sometimes 4-angled, sparsely to densely finely velvet-hairy.

85. *Oldenlandia umbellate* L

Family: Rubiaceae

Habit: Herb

Useful Parts: Leaves

Uses: It is used in combination with other herbs for the treatment of hepatitis, snake bites and tumours of the liver, lung, stomach, and rectum

Description: Small diffuse herbs; stem terete, sparsely pubescent towards nodal regions. Leaves 1-1.5 x c. 1 cm, narrow, linear; stipules sheathing, shortly pectinate on the margins. Flowers axillary, solitary or binate, sessile to shortly pedicelled. Calyx tube sub globose, 1.5-2 cm; lobes triangular, to 0.1 cm long. Corolla white; tube 0.1-0.15 cm long; lobes 0.05 cm long. Style shortly exserted; stigma papillate. Capsule ovoid, 0.2 cm long, laterally compressed, dehiscing loculicidally at tip; seeds minute, angular, reticulate.

87. *Boehmeria cylindrica* (L.)

Family: Urticaceae

Habit: Annual perennial herb

Useful Parts: Whole Plant

Uses: Used to cure cancer

Description: Strongly toothed margins are evident on the 4 to 18 cm long, simple, opposite, rough, lance to oval-shaped leaves of this tall perennial herb (to 1.5 m). It has a single stem with tiny hair***s.***

88. *Digitaria sanguinalis* (L.)

Family: Poaceae

Habit: Annual herb

Useful Part: Leaves

Uses: It is used to treat anti-ulcer, anti-helminitic, anti-inflammatory, anti-diabetic, anti-depressant activity.

Description: Digitaria sanguinalis is a sparse, tufted decumbent annual grass with weak and spreading, hollow stems, to 1 m long or more, usually rooting at the lower nodes, the erect portions up to 60 cm tall. The sheaths are shorter than the internodes and pubescent. The ligule is a thin, truncate membrane, 1-2 mm long, hairless and irregularly dissected. The leaf blades are soft and flat, 5-10 mm wide, pubescent on one or both surfaces to nearly glabrous. The inflorescence consists of 4-9 erect or spreading, digitate racemes at the apex of the stem and sometimes below. The rachis is narrowly winged. Spikelet, 3 mm long, are born in pairs, one stalked and the other stalkless; they are green or purple-tinged. Fruit is glume.

89. *Orysa sativa* L.

Family: Poaceae

Habit: Annual to woody perennial herb

Useful Parts: Stem and Leaves

Uses: rice grain is applied to the skin to treat boils, sores, swellings, and blemishes, and sticky rice is used in remedies for stomach upsets, heartburn, and indigestion. Brown rice extracts have been used as remedies for breast and stomach cancer, as well as indigestion, nausea, and diarrhoea.

Description: Basal sterile florets similar; barren; without significant palea. Lemma of lower sterile floret lanceolate; 2-3 mm long; 0.25 (-0.5) length of spikelet; membranous; without lateral veins; emarginate. Lemma of upper sterile floret lanceolate; 2-3 mm long; 1 length of lower sterile floret; membranous. Fertile lemma elliptic; laterally compressed; 8-11 mm long; coriaceous; keeled; 5 -veined. Lemma midvein ciliate; hairy above.

90. *Brachiaria brizantha (Hochst. Ex A. Rich)*

Family: Poaceae

Description: It is a tufted perennial grass, usually 60-120 cm high (up to 200 cm), with deep roots (down to 2 m) and short rhizomes. It has stout, erect or slightly decumbent culms and bright green leaves. Inflorescence is a panicle consisting of 2-16 racemes, 4-20 cm long. Spikelets are usually on a single row, elliptical, 4-6 mm long with a sub-apical fringe of long purplish hairs.

Table 4: Informant Consensus Factor (ICF) Values by Category for Treating Various Diseases

S. No	Category	Plant Species Used and Number of Citations	Total number		ICF
			Species	Use citation	
1.	**Skin Disease (SD)**	*Abutilon indicum* (11), *Achyranthes aspera (23), Leucas aspera (17), Ocimum tenuiflorum (27), Solanum trilobatum (14), Ricinus communis (27), Phyla nodiflora (8), Cardiospermum halicacabum (10), Cleome viscosa (5), Cleome gynandra (9), Luffa cylindrica (8)*	11	159	0.94
2.	**Hair Problem**	*Ocimum tenuiflorum (13), Tridax procumbens (7), Ricinus communis (27)., Phyla nodiflora (21), Cassia angustifolia (9), Cardiospermum halicacabum (12),*	6	89	0.93
3.	**Cold (C)**	*Leucas aspera (22), Ocimum tenuiflorum (61), Solanum trilobatum (61), Solanum nigrum (15)*	4	159	0.98
4.	**Head ache (HA)**	*Abutilon indicum (8), Leucas aspera (22), Ocimum tenuiflorum (37), Solanum trilobatum (32), Solanum nigrum (13), Tribulus terrestris (8), Momordica cymbalaria (5), Cleome gynandra (4), Commelina communis (5), Argemone Mexicana (4)*	10	138	0.93
5.	**Fever (F)**	*Abutilon indicum (9), Achyranthes aspera (6), Leucas aspera (21), Ocimum tenuiflorum (60), Solanum trilobatum (48), Solanum nigrum (15), Tribulus terrestris (7), Commelina communis (9), Argemone mexicana (1)*	9	176	0.95

6.	**Menstural issues (MI)**	*Abutilon indicum (4), Ocimum tenuiflorum (10), Solanum nigrum (22), Ricinus communis (8), Citrullus lanatus (13), Momordica cymbalaria (6), Cucumis melo (15), Momordica charantia (12), Lagenaria siceraria (11), Celosia cristata (2)*	10	103	0.91

Table 5: Ailment Type, Used Value, Relative Frequency of Citation, Relative Popularity Level and Rank Order Priority of Selected Medicinal Plants

S. No	Name of the plant	Ailment Type	UV	RFC	RPL	ROP
1.	*Abutilon indicum*	SD, HA, F, MI	0.125	0.372	0.046	46
2.	*Achyranthes aspera*	SD, F	0.039	0.593	0.023	23
3.	*Leucas aspera*	SD, C, HA, F	0.066	0.697	0.046	46
4.	*Ocimum tenuiflorum*	SD, HP, C, HA, F, MI	0.24	2.41	0.058	58
5.	*Solanum trilobatum*	SD, C, HA, F	0.21	1.80	0.047	64
6.	*Solanum nigrum*	C, HA, F, MI	0.061	0.755	0.046	47
7.	*Tribulus terrestris*	HA, F	0.134	0.98	0.026	26
8.	*Tridax procumbens*	HP	0.142	0.081	0.011	11
9.	*Ricinus communis*	SD, HP, MI	0.048	0.720	0.034	34
10.	*Phyla nodiflora*	SD, HP	0.068	0.337	0.023	23
11.	*Cassia angustifolia*	HP	0.111	0.104	0.011	11
12.	*Luffa cylindrica*	SD	0.125	0.093	0.011	11
13.	*Citrullus lanatus*	MI	0.075	0.151	0.011	11

14.	*Momordica cymbalaria*	HA, MI	0.181	0.127	0.023	23
15.	*Cucumis melo*	MI	0.066	0.174	0.011	11
16.	*Momordica charantia*	MI	0.083	0.139	0.011	11
17.	*Lagenaria siceraria*	MI	0.090	0.127	0.011	11
18.	*Celosia cristata*	MI	0.05	0.023	0.011	11
19.	*Cardiospermum halicacabum*	SD, HP	0.090	0.255	0.023	23
20.	*Cleome viscosa*	SD,	0.02	0.058	0.011	11
21.	*Cleome gynandra*	SD, HA	0.158	0.151	0.023	23
22.	*Commelina communis*	HA, F	0.142	0.162	0.023	23
23.	*Argemone mexicana*	HA, F	0.04	0.058	0.023	23

Skin disease - SD, Hair Problem HP, Cold C, Head Ache - HA, Fever - F, Menstrual Issues - MI

Table 6: Fidelity Level Percentage of Medicinal Plants Based on Various Categories of Diseases

S. No	Ailment	Plant	FL%
1.	Skin Disease	*Abutilon indicum*	12
		Achyranthes aspera	26
		Leucas aspera	19
		Ocimum tenuiflorum	31
		Solanum trilobatum	16
		Ricinus communis	31
		Phyla nodiflora	9
		Cardiospermum halicacabum	9
		Cleome viscosa	11
		Cleome gynandra	5
		Luffa cylindrica	9
2.	Hair Problem	*Ocimum tenuiflorum*	15
		Tridax procumbens	8
		Ricinus communis	24
		Phyla nodiflora	31
		Cassia angustifolia	10
		Cardiospermum halicacabum	13
3.	Cold	*Leucas aspera*	25
		Ocimum tenuiflorum	70
		Solanum trilobatum	70
		Solanum nigrum	17
4.	Head ache	*Abutilon indicum*	9
		Leucas aspera	25
		Ocimum tenuiflorum	43
		Solanum trilobatum	37
		Solanum nigrum	15
		Tribulus terrestris	33
		Momordica cymbalaria	5
		Cleome gynandra	4
		Commelina communis	5
		Argemone mexicana	4
5.	Fever	*Abutilon indicum*	10
		Achyranthes aspera	6
		Leucas aspera	24
		Ocimum tenuiflorum	69
		Solanum trilobatum	55
		Solanum nigrum	17
		Tribulus terrestris	8
		Commelina communis	10
		Argemone mexicana	1
6.	Menstrual issues	*Abutilon indicum*	4
		Ocimum tenuiflorum	11
		Solanum nigrum	25

		Ricinus communis	9
		Citrullus lanatus	15
		Momordica cymbalaria	6
		Cucumis melo	17
		Momordica charantia	13
		Lagenaria siceraria	12
		Celosia cristata	2

PLATE 1: LIST OF PLANTS BELONGING TO EUPHORBIACEAE

Ricinus communis

Jatropha gossypiifolia L.

Chrozophora brocchiana (Vis.) Schweinf.

Euphorbia peplus L

Euphorbia serpens Kunth

PLATE 2: LIST OF PLANTS BELONGING TO EUPHORBIACEAE AND VEBENACEAE

Croton bonplandianus Baill.

Euphorbia prostrate Aiton

Stachytarpheta jamaicens (L.)

Lantana camara L.

Lantana aculeate L.

PLATE 3: LIST OF PLANTS BELONGING TO VERBENACEAE, LEGUMINOSAE AND CAESALPINIACEAE

Phyla nodiflora (L.) Greene

Acacia tortilis (Forssk).

Senna occidentalis (L.) Link

Galactia spp.

Cassia angustifolia Vahl.

PLATE 4: LIST OF PLANTS BELONGING TO RUBIACEAE AND ASCLEPIADACEAE

Oldenlandia umbellate, L.

Pergularia daemia (Forsskal) Chiov.

Oxystelam esculentum R. Br.

Calotropis procera (Aiton) Dryand.

Calotropis gigantea (L.) Dryand.

PLATE 5: LIST OF PLANTS BELONGING TO RHAMNACEAE, COMPOSITAE, ASTERACEAE AND MALVACEAE

Ziziphus spina-christi (L.) Desf.

Tridax procumbens (L.)

Parthenium hysterophorus L.

Stoebe plumose L.

Sida spinosa L.

PLATE 6: LIST OF PLANTS BELONGING TO MALVACEAE AND ZYGOPHYLLACEAE

Sida cordifolia, L.

Abelmoschus ficulneus (L.)

Hibiscus calyphyllus Cav.

Abutilon indicum, (L) Sweet.

Tribulus terrestris, L.

PLATE 7: LIST OF PLANTS BELONGING TO CUCURBITACEAE

*Luffa cylindrica (*L.) M. Roem

Citrullus lanatus (Thunb.) Matsum & Nakai

Momordica cymbalaria (Hook & Fenzl)

Cucumis melo L.

Momordica charantia L.

PLATE 8: LIST OF PLANTS BELONGING TO CUCURBITACEAE AND AMARANTHACEAE

Citrullus colocynthis (L.) Schrad

Mukia maderaspatana, W.

Lagenaria siceraria (Molina) Standley.

Cucurbita maxima Duch

Achyranthes aspera L..

PLATE 9: LIST OF PLANTS BELONGING TO AMARANTHACEAE, SAPINDACEAE AND SOLANACEAE

Celosia cristata L

Cardiospermum halicacabum L.

Physalis peruviana L

Physalis angulata L.

Solanum pimpinellifolium L.

PLATE 10: LIST OF PLANTS BELONGING TO SOLANACEAE

Solanum linnaeanum Happer

Solanum trilobatum, L.

Capsicum annuum L.

Datura matel L.

Solanum incanum L.

PLATE 11: LIST OF PLANTS BELONGING TO SOLANACEAE, TURNERACEAE, PASSIFLORACEAE AND LAMIACEAE

Solanum melongena L.

Turnera ulmifolia L.

Passiflora foediax L

Passiflora edulis Sims

Leucas aspera (Willd.) Link

PLATE 12: LIST OF PLANTS BELONGING TO LAMIACEAE AND CONVOLVULACEAE

Ocimum tenuiflorum

Mesosphaerum suaveolens

Ocimum kilimandscharium Baker ex Gurke

Merremia disseca (Jacq.) Hallier f.

Merremia aegyptia (L.) Urb.

PLATE 13: LIST OF PLANTS BELONGING TO CONVOLVULACEAE AND ACANTHACEAE

lpomoea aquatic, Forsk.

Ipomoea quamoclit L.

Ipomoea carnea, Jacq.

Barleria volkensii Lindau

Hygrophila auriculata (K. Schum.)

PLATE 14: LIST OF PLANTS BELONGING TO ACANTHACEAE, PEDALIACEAE, ONAGRACEAE AND BORAGINACEAE

Dicliptera paniculata (Forssk.) I. Darbush

Pedalium mure,, L.

Ludwigia octovalvis Linn.

Ludwigia palustris (L.) Elliott.

Heliotropium indicum L.

PLATE 15: LIST OF PLANTS BELONGING TO SCROPHULARIACEAE, CYPEACEAE, COMPOSITAE, CLEOMACEAE AND CAPPARIDACEAE

Stemodia durantifolia (Linn.)

Schoenoplectus tabernaemontani (C.C. Gmel.)

Corchorus trilocularis, L.

Cleoserrata speciosa (Raf.) Iltis

Cleome viscosa L.

PLATE 16: LIST OF PLANTS BELONGING TO CAPPARIDACEAE, COMMELINACEAE, PAPAVERACEAE, GERANIACEAE AND EUPHOBIACEAE

Cleome gynandra L.

Commelina communis L.

Argemone mexicana L.

Impatiens balsamina L.

Phyllanthus polygonoides Nutt.

PLATE 17: LIST OF PLANTS BELONGING TO COMPOSITAE, ARALIACEAE, OLEACEAE, LYTHRACEAE AND RUBIACEAE

Canthillium cinereum

Hydrocotyle spp.

Jasminum sambac (L.) Aiton

Ammannia coccinea Rottb.

Scleromitrion diffusum (Willd.) R.J.Wang

CONCLUSION

The present study carried out in Athimarapatti village and has documented 90 plant species belonging to 34 families. The floristic and ethnobotanical survey of the village concludes that rural people of Thoothukudi district possess rich ethnobotanical knowledge about treatment of various diseases but this traditional knowledge is declining due to rapid urbanization and migration of rural people. Thus, it becomes necessary the documentation of ethnobotanical knowledge. This study also suggested that documentation of flora with various uses provides raw material for pharmacological investigation and resulting in discovery of drugs. This documentation of flora and ethnobotanical knowledge provides a catalog of useful plants of the village and will serve as a physical record for both student community those who are studying botany as well as for village people.

PLATE 18: LIST OF PLANTS BELONGING TO RUBIACEAE, URTICACEAE AND POACEAE

Brachiaria brizantha (Hochst. ex A. Rich.)

Citrus aurantiifolia (Christm.) Swingle

Boehmeria cylindrica (L.)

Digitaria sanguinalis (L.)

Orysa sativa L.

BIS (2-ETHYLHEXYL) ADIPATE DEGRADATION VIA A MICROALGAL APPROACH - A CRITICAL NECESSITY

A dissertation submitted to

ST.MARY'S COLLEGE (Autonomous), Thoothukudi

affiliated to

MANONMANIAM SUNDARANAR UNIVERSITY, Thirunelveli

in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE IN BOTANY

By

S. Loorthumini

Reg. No. 21APBO07

DEPARTMENT OF BOTANY

ST. MARY'S COLLEGE (Autonomous)

THOOTHUKUDI -628001

April - 2023

CERTIFICATE

This is to certify that this dissertation entitled, **"Bis(2-ethylhexyl)adipate degradation via a microalgal approach - A critical necessity"** submitted by **S. Loorthumini Reg. No. 21APBO07 to St. Mary's College (Autonomous)**, Thoothukudi in partial fulfilled for the award of the degree of "Master of Science in Botany' is done by her under my supervision. It is further certified that this dissertation of any part of this has not been submitted elsewhere for other degree.

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DECLARATION

I do here by declare that this dissertation entitled, **"Bis (2-ethylhexyl)adipate degradation via a microalgal approach - A critical necessity"** submitted by me in partial fulfillment for the award of the degree of "Master of Science in Botany', in the result of my original and independent work carried out under the guidance of Dr. G. Flora M.Sc., M.Phil., Ph.D., Assistant Professor, Department of Botany, St. Mary's college (Autonomous), Thoothukudi and it has not been submitted elsewhere for the award of any other degree.

S. Loorthumini
(S. Loorrthumini)

Station: Thoothukudi

Date: 12.04.2023

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Contents

INTRODUCTION

Plastic is derived from the Greek word *plasticos*, which means capable of being moulded into various shapes. Plastic materials have found versatile applications in every aspect of modern human life as a result of rapid technological advancement and the geometric progression of global population growth. Different substances are blended in various proportions in plastics to improve performance and reduce costs (Li *et al., 2005).* Plastic is a matter that is hard to destroy and degrade once manufacture that goes in contradiction to natures rule: consequently, it creates a catastrophe for the complete world (Gilmore *et al*., 2018). Plastic pollution has become one of the most wide spread recalcitrant environmental contaminants (Stabinkova *et al*., 2021). Plastic particles are ubiquitous pollutants in the living environment and even in the blood stream of human beings. Polyethylene terephthalate, polyethylene and polymers of styrene were widely encountered in the blood stream (Lesile *et al*., 2022). Plasticizers are among the most important additives needed in the processing of polymer materials, particularly polyvinyl chloride (PVC) plastics, which account for more than 60% of total plastic auxiliary yield (Erythropel *et al*., 2014; Rahman *et al*.,2004). Traditional petroleum-based phthalate plasticizers are the most commonly used around the world. Phthalate plasticizer yield and consumption account for a large proportion of total plasticizer production and sales, but they are gradually being phased out due to potential threats to human health and the environment. Strict environmental and safety regulations have been developed and implemented. A research focus has been on the development of environmentally friendly non-toxic plasticizers and biodegradable bio-based plasticizers to replace phthalates. Non-toxic green plasticizers with high performance, oil resistance, extraction and migration resistance are constantly being developed, produced, and applied in electrical insulation, food packaging, and medical and health products. Plasticizers are functional

additives used to improve polymer flexibility, plasticity, processability, and elongation, particularly in PVC products (Choi *et al*., 2004). Phthalates, also known as phthalic acid esters, are a type of xenobiotic organic compound that is widely used to make plastic goods more flexible. (Kashyap and Agarwal, 2018). They are colourless, odourless, and flavourless, and exist as liquids over a wide temperature range (25°C to 50°C) (Tran *et al.,* 2021). Di(2-ethylhexyl)phthalate (DEHP) was added to plastic polyvinyl chloride (PVC) in the 1930s to improve flexibility and elasticity. OECD (Organization for Economic Cooperation and Development, 2018), phthalate ester (PE) including di(n-butyl) phthalate (DBP) and di (2-ethylhexyl) phthalate (DEHP) is synthetic compound commonly used as a plasticizer to impart flexibility, workability, and durability to polymers such as polyvinyl chloride. Also this compound is used in a wide variety of products such as paints, adhesive in cosmetics (Ling *et al*., 2007; Babu and Jiunn-tzong-wu, 2010). The most commonly used plasticizers, phthalates and adipates, are hydrophobic and lipophilic and can accumulate in the soil. In this regard, an accurate toxicological assessment of these compounds is required. There is some analogy between the physiological and toxicological properties of chemically similar substances. Furthermore, when researching the toxicity of plasticizers, it is necessary to consider the toxicity of the substances that form them. The type of acid that forms, such as the alcohol radical, influences the physiological and toxicological properties of plasticizers. An increase in toxicity is associated with a decrease in the number of carbon atoms in the alcohol radical in the series of phthalic acid esters (Lazarev *et al*., 1976; Vikhareva *et al*., 2021). As a result, PE has become ubiquitously disturbed in the environment (Yuun *et al*., 2008). In 2019 annual plastic production was 368 million tons , and it is expected that is production will increase up-to 33 billion tons by 2050 (Bellesi *et al*., 2020; Plastic Europe, 2020). It is estimated that 76% of total plastics produced are land filled or

spread in the natural environment (Geyer, 2020). When plastic garbage is disposing in landfills, it appears to eliminate waste from the upper surface of land but it actually diminishes agriculture land (Zhang *et al*., 2004). Because natural decomposition takes so long, the landfill area cannot be used for other purpose (Tansel and Yildiz, 2001). In comparison to landfills incineration is a superior solution because as it requires less space and provide better energy recovery (Sinha *et al*., 2010). However it is not without restriction because it produces green house gases, as well as Polychlorinated biphenyls (PCBs) and free radical exposure (Astrup *et al*., 2009). Regardless, landfills and incinerated constraint can be addressed through recycling, though this procedure is costly and its end product is poor (Yamada-Onodora *et al*., 2001). In compare to other approaches, biodegradation is the most effective one. It is inexpensive and does not emit hazardous pollution in to the atmosphere.

The main polymer constituents of microplastics found in water have been identifiedas polyethylene, polypropylene, poly stryrene and polyethylene terephthalate accounting for 70% of the total but polyvinylchloride, polyacrylonitrile, rubber different copolymers are also common (Li *et al.,* 2020). MPS are constantly present in fresh and marine ecosystem and they easily leaching the plasticizers due to physical and chemical factors of the nature. Because some plastic additives are physically bound to the plastic and can easily be released into the environment, they will eventually become available to organisms. Heat and acidic or basic conditions (E.g., bisphenol A) can disrupt the hydrogen bonds of additives that are chemically bound to polymers, releasing the additives into their surroundings (Rani *et al*., 2015; Hermabessiere *et al*., 2017). Plastic additives have been found in varying concentrations and distributions in the biosphere (Hermabessiere *et al*., 2017; Hahladakis *et al*., 2018). As a result, plastic environmental pollution is caused not only by the plastic materials themselves, but also by the

chemicals used in plastic manufacturing to achieve the desired characteristics of each product. Plastic additives have been reported to be potentially toxic to mammals (e.g., endocrine disruptors, contributors to chronic health effects, and cancer risks) (Hermabessiere *et al*., 2017; Hahladakis *et al*., 2018; Groh *et al*., 2019). A lot of micro plastic debris was found even in oceanic surface water (Audrezet *et al*., 2020). It is considered that micro plastic have become a main source of anthropogenic pollution of the oceans (Bowly *et al*., 2021). MPS concentration in highly contaminated rivers could be up to 100mg/L. It is evident that the quantity of micro-plastic will increase over the next decade, so the fate, and biological impact on the environment of this contaminant are in focus of scientific research. Micro-plastic have been also in freshwater (Wagner *et al*., 2014), drinking water (Eerkes-Medrano 2019), soil (Guo *et al*., 2020), as well as in food particle (Rainieri *et al*., 2019). Di Methyle Phthalate (DMP) only has a modest level of toxicity, its metabolic intermediary mono-methyl phthalate (MMP) is not only poisonous but also and endocrine disrupter that can affect how animal and even human developed and reproduced by decreasing sperm counts and testosterone production (Brar *et al*., 2009). Microalgal biotechnology did not really take off until the middle of the last century. Now microalgae were used in many fields including bioremediation, bioaccumulation, etc. Microalgae are well-known pollutant scavengers for a wide range of chemicals emitted by the domestic, industrial, and agricultural sectors. Microalgae are photosynthetic microorganisms found at the base of aquatic food chains. Eco-pollutants are toxic to microalgae and, as a result, have a negative impact on all higher-level organisms in food chains, as well as human surroundings and humans. Metals and metalloids, organic solvents, pesticides, and detergents, as well as pharmaceuticals and personal care products, have all been shown to have a negative impact on micro-algal populations (Maizek and Brozek-Pluska, 2019). Many researchers found that microalgae

have the capacity to reduce the toxicity of plasticizers. Similarly the microalgae also do adsorption and absorption on emerging contaminants. These kinds of information evidenced that the microalgae certainly helps to eliminate the BEHA pollution in the aquatic environment. Hence the present study focused on the potential of microalgae in degradation of (BEHA) bis(2-ethylhexyl) adipate.

Massive plastic accumulation has been taking place across diverse environments like aquatic and terrestrial environments due to large-scale plastic production. Now a days, societies struggle with continuously increasing concerns about the subsequent pollution and environmental stresses that have accompanied this plastic revolution. Exposure to weather conditions and environmental micro-flora like bacteria and microalgae can slowly corrode the plastic and release the plasticizers. These are potential sources of negative effect on global food chains. In recent years, several studies have been targeting the utilization of micro algae for remediate the plastic and plasticizers pollution. Hence the bioaccumulation of di(2-ethylhexyl) adipate using *Chlorella sp.* was studied. Through this research the reduction of emerging contaminants will be studied meanwhile the adsorption, absorption and accumulation of Bis(2- ethylhexyl) adipate (DEHA) by microalgae was also established. This research definitely become a boon to our society.

Objectives

The main objectives of this study are as follows;

- To identify the microalgae which degrade plasticizers.
- To estimate the chlorophyll content of the microalgae with and without treatment of bis(2-ethylhexyl)adipate.
- To retrieve the level of degradation of plasticizer using GC-MS.

Micro-plastic pollution is a difficult issue (Windosr *et al*., 2019) with significant environmental and public health repercussions. This pollution problem is a classic transboundary of how land based pollution can spread rapidly, even into remote areas such as virgin mountainous regions, wilderness areas, and the Arctive (Bergman *et al*., 2019; Brahney *et al*., 2020), as well as the ocean's deepest trenches (Jamieson *et al*., 2019). Because plastic pollution is physically apparent, it has piqued the interest of a diverse group of stakeholders including scientists, policy markers, the media, and the general public. This subject has gotten a lot attention, possibly more than any other pollution concern in history of science (Sedlak, 2017).

Food, medicines, cosmetics, detergents and chemicals all employ synthetic polymers in their packaging. Plastics are utilized for packaging applications in about 30% of the world's population. The usage rate is still growing at a rapid rate of 12% each year. Because they offer greater physical and chemical qualities. Such as strength, lightness, resistance to water and most water-borne germs, they have supplanted paper and other cellulose-based products for packaging. Polyethylene (LDPE, MDPE, HDPE and LLDPE), polypropylene (PP), polystyrene (PS), polyvinyl chloride (PVC), polyurethane (PUR), poly(ethylene terephthalate) (PET), poly(butylenes terephthalate) (PET), poly(butylenes terephthalate (PBT), nylons are among the most commonly used plastics in packaging. Plastics are widely used not just because of their advantageous mechanical and thermal qualities, but also because of their stability and durability (Rivard *et al*., 1995). Because of their durability and visibility in litter, plastics (Polymers) have gotten more public and media attention than any other component of

the solid waste stream. In 1993, the global demand for plastics was around 107 million tones, and in 2000, it was anticipated to be above 146 million tones.

The dramatic increase in production and lack of biodegradability of commercial polymers, mainly commodity plastics used in packaging (e.g. fast food), industry and agriculture, has focused public attention on a potentially huge environmental accumulation and pollution problem that could persist for centuries (Albertson *et al.,* 1987). Plastic waste is dispose off through the process such as land filling, incineration and recycling. Because of the persistent presence of wasted plastic in our environment, several communities have become more aware of its negative impacts on animals and the aesthetic characteristic of towns and forests. Plastic that has been improperly disposed of has the potential to damage people by polluting the environment. Furthermore, the combustion of polyvinylchloride (PVC) polymers releases persistent organic pollutants (POPs) such as furans and dioxins (Jayasekara *et al.,* 2005).

Plastics have become an indispensable part of the society due to their lightweight, easy handling, durability, flexibility, resistance to water, and other microbial attacks. However, their extensive use has led to a plastic pollution problem. To avoid long-term environmental damage, degradation of plastic is the most preferred option. Microbial degradation using bacteria and fungi is an emerging strategy to manage plastic waste. This chapter highlights the benefits, concerns, and threats surrounding the use of plastics, including plastic production and plastic waste generation, environmental and health effects of plastic pollution, plastic waste management options, biodegradation of plastic polymers and the mechanism involved, biodegradable plastics, and challenges and constraints of plastic waste biodegradation (Yogalakshmi and Singh, 2020)

Microplastics concentration in the Manas River, china, was explored by a group of researchers (Wang *et al*., 2019). They observed a range of 21±3 to 49±3 items/L in which fibrous microplastics were dominant at all sites with a size range of 0.1 and 1.0 mm, while black and white were the dominant colours. IR spectral analysis showed that the dominant polymers were PP and PET. The study can help to understand the contamination features of microplastics in inland rivers.

Microplastic fibers constitute the largest component of plastics in aquatic environments. (Sait *et al*., 2021) investigated the photodegradation of PET, PA, PAN and their respective chemical profile, along with their potential for additive leaching. PET and PA fibers showed significant morphological changes upon exposure to UV. Chemicals identified in fibers and aqueous leachates include monomers, UV stabilizers and degradated polymers. Bisphenol A, Bisphenol S and Benzophenone-3 were quantified in all fibers and wool at concentrations between 4.3 and 501 mg/g, with wool displaying maximum concentration of BPs and BzPs at 863 and 27 mg/g, respectivetly.

Kosuth *et al*. (2018) investigated microplastic particles in tap water, beer, and commercial sea salt. Microplastics were present in 81% of the tap water samples which composed of fibers (98.3%) of 0.1± 5 mm. the abundance ranged at 0 to 61 particles/L. Likewise microplastics were present in beer and salt. The composition was dominated by fibers (99%). Beer was contaminated with 46.7 to 806 particles/kg. It was estimated that an average person consumed more than 5800 particles of microplastics from tap water, beer and salt. In another study, high and low cost commercial were purchased by Renzi and Blaskovic (2018) to analyse for the presence of microplastics. The microplastics ranged between 1.57- 31.68 items/g. the sizes of particles ranged within 4-4628 µm. the samples were purchase from Italy and Coratia and it was found that all samples from

both the countries had microplastic contaminated but varied in abundance which is dependent on several factors.

Gundogdu and Cevik (2017) reported the distribution of micro-meso plastics in the Northest Levantine coast of Turkey. The average of micro and meso plastics was determined to be 0.376 items/m². The highest value was determined in Mersin Bay at the mouth of the Seyhan river (906) items and the lowest level was found in Station No.4 in Iskenderun Bay (78 items).

Babu and Wu (2010) observed thephthalate esters are widely distributed pollutants that originate from synthetic plasticizer and are known to act as toxicants as well as environmental pheromones in aquatic ecosystems. This study revealed that sixteen species of freshwater algae and cyanobacteria were capable of producing di(n-butyl)phthalate (DBP) or mono(2-ethylhexyl)-phthalate (MEHP) or both. The incubation of the cells in culture medium containing $NaH_{13}CO_3$ confirmed that both phthalates were de novo synthesized by the studied cells.

Chang *et al*. (2021) reported that Phthalate esters (PAEs) are one of the most widely used plasticizers in polymer products and humans are increasingly exposed to them. Epidemiological studies found a consistent association between PAE exposure and a decrease in sperm quality in males and symptom development of ADHD in children. Future studies need to thoroughly perform in large-scale populations to increase the precision of the association and enhance the overall understanding of potential human health risks of PAEs.

Benjamin *et al*. (2015) investigated the phthalates are a group of xenobiotic and hazardous compounds used in plastics to enhance their plasticity and versatility. He

have shown endocrine disruption, hepatotoxic, teratogenic and carcinogenic properties, but usage continues due to their cuteness, attractive chemical properties, low production cost and lack of suitable alternatives. The major phthalates used in industry, routes of environmental contamination, evidences for health hazards, routes for *in situ* and *ex situ* microbial degradation, bacterial pathways involved in the degradation process, half-lives of phthalates in environments.

Ganta *et al*. (2020) reported the bisphenols and phthalates are two known plasticizers that have been found to cause health impairments in various organs and fetuses and newborns. Bioremediation is a low cost and eco-friendly solution that can accumulate and concentrate the toxin to the point of easy disposal. Many living systems or their components can be used as agents for toxin removal among which they have discussed a few bacteria, fungi, and plant biomass for the removal of bisphenols and phthalates.

Gaur *et al*. (2022) described the microplastics have become a major environmental and human health hazard, and microorganisms have evolved to degrade different classes of plastic polymers. Meta "omics" approaches have been used to identify the active microbiota and microbial dynamics involved in the mitigation of microplastic-contaminated sites. Protein engineering approaches have opened new avenues to tackle this alarming situation.

Selvaraj *et al*. (2021) reported the microalgae, especially *Chlorella sp*., have been used for centuries as food and currently their biotechnologicl potential is notable for the presence of several compounds relevant to the market, like PHB. Microalgae are appealing because of the increasing demand for biopolymers like PHB. Therefore, the

present study is aimed to determine the efficacy of biomass production and yield of PHB producing microalgae isolated and identified by phylogenetic analysis**.**

Gobas *et al*. (2020) explored the bioaccumulation behavior of several phthalate esters in aquatic food-webs, concluding that they do not biomagnify in the food-web. Higher molecular weight esters (DEHP, DnOP, and DnNP) show evidence of trophic dilution, which is consistent with findings from laboratory and modeling studies. Bioaccumulation patterns of DBP, DiBP, and BBP indicate no significant relationship with trophic position consistent with a lipid-water partitioning model. Low bioavailability of the high-molecular weight esters in natural waters is the main reason why the BAFs of the higher molecular weight phthalates are below the UNEP criteria.

He *et al*. (2016) studied the removal and biodegradation of nonylphenol (NP) by four freshwater microalgae, including three green algae (*Scendesmus quadriauda, Chlorella vulgaris,* and *Ankistrodesmus acicularis*) and one cyanobacterium *(Chroococcus minutus),* was studied in bacteria-free cultures exposed to different concentrations of NP for 5 days. All four algal species showed a rapid and high ability to remove NP, with *A. acicularis* having the highest NP removal rate (83.77%) at 120 h when exposed to different NP treatments. *C. vulgaris* had the highest NP biodegradation percentage (68.80%).The extracellular NP contents were lower than the intracellular NP contents in all tested algae, with the ratio of the extracellular and intracellular content ranging from 0.04 to 0.85. These results indicate that *A. Acicularis* and *C. vulgaris* are more tolerant to NP and could be used for treatment of NP contaminated aqueous systems effectively.

Chi *et al*. (2007) investigated the influence of major nutrients (N, P) on the biodegradation and bioconcentration of dibutyl phthalate (DBP) and di-2-

ethylexylphthalate (DEHP) by *Chlorella vulgaris* in lake water. It found that nutrient addition had a significant effect on biodegradation rate constants and BCFs of DBP and DEHP, with P addition being less pronounced than N addition due to N-limitation status of phytoplankton, while addition of both N and P more greatly affected biodegradation than addition of N or P. BCFs decreased with increasing algal exudate as measured by dissolved organic carbon (DOC) and a strong correlation between BCFs and DOC was obtained. This suggests that DOC plays an important role in the bioconcentrate of DBP.

Touliabah *et al*. (2022) proposed a new bioremediation method based on the diverse functionalities of algae that is more ecologically friendly and environmentally sustainable than prior methods with other bacteria. Algae-based wastewater treatment systems are becoming increasingly popular due to their environmental sustainability and lack of secondary pollutants. Phytoremediation is a cost-effective alternative to conventional treatments for degrading organic contaminants and can be an important part of the bioenergy value chain. They focuses on microalgae and cyanobacteria species, which may remove many organic contaminants from water systems.

Liu *et al*. (2022) investigated the plastics and microplastics are difficult to degrade in the natural environment due to their hydrophobicity, covalent bonds, and functional groups. In nature, they are more likely to attract other substances, which can be toxic and harmful. Degradation is an effective way to eliminate plastic pollution, but there are no mature and effective methods that can be applied in engineering practice or widely used in nature. There is an urgent need for research on the degradation of (micro)plastics.

Tan *et al*. (2023) reported the nonphthalate plasticizers (NPPs) are increasingly used for industrial needs, but knowledge is limited on their environmental occurrences,

fate, and human exposure risks. They investigated 45 NPPs along with major PAEs in house dust from five regions in the Asia-Pacific region and the US. The median total concentrations of NPPs ranged from 17.8 to 252 μg/g, while the mean ratios of ΣNPPs to ΣPAEs ranged from 0.19 (Hanoi) to 0.72 (Adelaide). Potential exposure risks cannot be overlooked due to lack of toxic threshold data, additional exposure pathways, and possible cocktail effect.

Palah *et al*. (2020) explored the potential of *Chlorella vulgaris* and Pretreatment to remediate plastic waste. Results showed that Pretreatment had a marked effect on the cracking and alteration of plastic polymer, which helped to grow microbial species on the cracked surface. GCMS analysis revealed that the microbial specie could produce biodegradation products such as alkanes ester, fatty acids, benzoic acid, and aromatics. The most toxic product of biodegradation is Bis (2-Ethyl hexylphthalate), which is the biodegradation product of toxic ingredient of plastics.

Moog *et al*. (2019) reported the biological degradation of plastics is a promising method to counter the increasing pollution of our planet and develop eco-friendly recycling strategies. *Ideonella sakaiensis*, a bacterium possessing the ability to degrade PET and use the degradation products as a sole carbon source for growth, was isolated in 2016. It expresses a key enzyme responsible for the breakdown of PET into monomers, PETase, which has potential for the development of biological PET degradation and recycling processes as well as bioremediation approaches of environmental plastic waste.

Khoironi *et al.* (2019) investigated the polyethylene terephthalate (PET) and Polypropylene (PP) are the most widely used plastics in manufacture of packaging, fibres, and drinking bottles. This study evaluated the interaction between microalgae

Spirulina sp. and microplastics in a 1 L glass bioreactor for 112 days. The results showed that the tensile strength of micro plastic PET decreased by 0.9939 MPa/day while the decreasing carbon in PET was higher than PP. The CO_2 evolution of cells imposed by PET microplastic was higher than imposed by PP. Biodegradation has important role in the degradation process of plastic.

Barone *et al*. (2020) studied the plastic accumulation has been taking place across diverse landscapes since the 1950s, leading to increasing concerns about pollution and environmental stresses. Degradation of used plastics is highly time-consuming and causes volumetric aggregation. Exposure to weather conditions and environmental microflora can slowly corrode the plastic structure. Cyanobacteria (e.g., *Synechocystis sp.* PCC 6803, and *Synechococcus elongatus* PCC 7942), which are photosynthetic microorganisms and were previously identified as blue-green algae, are currently under close attention for their abilities to capture solar energy and the greenhouse gas carbon dioxide for the production of high-value products. Microalgae are also suitable for environmental and biotechnological applications based on the exploitation of solar light. In recent years, several studies have been targeting the utilization of microorganisms for plastic bioremediation. Wild-type or engineered cyanobacteria may represent an interesting, environmentally friendly, and sustainable option.

Zhang *et al*. (2018) investigated the combined effects of UV-B irradiation and di-(2-ethylhexyl) phthalate (DEHP) on photosynthesis and antioxidant system of *Scenedesmus acuminatus*. Results showed that UV-B radiation decreased chlorophyll a fluorescence yield, photosynthetic activity (Fv/Fm), pigment content and superoxide dismutase activity, while DEHP increased ROS production and soluble protein and malondialdehyde contents. The highest degradation rate was 89.9% at an initial DEHP

concentration of 10 mg L−1 within 6 h. This result may be attributed to the regulation of ROS generated by *S. acuminatus*, and the addition of high DEHP concentration aggravated cell damage.

Gu *et al*. (2017) investigated the acute toxic effects and underlying mechanisms of dibutyl phthalate (DBP) at different concentrations (0–20 mg L−1) on two typical freshwater algae (*Scenedesmus obliquus* and *Chlorella pyrenoidosa*). The growth of both algae was inhibited by DBP exposure, and the 96-h median effective concentration values (96h-EC50) were 15.3 mg and 3.14 mg. The increased production of intracellular reactive oxygen species and malondialdehyde content was linked to oxidative stress and lipid peroxidation in both algae, as well as increased activity of antioxidant enzymes such as superoxide dismutase and catalase. The findings will contribute to the understanding of toxic mechanisms in PAEs and the evaluation of environmental risks for primary producers in aquatic ecosystems.

Li *et al*. (2020) reported the microplastics are ubiquitous in aquatic ecosystems, but knowledge on their impacts on phytoplankton, especially freshwater microalgae, is limited. To investigate this issue, microalgae *Chlamydomonas reinhardtii* was exposed to polystyrene (PS) microplastics with 4 concentration gradients and the growth, chlorophyll a fluorescence, photosynthetic activities, Fv/Fm, contents of malondialdehydes (MDA), soluble proteins, extracellular polymeric substances (EPS) and settlement rate were measured. Results showed that the density of microalgae decreased as the increase of PS microplastics concentrations, and the highest inhibitory rate (IR) was 45.8% on the 7th day under the concentration of 100 mg/L. The high concentration (100 mg/L) of microplastics also inhibited the content of EPS released by microalgae into the solution. The scanning electron microscope (SEM) images showed

that microplastic beads were wrapped on the surface of the microalgae and damaged their membranes, which could suggest the reduction of photosynthetic activity and the increase of soluble proteins and MDA content.

Gao and Chi, (2015) investigated the biodegradation of diethyl phthalate (DEP) by three marine algae. The first-order biodegradation rate constants of DBP in algal solutions were in the order of *Cylindrotheca closterium* > *Dunaliella salina* > *Chaetoceros muelleri*. When singly existed, DEP was degraded more quickly than in a mixture with DBP, indicating that DBP had inhibitory effect on the biodegradation of DEP. The degradation trends of DEP and DBP in both extra- and intracellular crude extracts were similar, and DEP was largely in water phase while DBP remained in both water phase and algal phase. It was concluded that algal extracellular enzymes played key roles in the degradation of DBP.

Zhang *et al*. (2016) investigated the biodegradation characteristics of dimethyl phthalate (DMP) by three freshwater unicellular organisms. All three organisms were capable of metabolizing DMP, with PCC7822 achieving the highest degradation efficiency. Phthalic acid (PA) was detected to be an intermediate degradation product of DMP and accumulated in the culture solution. The optimal initial pH value for the degradation was 9.0, which mitigated the decrease of pH resulting from the production of PA. After 72 hours' incubation, no more than 11.8% of the residual of DMP aggregated in Cyanobacteria cells while majority of DMP remained in the medium. Esterase was induced by DMP and the activity kept increasing during the degradation process, suggesting that esterase could assist in the degradation of DMP.

Ji *et al*. (2014) investigated the toxicity and cellular stresses of bisphenol A (BPA) to *Chlamydomonas mexicana* and *Chlorella vulgaris* and its

biodegradation/bioaccumulation by both microalgae. The 120-h EC50 of BPA was 44.8 and 39.8 mg L−1, respectively, and the dry cell weight and chlorophyll a content decreased with increasing BPA concentration. The highest rates of BPA biodegradation, 24 and 23% respectively, were achieved at 1mg L−1 BPA. Total nitrogen (TN) and total phosphorous (TP) removal was higher in C. vulgaris. Both microalgae were more tolerant to BPA and could be used for treatment of BPA contaminated aqueous systems.

Gao *et al*. (2021) reported that the marine diatom (*Phaeodactylum tricornutum*) was exposed to different concentrations of dimethyl phthalate and diethyl phthalate (DEP) for 96 h to investigate their toxicities. Results of this study showed that the diatom could remove DMP and DEP effectively with removal rates of 0.20–0.30 and 0.14–0.21 mg L−1 h−1, respectively. However, the two PAEs significantly inhibited photosynthesis and chlorophyll biosynthesis. Additionally, reactive oxygen species (ROS) level and antioxidant enzymes (SOD and POD) activity increased with the increase of PAEs concentrations. The results of this study help to understand the toxic mechanisms of PAEs and provide strong evidences for evaluating their ecological risks in the marine environment.

Chenchenshen *et al.* (2019) revealed di-2-ethylhexyl phthalate (DEHP) poses a great threat to aquatic ecosystems, with known hazards to aquatic species. This study investigated growth inhibition, oxidative damage and antioxidant enzyme activities in *Chlorella vulgaris* under DEHP treatment. Results showed that DEHP reduced superoxide dismutase and glutathione peroxidise activities, increased hydrogen peroxide level and MDA content in a concentration-dependent way, indicating that DEHP could have biochemical and physiological toxic effects in *C. vulgaris.* These findings helped to

understand the toxicity mechanisms of DEHP and the environmental risk assessment of primary producers of aquatic ecosystems.

Yan *et al*. (1995) studied the *Chlorella pyrenoidosa* has an ability to accumulate and biodegrade phthalate esters, with maxima of 162 at 24 h, 205 at 12 h, and 4,077 at 12 h. Average biodegradation rates of DMP, DEP, and DBF per day were found to be 13.4, 7.3, and 2.1 mg/L, respectively. A second-order kinetic equation was formulated as - dC/dt = KNr, with a factor r indicating the rate of algal growth.

Chi *et al.* (2005) investigated the microalgae have the ability to degrade and accumulate organic pollutants, but little is known about the biodegradation and accumulation of di(2-ethylhexyl) phthalate(DEHP). This study studied the accumulation and biodegradation kinetics of DEHP in *Chlorella vulgaris*. The initial concentration of DEHP was approximately 0.4 mg/L, and the alga was able to accumulate and degrade DEHP significantly. The amount of DEHP accumulation by the alga reached maximum of 107.5 mg/g dry weight at 0.5 h and bioconcentration factor reached maximum of 3.67Ã—105 at 6 h. First-order biodegradation constant was 0. 0021 h-1.

Rabet *et al*. (2018) described the effects of two plastic-derived chemicals, Bisphenol A (BPA) and di-2-ethylhexyl phthalate (DEHP), on the abundance and physiological responses of the marine toxic dinoflagellate *Alexandrim pacificum* were assessed during 7 days of exposure. Results showed that *A. pacificum* was highly sensitive to these contaminants, with a decrease in biomass and photosynthetic activity. However, recovery of contaminated cells activity depending on exposure time and BPA and DEHP contamination could be related to an adaptation to induced stress. Hu *et al.* (2022) described the *Gordonia sp.* GZ-YC7 is a new phthalate esters degrading strain isolated from soil of plastic film mulch culture. It exhibited the highest di-(2-ethylhexyl)

phthalate degradation efficiency under 1000 mg/L and the strongest tolerance to 4000 mg/L. Comparative genomic analysis revealed that there exist diverse esterases for various phthalates, which may contribute to its broad substrate spectrum, high degrading efficiency, and high tolerance. It has potential for bioremediation in polluted soil environments.

Kumar *et al*. (2017) investigated the conventional methods of polyethylene degradation are lethal to the neighboring environment, and a better solution is needed. It is also investigated the biological treatment of domestic polyethylene bags from three different sites in Chennai, Tamil Nadu, India. Microalgae like green algae, blue-green algae and diatoms were isolated from the polyethylene sheets and selected for the biological treatment. The most dominant microalgae were *Scenedesmus dimorphus* (Green microalga), *Anabaena spiroides* (blue-green alga) and *Navicula pupula* (Diatom). The scanning electron microscopical study revealed that the degradation was evident in the treatment of LD polyethylene sheet using the microalga *Anabaena spiroides*, which was found to grow feasibly rather than the other microalgae.

Bate *et al.* (2019) investigated the increasing incidence of cyanobacterial blooms in Southern African aquatic systems is raising concern about the potential for these microorganisms to contaminate potable water with toxic secondary metabolites. They focused on two lakes, an estuary and the sea in a small catchment in Maputaland, northern KwaZulu Natal, South Africa, fed by groundwater impacted by sewage effluent. Analysis of the microalgae in two freshwater lakes showed that cyanophytes made up over 88% of the phytoplankton in the larger Lake Mgobezeleni and over 50% in the smaller Lake Shazibe, raising concerns about the potential health risk to communities using this water for domestic, agricultural and recreational purposes.

Generation Sequencing analysis of bacterial 16S rRNA genes showed that cyanobacterial taxa closely related to species that are known to produce cyanotoxins were detected in all the water bodies sampled. These results highlighted the importance of identifying water systems at risk of experiencing cytotoxic cyanobacterial bloom and monitoring such vulnerable systems to ensure the safety of surrounding community.

Susanti *et al*. (2021) reported that the microalgae are oxygenic photosynthetic microorganisms and closely related to the heterotrophic oraganism. They have a variation in morphological features, biochemical composition, reproduction, cell organization, plastid structure, and habitat. Currently, algae are divided into 10 groups composed of either prokaryotic or eukaryotic. In terms of application, algae biomass is utilized as a valuable natural product worldwide, but biomass productivity needs to be enhanced using complementary growth media usage. Research during the COVID-19 pandemic about the utilization of anaerobically digested dairy manure wastewater as an alternative media for algal biomass production will also be reported.

Sandeep *et al*. (2018) studied the microalgal diversity and dynamics of a tropical estuarine ecosystem (Muttukadu, of Indian south east coast) and applies tools like isolation of useful species to utilize in aquaculture and conserve native strains. Selected diversity indices (Simpson index, Dominance index, Shannon- Weiner index, Pielou's evenness index and Margalef richness index) were used to describe trends of diversity in the estuary during the study period. Sixty three species of microalgae belonging to Chlorophyceae were identified, with Bacillariophyceae forming the dominant flora with twenty six species. The species diversity was increased after the flood during December-2015 in south east coast of India. Nutrient profiling of isolates revealed the presence of essential fatty acids (EPA & DHA) in high percentage in some of the isolates.

Rampinelli *et al*. (2022) isolated and identified the diatoms found in the Paranaguá Estuarine Complex (PEC). The diatoms were purified and analyzed with light and scanning electron microscopy for morphological identification, while DNA sequences were used for molecular identification. The two best-selected strains were identified as belonging to two genera, *Nitzschia* and *Navicula*. The rbcL region was found to be the most informative for species identification.

Arsad *et al*. (2022) aimed to assess the diversity of microalgae in several different sub-habitats at Siwil Beach and Sempu Island. It used a quantitative descriptive method with data collection techniques, incorporating the purposive sampling method, and non-metric multidimensional scaling. The results showed that the composition of the microalgae species was dominated by Bacillariophyceae, with a total abundance of 5,423,073 cells/cm2, while the highest abundance in Sempu Island was 1,986,252 cell/cm2. Factors that mainly affected the abundance were environmental, as evidenced by the measurement of water quality.

Trivedi and Mitra, (2021) revealed the pelagic environment of the ocean supports two basic types of marine organisms: plankton and nekton. Phytoplankton are free floating tiny floral components that require sunlight, nutrients or fertilizers, carbon dioxide gas and water for growth. Nekton are freefloating animals that are strong enough to swim against independent of water movements. Marine phytoplankton was the dominant producers in the ocean, and their role in the marine food chain is of paramount importance. Approximately 4000 species have been described, and they exhibit remarkable adaptations to remain in floating condition in the seawater. These adaptations include their small size and general morphology, colony or chain formation, and ionic

regulation. A field study was conducted in September, 2017 in the Thakuran River to identify 73 species in a salinity range between 12psu to 18psu.

Ge *et al*. (2022) examined the phytoplankton alpha and beta diversity using investigation data in May (springtime), August (summer) and November (autumn) 2009 in China's Jiulong River estuary, where it was easily polluted due to human population and low self-purification capacity. Potential influencing factors were explored, including dissolved oxygen, salinity, nutrients, nutrient ratios, geographic and hydrologic distance, and so on. Results showed that Shannon's index (H') and Pielou's index (J) decreased from the estuary's upper to middle and then increased from middle to lower reaches, Simpson's (D) observed the opposite trend and species number (S) gradually increased. Beta diversity also showed a gradual decrease trend from the upper to lower reaches. Nutrients and nutrient ratios were characterized by excess nitrogen (N) and silicon (Si) and limited phosphorus (P), which could potentially cause diatom blooms. This study advocates for the protection of the entire estuary system with particular emphasis on its upper reaches, and greater attention should also be paid to impacts associated with N input and nutrient ratio trade-offs to the prospective watershed management of this estuary.

Maltsev and Maltseva (2021) studied the possibility of obtaining commercially valuable products from microalgae stimulates scientific research in this direction. Fatty acids (FAs) are involved in the metabolic pathways of formation and conversion of most lipid classes, and their composition largely determines their properties and practical use. They summarizes information on the diversity of the fatty acid composition of microalgae and cyanobacteria, taking into account their rare and unusual categories. It is formed by 135 FAs, distributed into several groups based on the length of the

hydrocarbon chain, its structure and the presence of substituents. There are both saturated and unsaturated FAs with different numbers of double bonds, rich in omega-3 and omega-6 fatty acids. They also considers the use of fatty acids as an industrial resource, as well as a biomarker.

Hasan *et al.* (2022) studied the Pasur River estuary (PRE) provides vital fishery resources and supports millions of livelihoods in the southwestern coastal region of Bangladesh. This research focused on phytoplankton community assemblages, alpha diversity indices, and the seasonal succession of major phytoplankton species in relation to physicochemical parameters in the tidal mangrove creeks of the PRE. Spatial and temporal variations were assessed by water sampling at 17 stations in the study area from January to December 2019. The mean salinity level was significantly higher during the dry season than during the wet season, and no significant variation was observed in the dissolved inorganic nitrogen and dissolved inorganic phosphorus. Spatially, no significant variation in the alpha diversity was observed, but significantly ($p < 0.05$) varied temporally. The study classifies the study areas as highly diversified zones, and the succession from diatoms (dry season) to blue-green algae (wet season) is attributed to changes in the physicochemical and nutrient parameters depending on seasonal environmental parameter fluctuations.

Petal *et al*. (2021) examined the phytoplankton abundance and diversity from site 1 (downstream) and site 2 (upstream) of the Auranga Estuary (20˚63' N and 72˚82' E). A total of 44 species were recorded, 35 species from downstream and 24 species from upstream. *Dinophyceae* and *Chrysophyceae. Nitzschia, Coscinodiscus* and *Ceratium* were abundant genera at site 1 and *Spirogyra, Microcystis, Chlorella* and *Oscillatoria* were abundant at site 2. Spatially, downstream had higher species diversity and

abundance than upstream, while winter season was favorable for plankton growth compared to summer and monsoon. The Shannon diversity index was 1.417 and 1.268 for downstream and upstream, respectively, indicating less diversity level in this estuary.

Darmarini *et al*. (2023) aimed to evaluate the diversity and abundance of phytoplankton in mangrove habitats at Lubuk Damar, Aceh Tamiang Regency, via plankton net with a 20-micron mesh size. Results showed that in August 2017, the diversity was higher than in January, with an Index Diversity of 1.24-2.83 and an Index Dominance of 0.17-0.48. In January, *Chaetoceros sp*. was dominant in water, followed by *Bacillaria sp*, and *Biddulphia sp.* In August, *Leptocylindrus sp*. was the dominant in diversity, and diatoms were dominant in abundance.

Susanti *et al*. (2014) observed the phytoplankton role in aquatic ecosystems and is one of the bioindicators used to describe conditions, quality, and environmental changes. Jakarta Bay is polluted by various wastes originating from industry, domestic, and sea transportation for fishing activities, which causes a decrease in water quality. This study revealed the composition of phytoplankton and its relationship with the physical and chemical parameters of the waters that empty into Jakarta Bay. The study was conducted in September 2021 at three sampling stations in estuary areas. Station 3 was the most polluted area from industrial waste, but only found the group of Cyanophyta (61.57%), Euglenophyta (38.43%), Chrysophyta (39.74%), and Chlorophyta (19.21%). Station 2 had the highest concentration of chlorophyll-a, but the water quality of the two sampling stations greatly affected the composition and abundance of phytoplankton.

Varghese *et al.* (2022) collected the phytoplankton samples from ten stations in the Kadalundiestuary during July 2018 to June 2019 were studied. 87 species were

recorded, 43 belonging to the Phylum Bacillariophyta (Diatoms) and 24 belonging to Miozoa (Dinoflagellates). *Tripos furca* contributed maximum with 7%, followed by *Trieres chinensis* (6%), *Skeletonema costatum* and *Tripos muelleri* (5% each). An average density of 25130 cells/m3 was recorded from the study area with a maximum of 25% in September and a minimum of 1.7% in July. Station wise concentration varied from 9% to 11%.

Balakrishnan *et al*. (2018) reported the halophilic microalgae were collected from the salt pans of Tuticorin, Southeast coast of India, of which 3 were Bacillariophyceae, 4 were Chlorophyceae and 6 were Cyanophyceae. The species diversity decreased sharply when the salinity of the water increased, with *Dunaliella sp.* being the most prominent species in the crystallizing area forming orange-red patches on the salt crystals. Most of the species failed to grow except *Oscillatoria sp*. and *Nitzschia sp*. in the hypersaline region.

Viji *et al*. (2018) studied the physio-chemical properties of water samples collected from various ponds in Tuticorin District, Tamil Nadu. The results revealed that overall water quality was unfit for drinking and irrigation purposes due to industrialization and population expansion. To prevent these problems, an understanding of fundamental water chemistry and other physical parameters is necessary.

Elumalai *et al*. (2013) reported is used Fourier transform infrared spectroscopy (FTIR) and GC-MS to identify and quantify lipids in freshwater microalgae from Cement factories, Ariyalur district. The lipid fractions were extracted from the biomass through different solvent extractions and analyzed for biodiesel. Results of this study showed that eight Microalgal groups produced SFA in high percentage; seven groups had high yields of PUFA and only one group of microalgal contain MUFA.

Oyewumi *et al*. (2018) collected the water samples from ponds at the Federal University of Technology Akure, Nigeria (FUTA) and a pond in Oda Road, Akure in Ondo State, Nigeria. Microalgae were cultured and identified in the laboratory using a microscope, identification keys and algae compendium. Seven (7) microalgae were identified and the growth rates estimated were observed to increase from day 6 to 10 with the maximum peak at day 8. The pH and temperature values on microalgal growth ranged from 7.0 to 8.29 and 23^{o}C to 32^{o}C respectively.

Aragaw *et al*. (2017) investigated the production of microalgae (mixed culture) in photobioreactor configurations using two different media formulations in batch cooperation. The predominant co-cultured freshwater microalgae species *Scenedesmus sp., Chlorella sp., Synedra sp.* and *Achanthidium sp*. were investigated in batch culturing media and the effect of culturing media (BB Medium and BG-11 Medium) for effective algal growth was determined. The maximum biomass concentration was found 0.608 g/L in the Bolt basalt medium and 0.5624 G/L in BG-11 medium for 15 day cultivation time. The amount of time required to adapt the culturing environment is not significantly different and PH range has an effect on mass productions of algae. The optimum pH for high productions of mixed culture microalgae was investigated at pH 8.

Mohanapriya *et al.* (2014) identified microalgae are the most widespread microorganism in freshwater environments and play a vital role in nutrient recycling. Water pollution due to industrialisation has led to the extinction of some species, while eutrophication has caused some microalgal species to overgrow and form algal bloom. They examined the isolation and identification of microalgae from freshwater samples collected from different regions of Noyyal River. 35 green algae, 10 blue green algae, and 4 brown algae were isolated and described.

The present study was carried out in the Korampallam freshwater channel of Puthukottai village, Thoothukudi. Pudukottai is a juction of two main cities (Tuticorin, Thirunelveli and other nearby villages).It is a best commercial and residential place with all facility. Pudukottai is a small Village/hamlet in Thoothukudi Block in Tuticorin District of Tamil Nadu State, India. It comes under Kumaragiri Panchayath. It is located 13 KM towards west from District head quarters Thoothukudi. 11 KM from Thoothukudi Rural. 627 KM from State capital Chennai It is near to bay of bengal. There is a chance of humidity in the weather. The samples were collected from the freshwater channel located nearby bridge.

I. Water Analysis

Physical parameters

The taste, colour, odour and temperature of the water sample were studied.

Chemical constituents

Test for pH

Warm up the instrument for 15min.Calibrate the instrument with the known buffer solutions. (Calibration is done by a buffer solution whose pH is close to that of the sample). Immerse the electrode in the unknown sample, stir for 3min and note the pH.

Test for Alkalinity

Pipette 50ml sample to a Erlenmeyar flask and add two drops of phenolphthalein indicator. If a slight pink color appears, titrate with acid, titrant to a colorless end point and note the reading as 'P' (ml of titrant used for phenopthalein alkalinity). Now add two drops of methyl orange to the same flask and continue to titrate further till the color changes from yellow to orange. Note this reading as 'T' Titrant value of the titrant used for both the titration

Test for Chloride

Pipette 50ml of the water sample (if sea water, take 0.5ml of sample) into a conical flask. Pipette into it, 0.5ml of K_2CrO_4 indicator. This gives yellow color to the sample. Titrate the solution with shaking, against standard silver nitrate solution till the appearance of reddish brown color perform a duplicate titration in an identical

manner.Carry out a blank titration using 50ml of delonized, chloride free water and 0.5ml of the indicator. Subtract this titre value from that obtained for the water sample.

Test for Nitrite

Take a known volume of sample in a 50ml volumetric flask. To it add 1ml of sulphanllamide reagent and mix well. Make up the contents to 50ml by adding distilled water. Shake throughly and measure the absorbance at 543nm against a distilled water blank. Pipette out known concentrations from the standard solution (10 to 100mg). Add reagents as above and draw a standard graph. From the standard graph deduce the amount of nitrite content.

Test for Inorganic phosphorus

Pipette 10ml of the water sample to a test tube. To it add 2ml of mixed reagent and make it up to 15ml with distilled water. Vortex the contents. After 10min. measure the absorbancy at 882nm in a spectrophotometer. Estimate the amount of inorganic phosphorous from the standard curve.

Test for Ammonia

To the sample add 0.4ml of phenol reagent (6) and 0.4ml of nitroprusside reagent (4) and mix well. To it add 1ml of the oxidizing reagent (5) and stopper the tubes immediately. Vortex and incubate for 1hr at room temperature in the dark. Measure the absorbancy at 640nm in spectrophotometer. Prepare a standard graph using different dilutions of the standard graph using different dilutions of the standard solution (1). [Conc. 1 to 10µg] From this find out the ammonia concentration of the sample.

Test for Total phosphorous

Sample digestion for total phosphorous

Take 50 mL of the sample and heat the contents until the volume is reduced to 15ml. Add 1ml of perchloric acid and heat it until the volume is reduced to 5ml. Add 2ml of phenopthalein indicator solution. Then add saturated NaOH solution drop by drop until the solution is turned to pink colour. Make the solution to 50ml with distilled water. Use this sample for the estimation of total phosphorus.

Pipette out known volume of stand and solution to test tubes. (10 to 100µg). Then add 2ml of mixed reagent followed by 2ml of potassium per sulphate reagent and mix well. Incubate for 10mins at room temperatures and read the absorbance of the solution 882nm. Pipette out a known volume as above and measure the absorbance. Fine out the concentration of phosphorous of the unknown sample from the standard curve.

Test for Sulphide

To 7ml of acetate buffer (3.5pH), add 3ml of phenanthroline monohydradte (0.1%) solution. To it add 10ml of water sample. Then make up to 25ml with distilled water and incubate at $25^{o}C$ for 1hr. Run parallel experiment with standard sulphide solutions (10 to 50mg). Read the developed colour at 510nm using suitable blank. Calculate the amount of sulphide by using standard curve drawn, with sodium sulphide.

Test for Calcium and magnesium

Pipette 5 ml of water sample to a 250ml conical flask. To this add 5ml of ammonium buffer and dilute to 100 ml with distilled water. Add a pinch of Erichrome black T and warm the solution to $60^{0}C$. Titrate against EDTA until the red colour turns to blue. Note the end point ‘B’.

II. Degradation of bis(2- ethylhexyl) adipate

1. Media preparation

Chu's medium No. 10 was used to grow the microalgae. The media composition is as follows.

Chemicals	**g/L**
Calcium nitrate	0.232
Dipotassium hydrogen phosphate	0.01
Magnesium sulphate	0.025
Sodium carbonate	0.02
Sodium silicate	0.044
Ferric ammonium citrate	3.5 mg
Citric acid	3.5 mg
Trace metal- 1 mL	
Boric acid	2.4
Manganese chloride	1.4
Zinc chloride	0.4
Calcium chloride	0.02
Copper chloride	0.1
Distilled water	1000 mL

The pH of the medium should be 7.1. The prepared media was autoclaved and stored in glass containers for future use.

2. Identification of microalgae

The microalgal samples were collected from the study area and maintained at refrigerated condition for short term storage. The collected microalgal samples were kept on the glass slide and covered with cover slip and studied under light microscope at 10X, 40X and 100X magnification. The diameter of the microalgae were measured and identified using Floras, and research articles.

3. Isolation of microalgae

The microalgae samples were cultured on agar plates containing Chu's medium No. 10 using spread plate method. The culture plates were incubated at incubation room at 25±2°C with 16 hours light and 8 hours dark cycle provided by cool white fluorescent lights. After 5 to 7 days the grown cultures were seen under microscope and picked the pure colonies. The picked colonies were again kept on agar plates. Repeated this procedure till the pure culture was obtained. The pure cultures were grown massively using Chu's broth medium.

4. Culture condition

The freshwater microalgae *Chlorella sp.* was isolated from the study area.Initially isolated pure colonies were transferred into 10 mL sterile Chu's medium No. 10 and kept inside the incubation room at 25±2°C, under a 16:8 light:dark cycle provided by cool white fluorescent lights. After 3 days of incubation, the algal cells attained sufficient growth; then transfer the whole broth culture 50 mL of fresh broth medium and incubate. After 5 days of incubation, transfer to 100 mL of fresh broth medium and incubate till well growth of algae. Use this as a mother culture and sub-cultured at regular intervals.

5. Experimental setup

The experiment was carried out in triplicate manner in 250 mL Erlenmeyer flasks containing 100 mL of Chu's Medium No. 10 which inoculated with 10% of the actively growing culture of *Chlorella sp.* cells. When the culture attained optical density 0.2 in the absorbance wavelength 750 nm, then it was treated with different concentration of DEHA (20 mg L^{-1},60 mg L^{-1}, 100 mg L^{-1}). A control and solvent control flasks were also maintained. Flasks were manually shaken thrice a day to avoid the adherence of the cells

to the surface of the flasks. Totally 15 days incubation period was given to the culture. Morphological changes in the cells were observed under light microscope (Pancha *et al*., 2013).

6. Growth analysis

Microalgal growth was monitored at regular intervals (0 day, 3rd day, 6th day, 9th day, 12th day and 15th day) by measuringoptical density at 750 nm using UV-vis spectrophotometer.

7. Pigment estimation

For the analysis of pigments content 2ml culture was centrifuge at 10000 rpm for 5 minutes, the supernatant was discarded and 2 ml of absolute methanol was added to the pellet. The content was mixed properly and incubated at 45^0C for 24 h in the dark. The absorbance of the supernatant were read at 470, 652.4, and 665.2 nm and corrected for the turbidity by substracting the absorbance at 750nm. The pigments contents were calculated using the equation (Pancha *et al*., 2013).

$$Chlorophyll\ a(mgL^{-1}) = 16.72\ A_{665.2} - 9.16\ A_{652.4}$$

$$Chlorophyll\ b(mgL^{-1}) = 34.09\ A_{652.4} - 15.28\ A_{665.2}$$

$$Carotenoid\ (mgL^{-1}) = \frac{1000A_{470} - 1.63\ Chlorophylla - 104.9\ chlorophyllb}{221}$$

8. Liquid- liquid extraction

Take 100 ml of water sample in a separating funnel of 2L capacity. Add 10g of NaOH and shake till it get dissolved. Add 50ml of n-Hexane and shake well for 10 minutes. After 30 minutes, discard the n-Hexane layer (impurities) and collect the water

phase. Adjust the pH of water phase to 2 by adding 6M HCl. Add 50 ml of n-Hexane and shake well for 10 minutes for extraction and wait for 30 minutes. Separate n-Hexane layer and stored it. Again add 50ml n-Hexane to the water phase and shake for 10minutes and wait for 30 minutes and separate n-Hexane layer and pool with the previous hexane extract. Add 3g of Na_2SO_4 (anhydrous) into n- Hexane layer for dehydration and leave undisturbed for 20 minutes. Transfer the n-Hexane layer to condensation flask. Concentrate the extract to 5ml by Rotary Evaporator at 35°C and clean up the sample in silica gel column using n-Hexane. Condense elute to 50 ml using rotary evaporator and further to 1ml by passing nitrogen gas. Collect the final extract in a glass vial and store at 4°C prior analyses. Inject 1µl of the sample into GCMS using auto injector for qualitative and quantitative analysis of intermediates of DEHA degradation.

9. GC-MS analysis

The samples were analyzed using a DB 5 MS column (30 m x 0.32 mm ID x 0.25 µm film thickness) using GC–MS. The initial oven temperature was set at 130°C for 5 min, then increased to 200°C at a rate of 8°C per minute. After maintaining at 200°C for 2 min, the temperature was increased to 280°C at a rate of 5°C/min and maintained for 15 min. The injector port and the detector temperatures were 240°C and 250°C, respectively. The peaks were tentatively identified based on the library search report.

RESULT AND DISCUSSION

Physical and chemical constituents of water were essential one to study the quality of water. The survivability of living organisms depends on the physio-chemical composition of water. Water analysis helps to estimate the mineral components of water that is essential to the growth of microalgae. The present study was done in Korampallam channel of Puthukottai, Thoothukudi. The water sample was collected and studied its constituents before the study of phytoplankton. The physical parameters like temperature, odour, taste, and colour were studied. The collected sample was odourless, colourless, tasteless liquid and temperature was 29^{o}C in December, 2022. The pH of the sample was 7.3 and salinity was 13ppt (Table 1). Chemical parameters like chloride (40.9 mg L^{1}), nitrite (5-24 mg L^{-1}), inorganic phosphorous (27.3mg L^{-1}), ammonia (16.7 mg L^{-}), sulphate (0.145 mg L^{-1}), shulpide (21.9 mg L^{-1}), calcium (440 mg L^{-1}), and magnesium (150 mg L^{-1}) were studied and the experiments confirmed that the presence of all above mentioned chemicals but at different ratios. Ammonia, nitrate, and phosphate are nutrients that support the fertility of water, one of the factors that determine water quality. Enrichment of nutrients in the waters causes an increase in the population of phytoplankton (Gypens *et al.,* 2009; Jones-Lee and Lee, 2005), reduces the concentration of dissolved oxygen, decreases biodiversity, and sometimes increases the potential for the growth of harmful phytoplankton species (harmful algal blooms) (Susanti *et al.,* 2022; Jones-Lee and Lee, 2005).

The microalgal strains of the study area were studied. The algal samples was collected by scraping rocks, fine gravel, waste clothes, moist soil, pebbles, and twigs. Totally 13 genera was identified by using light microscope. Of these identified microalgae 8 were from Basillariophyceae, 4 from Chlorophyceae and 1 from Cyanophyceae (Plate 1).

Table 1. Physio-chemical parameters of freshwater sample collected from Korampallam channel of Pudhukottai

Physical parameters	
Colour	Transparent to pale yellow in colour
Taste	Tasteless
Odour	Odourless
Temperature	28^{o}C
Chemical parameters	
pH	7.3
Salinity	13 ppt
Alkalinity	92 mg/L^{-1}
Sulphide	21.9 mg/L^{-1}
Ammonia	16.7 mg L^{-1}
Nitrite	5.24 mg L^{-1}
Inoranic phosphorous	27.3 mg L^{-1}
Sulphate	0.145 mg L^{-1}
Chloride	40.9 mg L^{1}
Calcium	440 mg L^{-1}
Magnesium	156 mg L^{-1}

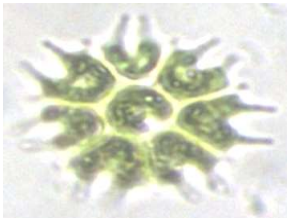
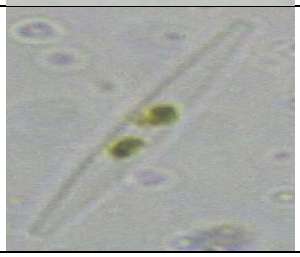
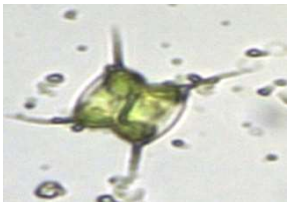
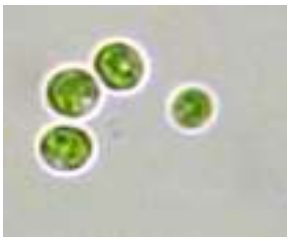
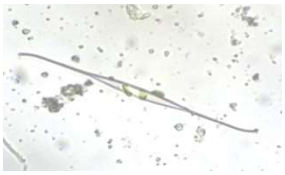
Cyclotella sp.		*Gomphonema sp.*	
Cymbela sp.		*Nitszchia sp.*	
Navicula sp.		*Ocilatoria sp.*	
Naviculv sp.		*Pediastum sp.*	
Syndera sp.		*Scenedesmus sp.*	
Amphora sp.		*Chlorella sp.*	
Closterium sp.			

Plate 1. Microalgal diversity of Korampallam channel of Puthukottai under light microscope with 100 X magnification

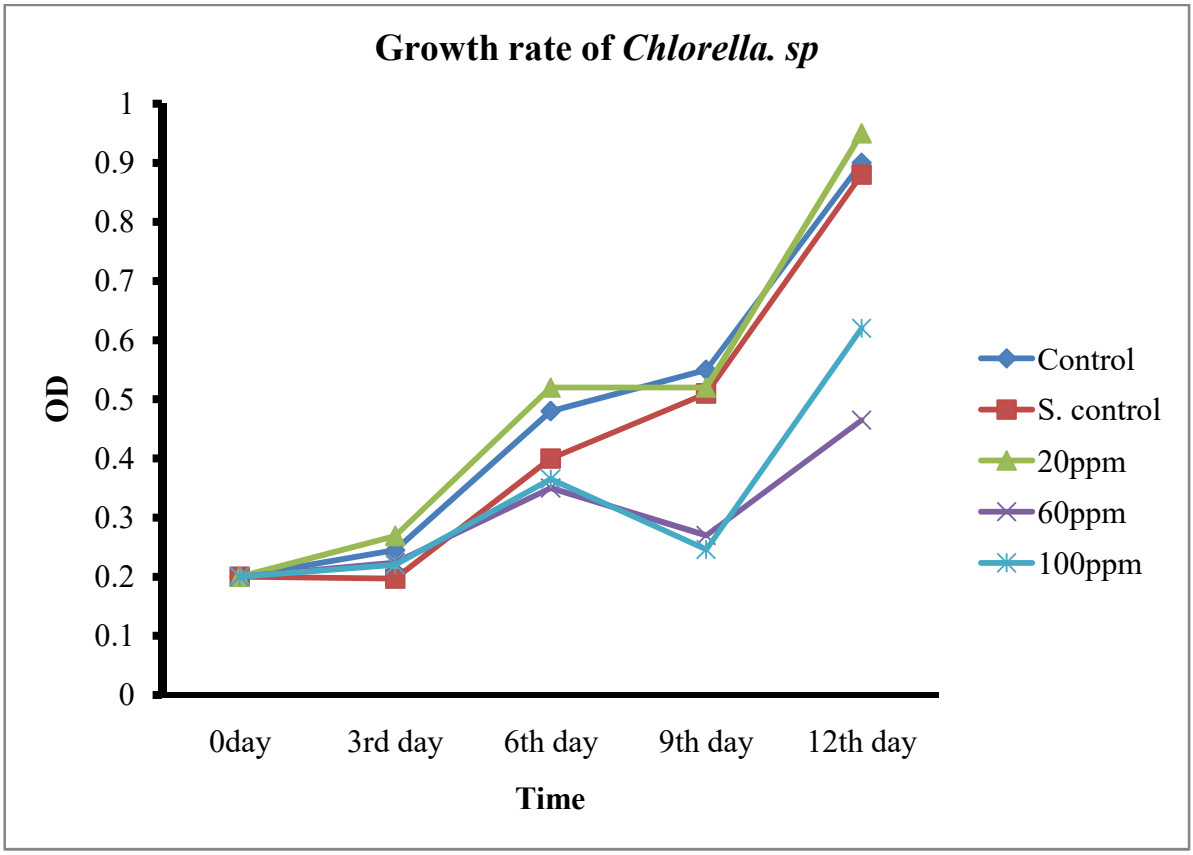


Fig. 1 Determination of growth rate of *Chlorella sp* after BEHA treatment

Table 2. Estimation of pigments from bis(2-ethyl hexyl)adipate treated *Chlorella sp.*

Samples	Chlorophyll a	Chlorophyll b	Carotenoid
3rd day			
Control	3.11±0.98	6.54±3.02	1.56±0.50
Solvent control	2.98±1.19	3.24±1.79	0.84±0.25
20 mg L^{-1}	2.84±1.32	4.54±1.08	0.76±0.05
60 mg L^{-1}	2.28±0.06	4.45±0.28	0.87±0.01
100 mg L^{-1}	2.24±0.68	4.961.84	1.06±0.45
6th day			
Control	4.92±0.77	10.07±0.32	2.10±1.11
Solvent control	3.18±0.54	6.45±2.17	0.72±0.69
20 mg L^{-1}	2.55±1.61	5.67±2.38	1.47±1.36
60 mg L^{-1}	2.03±0.49	3.81±1.09	0.84±0.31
100 mg L^{-1}	2.51±0.16	4.46±0.31	0.79±0.12
9th day			
Control	3.35±0.79	3.72±0.78	0.67±0.40
Solvent control	2.32±0.94	4.34±1.78	0.71±0.39
20 mg L^{-1}	4.6±2.15	5.79±3.62	1.04±0.27
60 mg L^{-1}	2.09±0.54	2.70±1.54	1.12±0.79
100 mg L^{-1}	1.05±0.75	3.36±0.75	0.72±0.34
12th day			
Control	6.33±2.18	4.79±2.19	0.79±0.14
Solvent control	2.30±1.87	6.58±3.49	1.45±1.92
20 mg L^{-1}	3.90±2.45	6.07±0.44	0.69±0.76
60 mg L^{-1}	0.86±0.89	4.8±1.28	1.27±0.65
100 mg L^{-1}	1.64±0.14	3.80±0.16	0.74±0.34

Similar results were obtained by Mohanapriya and Geetharamani, 2014 and Selvaraj *et al.,* 2021. The microalgal strains were cultured using Chu's medium No. 10 for isolation of pure colonies. *Chlorella sp.* was isolated through spread plate method. The isolated pure culture was grown in broth media for further uses. Totally 39 genera of 70 species were identified by Narchonai *et al.,* 2019 and they resulted that, most of the species fall under Chlorophyceae followed by Cyanophyceae and Bacillariophyceae. Halder *et al.,* 2019 reported that the members of Chlorophyceae and Cyanophyceae were dominant in winter season than in late summer. Hence based on the seasons the species richness may change.

Plasticizers are the emerging contaminants present in aquatic environments. The main aim this current investigation was to degrade the plasticizer using microalgae. Hence the microalgae like *Chlorella sp.* was isolated as a pure strain from the study area and treated with different concentrations (20mg L^{-1}, 60 mg L^{-1} and 100 mg L^{-1}) of bis (2 ethylhexyl) adipate (BEHA) and incubated for 12 days (Plate 2). The growth rate and pigments were estimated at regular intervals and the data revealed the tolerance of *Chlorella sp.* on BEHA pollution. 20 mg L^{-1} concentration of BEHA showed good and consistent growth throughout the incubation period while other shows less growth (Fig. 1). The pigment content of BEHA treated *Chlorella sp*. showed different amount of Chlorophyll a, b and carotenoids. The leaching and production of pigments were based on the toxicity. 20 mg L^{-1} BEHA treated *Chlorella sp.* showed high chlorophyll a (4.6 µg ml^{-1}), chlorophyll b (5.79 µg ml^{-1}), and carotenoids (1.04 µg ml^{-1}) content than control, solvent control and all other concentrations at 9 days incubation shown in Table 2. Similar results were obtained by Chi *et al.,* 2019. According to them, the DEP and DBP degradation was more quickly done by a microalgae *C. closterium* than by *C. muelleri* and *D. salina*. The concentration of the plasticizer should be relevant to the

environmental concentration (0.1 mg L^{-1}) of pollutant. The removal of organic pollutants involved a rapid initial passive physiochemical adsorption followed by active absorption, accumulation and degradation (Gao *et al.,* 2011). The degradation of plasticizer revealed that the microalgae were one of the best agents to remove the contaminants from the aquatic environment. Therefore, understanding of metabolic pathway of pollutants by algae is useful for risk assessments. In this work, freshwater microalgae were applied for the investigation of biodegradation pathway of BEHA in aquatic environment. When BEHA was treated with the *Chlorella sp*, high removal rate was observed. It was detected that cellular uptake was the predominant mechanism for the depletion of BEHA by *Chlorella sp*, while biotransformation accounted for the elimination of BEHA by the other species. The GCMS analysis revealed that *Chlorella* sp. have the capability to degrade the toxic plasticizers to less toxic compounds and it also release methane, CO_2 during degradation process. The microalgae either involve bioaccumulation, or biodegradation of plasticizer during the incubation period and also using the chemical constituents of BEHA as a energy source. Due to this effect, the toxicity of the chemical was reduced meanwhile the microalgae was benefitted.

SUMMARY AND CONCLUSION

The increased usage of plastic materials has led a severe threat to aquatic environment. The micro and nano-plastics are one of the major pollutants arose from macroplastics, meanwhile the additives of plastics especially plasticizers were leached out from macroplastics. The negative impacts of these materials are eliminated using remediation approaches. But the degradation of such emerging contaminants was challenging one. Hence the ultimate aim this present study to degrade the BEHA using microalgae. Initially the water sample from the study area was analyzed and found that the presence of nutrients that were essential for microalgal species richness. After that the microalgal strains (13 species) were identified through this study and pure *Chlorella sp.* was also isolated. Further study was done with BEHA at various concentrations with *Chlorella sp.* and discovered that the selected strains have the ability to degrade BEHA at 20 mg L^{-1} concentration, whereas beyond this level the toxicity of BEHA helped to bleach the algae. The pollutants generally enter into different bio-cycles hence broken into threat less compounds due to the interference of microorganisms. This research summarizes that the microalgae has the potentiality to degrade the emerging pollutants present in aquatic environment while at higher concentration the algal culture windup due to heavy toxicity. Furthermore research should be needed to understand the large scale degradation of BEHA and genes responsible for degrading BEHA.

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BIGNONIACEAE: A COMPARATIVE MORPHOANATOMICAL EVALUATION

A dissertation submitted to

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affiliated to **MANONMANIAM SUNDARANAR UNIVERSITY**

in partial fulfilment of the requirements for the Degree of

Master of Science in Botany.

By

B. MARIA ANSHEELA

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ST. MARY'S COLLEGE (Autonomous)

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2022-2023

CERTIFICATE

This is to certified that is this short term project work entitled dissertation entitled **BIGNONIACEAE: A COMPARATIVE MORPHOANATOMICAL EVALUATION** submitted to **ST. Mary's College (Autonomous)** affiliated to **MANONMANIAM SUNDARANAR UNIVERSITY** in partial fulfilment of the requirements for the Degree of Master of Science in Botany and is a record of work done in the department of Botany **ST. Mary's College (Autonomous)** THOOTHUKUDI during the year 2022-2023.

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DECLARATION

I do here by declare that this dissertation entitled **BIGNONIACEAE: A COMPARATIVE MORPHOANATOMICAL EVALUATION** Submitted by me in partial fulfilment for the award of the degree of **'Master of Science in Botany',** in the result of my original and independent work carried out under the guidance of **Dr. Mrs. S. Beulah Jerlin M. Sc, M.Phil., Ph.D.** Assistant Professor. Department of Botany, St. Mary's College (Autonomous) THOOTHUKUDI and it has not been submitted elsewhere for the award of any other degree.

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B. MARIA ANSHEELA

CONTENT

INTRODUCTION

Today, it is very important to identify plants that exist in nature and to know their species. It is important to determine the type of plant in various fields such as horticulture, botanical research, aromatic herbs, farming, floristry, and so on. Plants are found all over the world, even where humans don't live. Plants are an integral part of the ecosystem. Many plant species are at risk of extinction due to deforestation. Plants are essential for human and other living things to exist. They are useful as a foodstuff, medicine, and in many other industries (Amlekar *et al*., 2014).

Identifying plants can help preserve and survive all natural life. Plant identification can be accomplished with the leaves of the plant using many different techniques. Leaves are more useful for classifying plants, such as flowers that are available for a short time as they are more readily available than other biometric components. Plant classification using leaves requires the knowledge of different biometric properties of the leaf such as color, shape, texture and vascularization. This definition manually is time consuming and expensive. Leaves can be classified by color including the similarity between the two images with the help of a color histogram, but color-based classification depends on the season and the influence of sunlight (Amlekar *et al*., 2014).

Ornamental plants are plants that exhibit visual activity with their flowers, leaves, fruits and forms or stand out with these features and are grown for ornamental purposes. Though it is not correct to examine the exact history of ornamental plants in certain periods, by the same token its use as an object of beauty in past times goes back to a time as old as the existence of civilizations in shaping gardens as an art form, expressing philosophical and religious beliefs (Jaggi, 2013; Baktır, 2013). Albeit cultivation of plants in culture for use as ornamental plants dates back to ancient times,

ornamental plants became an important sector in the world at the beginning of the 20th century. Ornamental plants are divided into 4 groups according to their intended use - cut flowers, outdoor ornamental plants, indoor ornamental plants and flower bulbs (Ay, 2009; Kazaz, 2012)

Ornamental plants can provide multiple profits with regards to environment beauty, economy concern and human lifestyles. Ornamental plants attract pollinators which can feed them through nectar present in their flowers that has high nutritive value for humans too. Despite the increasing interest of ornamental plants, some of them are also cultivated for their medicinal use as they have many bioactive compounds like phenolic compounds, carotenoids, antioxidants, essential oils and other secondary metabolites(Goleniowski M *et al*., 2013 and Kaushik P *et al*., 2015) Ornamental plants like *Ocimum* sp., *Nicotiana* sp., *Ixora, Aloe vera, Agave,* etc. and ornamental flowers like roses, nasturtium, hibiscus, marigold, *Calendula,* etc. are commonly grown in homes which also have many medicinal applications. Along with this, remedies from plants can be much cheaper and protect against free radicals without any side-effects than medicines formed by pharmaceutical companies.

Trees, shrubs, seasonal annual and perennial plants, other species used as ground cover and ornamental grasses are considered in this class (Karagüzel *et al*., 2010; Kazaz, 2012).

Indoor and outdoor spaces, with their visual-aesthetic aspects, are the main areas of use of ornamental plants. It has aesthetic and renovation uses in landscape works. Some plants have medicinal values due to their biochemical content of their underground parts (root-tuber bulb), as well as their leaves, flowers and fruits. Some ornamental plants are at risk in terms of biodiversity due to the above mentioned characteristics. In order for such plants to continue their generations in first place, it is

necessary to take them under protection, to establish a gene bank, to apply and develop methods for production. In this sense, determining and identifying the natural habitats of plants is very important as a first step.

Plant morphology or Phyto morphology is the study of the physical form and external structure of plants, whereas plant anatomy is the study of internal plant structure, mostly at the cellular/microscopic level (Armando Carrillo-Lopez and Elhadi M. Yahia. 2019)

Unlike botanists, who make a clear and formal distinction between plant morphology and plant anatomy (the former referring to whole-plant form and the organization of parts, the latter to the fine structure, or histology, of parts), zoologists are uncharacteristically vague in their usage of these terms.

Morphology "deals with the form of living organisms, and with relationships between their structures" (from the Greek term morpho), whereas anatomy is "the science of the structure of the bodies of humans, animals, and plants" (derived from the Greek terms anaand -tomy, meaning "repeated cutting") (Brown, 1993). Morphology is the study of "form," which can be generalized to all hierarchical levels, from organelle to whole organism. It is also concerned with the relationships among structures; hence it includes emergent features of form such as relative size, allometry, and even function and physiology.

Morphology of plant species is extremely essential field for all types of research because without it no one can understand about the plant and its characters.Similarly anatomy is also very needed as in the old days scientists were used this tool for the identification of the plants (Phillips *et al.,* 2009).

Anatomy, a field in the biological sciences concerned with the identification and description of the body structures of living things. Gross anatomy involves the

study of major body structures by dissection and observation. “Gross anatomy” customarily refers to the study of those body structures large enough to be examined without the help of magnifying devices, while microscopic anatomy is concerned with the study of structural units small enough to be seen only with a light microscope. Dissection is basic to all anatomical research. The earliest record of its use was made by the Greeks, and Theophrastus called dissection “anatomy,” from anatemnein, meaning “to cut up.”

Comparative anatomy, the other major subdivision of the field, compares similar body structures in different species of animals in order to understand the adaptive changes they have undergone in the course of evolution.

Bignoniaceae is a family of flowering plants commonly known as the bignonias or trumpet veins and also noted for ornamentals. Bignoniaceae is a moderate size group of shrubs, lianas, climbers and trees that are distributed in pantropical regions, especially in the new world tropics, with a few temperate taxa. The family consists of 110 genera and 650 species (WISC 1999).

The major economic contributions of the Bignoniaceae are to medicine and ornamental wood uses in various countries. The medicinal value of some members of the Bignoniaceae in West Africa is well documented by authors such as Oliver (1960), Irvine (1961), Ayensu (1978) and Burkill (1987), among others.

The Bignoniaceae belong to the Lamiales (core eudicots; APG III 2009) and forms a monophyletic group of plants (Spangler and Olmstead 1999; Olmstead *et al*., 2009) with a pantropical distribution and center of diversity in the Neotropics (Gentry 1980). Only a few species occur naturally in temperate or Neotropical montane regions (e.g., *Bignonia capreolata*, Catalpa section Catalpa spp.; Fischer *et al*., 2004; Lohmann 2004; Li 2008). The systematic relationships among members of the

Bignoniaceae have been carefully explored in the last few years (Zhjra *et al.*, 2004; Lohmann 2006; Grose and Olmstead 2007a, 2007b; Li 2008), and a robust phylogeny and classification for the entire family is now available (Olmstead *et al*., 2009). The Bignoniaceae is well known, since many of its members have economical (Record and Hess 1943; Mainieri and Chimelo 1989) and ornamental importance due to their showy tubular flowers (eight countries have Bignoniaceae as their national trees; Gentry 1992; Lohmann 2004). The family is composed mainly of trees and lianas, with a few shrubs and herbs (e.g., Argylia in the Andes and Incarvillea in the Himalayas, both herbaceous; Fischer *et al.*, 2004).

Three ornamental plants from the Bignoniaceae are the subject of the current study, which focuses on their morphological and anatomical characteristics. Plant biologists employ plant morphological traits to distinguish between diverse plant species, classify them, and describe them as well as how they are related to one another. Anatomical characteristics can also be used to identify plant species.

SCOPE AND OBJECTIVES

Plants have been used for centuries to treat a variety of ailments. Ornamental plants are plants grown primarily for their aesthetic value; however, many ornamental plants are used as folk medicine in the treatment of various diseases. As a result, it is critical to gain a thorough understanding of the medicinal properties of these ornamental plants (Larbie and Abboah-Offei, 2014). Herbal medicines are becoming more popular in the modern era as people's faith in natural therapies develops. Understanding the chemical constituents of plants can aid in the discovery of therapeutic agents. However, it is critical to work with locally available resources to bring out their pharmaceutical values for use as a source of nutrient supplements. However, it is critical to focus on locally available resources in order to maximise their pharmaceutical value for use as nutrient supplements, minerals, vitamins, enzymes, and antimicrobials in medicine. As a result, the current investigation was launched with the following goals in mind.

- To identify morphological characteristics.
- To assess various pharmacognostical parameters such as microscopic and macroscopic characteristics of *Tecoma stans, Tabebuia rosea,* and *Millingtonia hortensis*, this can then be used to provide useful information about the drug's quality.
- To understand the anatomical characteristics of plants.

LITERATURE REVIEW

Bignoniaceae are most noted for ornamentals, such, *Tabebuia* and *Spathodea*, grown for their conspicuous, tubular flowers. George *et al.*, (2005).

Bignoniaceae Juss. are widely distributed in tropical and subtropical regions. Most of their Neotropical species have a climbing habit, which makes the family one of the most ecologically important in the Americas. This study of pollen morphology from 23 Bignoniaceae species in Brazilian forest fragments aimed to investigate new pollen characteristics of the family by light and scanning electron microscopy, which can assist in delimitating Bignoniaceae species and taxonomy. Another aim of the study was to analyze the evolution of the family's pollen characteristics. The pollen grains were acetolyzed, measured and photographed. We describe the pollen grains based on qualitative data, and quantitative data were analyzed using descriptive and multivariate statistics. Based on the qualitative data, an ancestral reconstruction of pollen morphology characteristics was performed for the taxa analyzed. We used the pollen data to understand the relationships established by previous phylogenies. The pollen grains are monads or tetrads, apolar or isopolar, and medium to large, with circular, subcircular to subtriangular amb; oblate to oblate-spheroidal; inaperturate, 6-colpate, (7)-8-(9)-colpate or 3-colporate with short or long colpi that are narrow, wide or very wide and with or without margo; lalongate endoapertures sometimes not evident; and psilate perforate, microreticulate or reticulate exine, homo- or heterobrochate, with simpli or duplicolumellate muri. This result confirms Bignoniaceae as a eurypalynous family, and it allows the identification of pollen morphological characteristics shared by genera and species studied. Cintia *et al.*, (2019).

Most Madagascar Bignoniaceae have indehiscent fruits and have been assigned to the tribe Crescentieae. As thus constituted, the Crescentieae are remarkably disjunct between Central America (and the West Indies) and Madagascar with a single monotypic genus also on continental Africa. This paper analyzes the evolutionary relationships of the Madagascar Crescentieae and concludes that they are descended from a different ancestral stock than the New World Crescentieae. A revised tribal taxonomy is proposed to reflect phylogenetic relationships with Crescentieae restricted to the neotropic species and Coleeae resurrected for the indehiscent-fruited Madagascar species. Alwyn (1976).

Nuptial nectary characteristics were investigated in 37 taxa of Bignoniaceae. A nuptial nectary associated to the floral axis was found in all species. Two main types can be distinguished according to their degree of development and functionality: 1) vestigial and non-secretory and 2) well-developed and secretory. The former is characteristic of Clytostoma spp., while the latter is found in the remaining species. Two subvarieties of the secretory type of nectary can be discerned according to their position and shape: 1) annular, found in *Adenocalymma, Amphilophium, Anemopaegma, Arrabidaea, Dolichandra, Eccremocarpus, Macfadyena, Melloa, Pithecoctenium, Tabebuia,* and *Tecoma,* and 2) cylindrical, found in *Argylia, Cuspidaria, Jacaranda, Mansoa, Parabignonia, Pyrostegia,* and *Tynnanthus*. Anatomically, two tissues are distinguished: 1) a single-layered epidermis covered by a cuticle and a variable number of stomata, and 2) a secretory tissue composed of compactly arranged parenchyma cells. Both nectary size and nectary/ovary ratio were usually larger in lianas (Bignonieae) than in trees (Tecomeae). Nectary type proved to be consistent among species of same genus but not among genera of same tribe. Nectary features such as vascularization, presence of trichomes and nectary type were

constant within the analyzed species and therefore have a reliable taxonomic value. Guillermo (2000).

Ogundipe and Wujek (2003) Studied and compared the foliar anatomy of the 12 genera (16 species) of Bignoniaceae family members. Across the family, anatomical characters found to be most useful are: stomata typs, trichomes in the adult material, presence of peristomatal folds, type sinuosity of epidermal anticlinal wall, veinlet termination number, cuticular striation, presence of sclerenchymatous idioblasts in the mesophyll, presence of hypodermis, presence of sclerenchymatous fibres in the ground parenchyma and presence of collenchyma in the outer tissue of petiole.

Usually trees or lianas, sometimes shrubs, rarely herbs; lianas mostly with unusual vascular structure. Leaves opposite, sometimes verticillate, rarely alternate, 2-3-foliolate, pinnate or palmate, less often simple, terminal pinna of liana species often modified into a tendril; axillary bud scales (pseudostipules) often present. Flowers in thyrses, racemes or solitary, terminal or axillary, usually conspicuous. Calyx with five sepals, sometimes bilobed or unlobed, rarely with calyptra. Corolla with five petals, often 2-lipped, rarely subrotate, imbricate or rarely valvate. Androecium attached to tube, stamens 4, didynamous in 2 pairs, the fifth (adaxial) stamen staminodial or absent, rarely all 5 stamens fertile or 2 fertile and 3 staminodial. Ovary withannular or cupular nectary disc, style with 2-lobed stigma, ovary superior, 2-carpellate, bilocular with a separating septum, sometimes unilocular or 4-locular, placentation axile or in unilocular ovary with 2 or 4 intruding parietal placentae, ovules numerous, anatropous or hemitropous. Fisher *et al.,* (2004).

Ugbabe *et al.,* (2008) compared the leaf epidermal features of eleven species of the family Bignoniaceae in Nigeria. The species are relatively uniform in the

qualitative macro morphological characters except in the leaf shape, which varies from ovate, elliptic, oblong-elliptic, oblong, oblanceolate to obovate-lanceolate. A more constant macro character for the species is the leaflet length /leaflet width ratio, which ranges from 2:1 to 4:1. The epidermal morphology of the adaxial and abaxial surfaces of the species was studied with the light microscope. The epidermal cells are polygonal, irregular or both. Anticlinal walls are straight, curved or undulate/ wavy. Leaflets of all species are hypostomatic with stomata restricted to the abaxial surface. The Anomocytic stomata type is most prominent except *Kigelia africana*, which has diacytic stomata. Striae are present on the adaxial surface of *Oroxylum indicum* and abaxial surface of *Spathodea campanulata*. Knobs are present on the abaxial and adaxial surfaces of *Markhamia lutea, Markhamia tomentosa*, abaxial surface of *Stereospermum kunthianum* and adaxial surface of *Tabebuia rosea*. Other features of the epidermis that show variation include stomatal size, shape and frequency. Epidermal cell shape, anticlinal wall undulation, striation on the epidermis, stomata type, distribution and stomata index are of taxonomic importance in the family while epidermal size and number are of little diagnostic value. The significance of these observations is discussed in relation to the taxonomy of the family.

Choudhary *et al.*, (2009) observed the studies on leaf epidermal micromorphology wood element character and phytochemical screening of three medicinally important taxa of the family convolvulaceae.

Bignoniaceae are woody, trees, shrubs, and lianas found in all tropical floras of the world with lesser representation in temperate regions. Phylogenetic analyses of chloroplast sequences (rbcL , ndhF , trnL-F) were undertaken to infer evolutionary relationships in Bignoniaceae and to revise its classification. Eight clades are recognized as tribes (*Bignonieae, Catalpeae, Coleeae, Crescentieae, Jacarandeae,*

Oroxyleae, Tecomeae, Tourrettieae); additional inclusive clades are named informally. *Jacarandeae* and *Catalpeae* are resurrected; the former is sister to the rest of the family, and the latter occupies an unresolved position within the "core" Bignoniaceae. Tribe *Eccremocarpeae* is included in *Tourrettieae*. Past classifications recognized a large *Tecomeae*, but this tribe is paraphyletic with respect to all other tribes. Here *Tecomeae* are reduced to a clade of approximately 12 genera with a worldwide distribution in both temperate and tropical ecosystems. Two large clades, *Bignonieae* and *Crescentiina*, account for over 80% of the species in the family. *Coleeae* and *Crescentieae* are each included in larger clades, the Paleotropical alliance and Tabebuia alliance, respectively; each alliance includes a grade of taxa assigned to the traditional *Tecomeae*. Parsimony inference suggests that the family originated in the neotropics, with at least fi ve dispersal events leading to the Old-World representatives. Richard *et al.,* (2009).

Atiq Mehsud *et al.,* (2013) studied the morphology and anatomy of seven most common weed species infesting agricultural and non-agricultural lands of rainfed area of Bannu region were investigated during 2012. The study included *Datura metel L., Euphorbia hirta L., Fagoniacretica L., Heliotropium europaeum L., Parthenium hysterophorus L., Solanum surattenseBurm f. and Withania somnifera (L.) Dunal.* Due to some special morphological and anatomical features, the capacity of rapid absorption of water along with minerals from the soil may be facilitated to compensate the rapid water loss, and thus can also be regarded as common xerophytes. Their morphological, anatomical and histological characteristics are suitable for their successful growth in rain-fed condition of the region.

Pace *et al.,* (2013) Review the important aspects in classical wood anatomy and evolution and test hypotheses regarding patterns of wood evolution in

Bignoniaceae members. Altogether, 85% of the genera currently recognized in Bignoniaceae were sampled, and 30 characters were delimited and mapped onto a robust phylogeny of the family. Some patterns of wood evolution within the Bignoniaceae seem to have been shaped by ecophysiological and habit aspects in the family. For example, vessels increase in diameter in the lianoid lineages but decrease in trees and shrubs during evolution. Rays in trees have evolved from a mixture of homo- and heterocellular to exclusively homocellular and storied in some lineages, while in the lianas the opposite pattern was recorded.

Shinde *et al.,* (2014) studied some parameters such as morphological, microscopical, physicochemical evaluation, florescence analysis; preliminary phytochemical analysis, thin layer chromatographic study and antimicrobial potential of alcoholic extract of *P. grandiflora* were carried out. Macroscopically the leaves are fleshy leaves, watery, needle shape. Flowers are racemes form; fruits are ovoid with small black coloured seed. Chemomicroscopy revealed the presence of Rubiaceous stomata in leaf; Rosette calcium oxalate crystals and protoplast in mesophyll of leaf, cortex and pith of stem and root; pink colored cuticle of stem, collateral vascular bundle with lignified xylem, abundant of starch grains and mucilaginous cells in all aerial parts.

Silvia Netala *et al.,* (2014) compared the structural features and physicochemical properties of three species of *Portulaca.* Different Parts of *Portulaca* were examined for macroscopical, microscopical characters. Physicochemical, phytochemical and fluorescence were also analysed. The plants are succulent, prostrate herbs. Usually roots at the nodes of the stem. Leaves are opposite with paracytic stomata and characteristic Kranz tissue found in C-4 plants. Abundant calcium oxalate crystals are present in all vegetative parts of the plant. Quantitative

determinations like stomatal number, stomatal index and veinlet number is were performed on leaf tissue.

Hameeda *et al.,* (2014) studied the morphological and anatomical adaptations of the plant *Acacia nilotica* (Mimosaceae) and *Dodonaea viscose* (Sapindaceae). The morphological adaptations were observed by dense hair, powder and cuticle layers on leaves and stem. The leaves were found to be leathery, needle like and elongated shapes. Sometimes, the leaves were found to be modified into thorns and spines. Similarly, thick and short rhizome, compact epidermis, wide cortex and many water storing tissues were observed.

InSun Kim (2014) studied the vegetative and reproductive morphology and anatomy of two Hawaiian endemic *Portulaca* species were examined. Specifically, *P. molokiniensis* and *P. sclerocarpa* were compared to closely related species in the genus. The comparisons were both qualitative and quantitative, using characteristics of leaves, stems, roots, and fruits. Tissue organizations of vegetative and reproductive parts of the plants were assessed using micro technique procedures, statistical analysis, and scanning electron microscopy. The most notable features of these two species were (1) the size and frequency of stomata in *P. molokiniensis*, and (2) the large number of sclerenchymatous cell layers in the thickest fruit walls of *P. sclerocarpa*. These findings may imply that stomata development in *P.molokiniensis*and thick fruit wall development in *P. sclerocarpa* are evolved features of survival.

The circumscription of Bignoniaceae genera and tribes has undergone major changes following an increased understanding of phylogenetic relationships within the family. While DNA sequence data have repeatedly reconstructed major clades within the family, some of the clades recovered still lack diagnostic morphoanatomical features, complicating their recognition. In this study we

investigated the wood anatomy of all major lineages of Bignoniaceae (except Tourrettieae) in search for anatomical synapomorphies for clades. We sampled 158 species of Bignoniaceae, representing 67 out of the 82 genera currently recognized. Detailed descriptions of quantitative and qualitative wood anatomical features are presented for each clade and interpreted in the light of a molecular phylogeny for the family. Jacarandae are characterized by a paratracheal winged-aliform parenchyma, with the traditional subdivision of Jacaranda into sections Monolobos and Dilobos supported by the uniseriate and homocellular rays of Monolobos versus the wide and heterocellular rays of Dilobos. Tecomeae s.s. are characterized by scanty paratracheal parenchyma, septate fibers, and heterocellular rays, traits also found in Delostoma, a genus previously included in Tecomeae s.l., but recently shown to represent a separate lineage. Crescentiina includes two subclades, the Tabebuia alliance and the Paleotropical clade, which share abundant aliform parenchyma, short and mainly homocellular rays, less commonly with heterocellular rays with body procumbent and one row of marginal square cells. Members of the Tabebuia alliance and the Paleotropical clade can be distinguished from each other by the narrow vessels with a widespread storied structure found in members of the Tabebuia alliance, versus the vessels with medium to wide width and a non-storied structure found in members of the Paleotropical clade. Oroxyleae are characterized by a combination of simple and foraminate perforation plates and homocellular rays, while Catalpeae are characterized by scanty paratracheal parenchyma, abundant tyloses and vessel-ray pits simple to semi-bordered. Bignonieae differ from all other clades by a variant secondary growth and a typically lianoid wood anatomy. Overall, wood anatomical characters are not very labile within the family, being distributed across clades in a very predictive manner. Several anatomical characters represent good anatomical

synapomorphies and provide further support to clades identified in molecular phylogenetic studies Marcelo *et al.,* (2014).

Agarwal *et al.,* (2014) described the micro morphology and physio chemical parameters of the leaves of *Tecoma stans* L. (Bignoniaceae). Leaves are opposite, compound and imparipinnate with 2 to 5 pairs of leaflets and a larger single terminal leaflet. Leaflet is lanceolate up to 10cm in long with serrate margin, mild green in colour. Microscopic evaluation revealed the abaxial epidermis is stomatiferous, the stomata are actinocytic type. Each stoma has 5-10 µm, wedge shaped radiating subsidiary cells all ground the guard cells are elliptic with wide opening. The vascular system is quite prominent and occupies entire midrib. It consists of a wide deep bowl-shaped main strand and a flat thick adaxial plate of strand. The trichome has a short central stalk, and a thin plate of glandular cells, lateral vein surrounded by a thin layer of parenchyma cells which extend in to adaxial vertical pillar of cells. Microscopic analysis was informative and provides useful information in the botanical identification, standardization for purity & quality and immense value in authentication of the leaf.

The most recent classification of Bignoniaceae recognized seven tribes, Phylogenetic and monographic studies focusing on clades within Bignoniaceae had revised tribal and generic boundaries and species numbers for several groups, the portions of the family that remain most poorly known are the African and Asian groups. The goal of the present study is to identify the primary lineages of Bignoniaceae in Egypt based on macromorphological traits. A total of 25 species of Bignoniaceae in Egypt was included in this study along with *Barleria cristata* as outgroup. Parsimony analyses were conducted using the program NONA 1.6, preparation of data set matrices and phylogenetic tree editing were achieved in Win

Clada Software. The obtained cladogram showed that within the studied taxa of Bignoniaceae there was support for eight lineages. Usama and Abdel (2014).

Mass flowering is a widespread blooming strategy among Neotropical trees that has been frequently suggested to increase geitonogamous pollination. We investigated the pollination ecology of the mass-flowering tree *Handroanthus impetiginosus*, addressing its breeding system, the role in pollination of different visitors, the impact of nectar robbers on fruit set and the function of colour changes in nectar guides. This xenogamous species is mainly pollinated by *Centris* and *Euglossa* bees (Apidae) seeking nectar, which are known to fly long distances. The flowers favour these bees by having: (1) a closed entrance in newly opened flowers which provides access only to strong bees capable of deforming the flower tube; and (2) a nectar chamber that is accessible only to long-tongued bees. Only first-day flowers with yellow nectar guides produce nectar. Pollinators prefer these flowers over second and third-day flowers with orange and red nectar guides, respectively. Nectar robbers damage two-thirds of the flowers and this robbing activity decreases fruit set by half. We attribute the low fruit set of *H. impetiginosus* to the intense nectar robbing and hypothesize that visual signalling of nectar presence in newly opened (receptive) flowers reduces geitonogamy by minimizing bee visits to unrewarding (non-receptive) flowers. Clemens *et al.*, (2014)

Sohail Ahmad *et al.*, 2014 studied the crude and numerous fractions of leaves, stem, and roots of the plant were investigated for phytochemical analysis and DPPH radical scavenging activity. Phytochemical analysis of crude and fractions of the plant revealed the presence of alkaloids, saponins, tannins, steroids, terpenoids, flavonoids, glycosides, and phenols. The antioxidant activity of various extracts was resolute against DPPH radical with the avail of UV at 517 nm. The stock solution

(1000mg/mL) and then several dilutions of the crude and fractions were prepared. Ascorbic acid was used as a standard. The plant leaves (52.59 ± 0.84 to 90.74 ± 1.00), stem (50.19 ± 0.92 to 89.42 ± 1.10), and roots extracts (49.19 ± 0.52 to 90.01 ± 1.02) divulged magnificent antioxidant activities. For the ascertainment of the fatty acid constituents a gas chromatograph hyphenated to mass spectrometer was used. The essential fatty acids for growth maintenance such as linoleic acid (65.70%), eicosadienoic acid (15.12%), oleic acid (8.72%), and palmitic acid (8.14%) were found in high percentage.

Chaitanya MVNL *et al.*, (2015) investigated the microscopical, Phytochemical and *In-vitro* antioxidant studies of hydro alcohol, Aqueous, Ethyl acetate, Chloroform, Pet - ether and total saponin fractions of aerial parts of *Cestrum aurantiacum* and *Solanum mauritianum*. In this study, they founded that the selected plants posses' good amount of phenolics, alkaloids, flavanoids and saponins, among all the fractions the total saponin fractions showed better antioxidant activity in compare to other fractions ,The fractions also showed a good *In-vitro* cytotoxic activity on MCF-7 cell lines and moderate against HCT-116 cell lines in comparison to standard Salvicine.

Comparative morphological and stem anatomical studies were carried out in two varieties of *Tecoma stans* L. (Bignoniaceae) in Kerala. The species taxonomically complicated, earlier various taxonomist identified the species as *Tecoma stans* due to its morphological variability. Critical morphological study resulted determination two sub species of *Tecoma stans* var. *angustata* and *Tecoma stans* var. *stans*. Stem anatomical characters are also found important to distinguish the varieties within the species. Anish and Antony (2016).

Majgaine, Shweta and D.L Verma, 2017 studied that *Boerhaavia diffusa* have some pharmacological activities and used as a medicine with multi action such

andiuretic, entrasthamatic, diaphoretic, anthelmintic, antiasthamatic, diaphoretic antihepatotoxic antiurethritis febrifuge, antiscabies these pharmacological activities are due to the presence of chemical constituents.

Deepak Kumar *et al.,* (2018) studied anatomical and morphological features of different vegetative organs and reproductive organs were studied along with medicinal importance of the plant. *Portulaca oleracea*. In the morphological study it was observed that the plant have sessile leaves which were ovate, smooth, succulent, arranged in opposite manner, stem was aerial, weak and cylindrical, root consists of a long thick tap root as well as many fibrous lateral roots, flowers were single in leaf axils, fruit consists of round to egg-shaped capsules with glossy brown and black seeds. In anatomical studies, cross sections of the leaf, stem and root were examined. Purslane has better nutritional quality than the major cultivated vegetables with higher beta-carotene, ascorbic acid, and alpha-linolenic acid along with high nutritive and antioxidant properties.

Pachyptera DC. is a small genus of neotropical lianas included in tribe *Bignonieae* (Bignoniaceae). The genus has a complicated taxonomic history but currently includes species distributed from Belize to Southern Amazon. *Pachyptera* is characterised by four main synapomorphies, namely, a papery peeling bark, prophylls of the axillary buds organised in a series of three, patelliform glands arranged in lines in the upper portions of the calyx and corolla tube. Furthermore, members of the genus also have stems with four phloem wedges in cross-section and conspicuous extra floral nectaries between the interpetiolar region and at the petiole apex, although these characters are also shared with other genera of tribe *Bignonieae*. Here, we present a taxonomic revision of *Pachyptera*, which includes a complete list of synonyms, detailed morphological descriptions of species and an identification key, as

well as information on the habitat, distribution and phenology, nomenclatural notes, taxonomic comments and illustrations of all the species. In addition, we designate three lectotypes, propose one new combination, raise one variety to species status and describe a new species. After these adjustments, a *Pachyptera* with five well-defined species is recognised. Jessics and Lucia (2018).

Pranabesh Ghosh *et al*., 2019 studied five common medicinal herbaceous weeds of West Bengal, and India namely; *Heliotropium indicum*, *Tridax procumbens*, *Cleome rutidosperma*, *Commelina benghalensis*, and *Euphorbia hirta* have been chosen from five different families describe their phytochemical, and anti-oxidant properties. This investigations have concluded that *Euphorbia hirta* possesses a significant amount of phytochemicals, and it exhibits the highest anti-oxidant activities in comparison with the other four medicinal weeds. In *Euphorbia hirta* leaves acetone extract highest amount of phytochemicals were detected by qualitative assays.

Al-Newani, (2019) investigated the *Portulaca oleracea* is a succulent plant in Portulacaceae family distributed around different regions of Iraq as collected widely in the gardens of Baghdad governorate. The results of this study shown that a systematic significant of morphological and anatomical data, purslane with branched shoot stems. Stems and leaves are glabrous and leaves are alternate, the petiole is absent. Inflorescences were viewed as clustered in the form of small one carrying many male and female flowers as the inflorescences take the form of long stemmed. Anatomical techniques revealed two patterns of stomatal complex, paracytic which is the most common followed by tetracytic is limited distributed type and it is recorded for the first time in this species. Druses crystals have been found distributed in the stem with angular collenchyma alternating with xylem parenchyma cells with large

intercellular space. As well as, root anatomy has been done and the results showed casperian strips cells clearly in section with xylem and phloem region.

Khan (2020) investigated the Various morphological components of *Millingtonia hortensis* Linn. f. (cultivated in Dubai as road-side ornament) were studied for their surface micromorphology. The surface of this plant presented three important structures – much distributed trichomes on the surface of all organs of this plant, stomata present on leaf, pedicel, petals and fruit and tracheoidal system making the wing surface. The leaf was characterized with peculiar cuticular striation running parallel to each other. Stomata were anomocytic type, raised above epidermis and with well-defined rim around. Guard cells outer ledges were very prominent. Contiguous stomata were frequent. Occasionally, triplet stomata were also present. Seed wing was thin and papery and composed of tracheoids running all over the seed surface in a fan like manner. Seed wing was porous with air spaces of varying sizes, well-suited for dispersal of seeds.

Martinella Baill is a genus of Neotropical lianas in tribe Bignonieae (Bignoniaceae). The genus is monophyletic and well supported by morphological and molecular characters. Members of *Martinella* are characterized by a continuous interpetiolar ridge surrounding the stem, bilobed or 4–5-parted calyces, and minute triangular prophylls of the axillary buds. Generic circumscription remained unchanged since the description of the genus, although unclear species limits remained. Based on extensive fieldwork, herbarium work, and a molecular phylogenetic hypothesis for the genus, we here recognize five species of Martinella. Of these, three were recognized in earlier treatments for the genus, while two represent new species described here, *Martinella lanuginosa* Kataoka and L.G.Lohmann, sp. nov. and *Martinella tomentosa* Kataoka and L.G.Lohmann, sp.

nov. *Martinella iquitoensis* A. Samp. is treated as a synonym of *M. insculpta* Sprague & Sandwith. In addition, one second-step lectotype is designated for Bignonia *martini* DC, and neotypes are designated for *Doxantha longisiliqua* Miers and *Martinella gollmeri* K. Schum. This work provides a full taxonomic treatment for Martinella, including a complete list of synonyms, morphological descriptions, illustrations, photographs, distribution maps, conservation status, and comments for all five species recognized. Eric (2021).

Palonological investigation of 49 species of the family Bignoniaceae distributed over 32 genera has been made. There is a great diversity in pollen morphological characters. The pollen grains may be united in tetrad as in *Chilopsis saligna* or free as in rest of the species.

The present palynological investigation of Bignoniaceae in India highlights the presence of heteropolarity, multi-orate colpi, dimorphism & polymorphism among the members. These palynological characters are not common among the dicots and studies on them are quite meagre. So, the present study has been undertaken to explore more of them as they are significant in taxonomic deductions, in recognizing several taxons & also throws light on the evolutionary relationships. The freshly collected polleniferous material of 13 species of Bignoniaceae, belonging to 12 genera, from West Bengal were acetolysed & thoroughly investigated using bright field light microscopy and their respective photographs were taken. The surface pattern of the pollen grains is mostly reticulate & few are psilate. The apertural pattern ranges from inaperturate to tri-tetrazonocolpate to tri-zonocolpor (oid) ate to hexazonocolpate, thereby exhibiting a high degree of eurypalyny in the taxon. Multi-orate colpi are observed in *Fernandoa adenophylla, Jacaranda obtusifolia,*

Markhamia lutea, Parmentiera cereifera, Tabebuia heterophylla and *Tabebuia aurea*. Polymorphic pollen (*Parmentiera cereifera* & *Pyrostegia venusta*), dimorphic pollen (*Oroxylum indicum*) & heteropolarity (*Pyrostegia venusta*) has been observed in the family. Pollen grains of *Fernandoa adenophylla, Jacaranda obtusifolia, Mansoa alliaceae* and *Tabebuia* heterophylla has been investigated for the first time in the present study. Heteropolarity in *Pyrostegia venusta*, multi-orate colpate pollen grains in *Fernandoa adenophylla, Jacaranda obtusifolia, Markhamia lutea, Parmentiera cereifera, Tabebuia heterophylla, Tabebuia aurea* & polymorphism in *Parmentiera cereifera* & *Pyrostegia venusta* has been surfaced for the first time in the present study. Few suggestions are made regarding the significance and evolutionary trend of the multi-orate features of the pollen grains. The presence of both types of grain in *Parmentiera cereifera*, i.e. a) *Pancolpate apertural* pattern, lacking ora and b) *Pancolpate* with multi-orate features, indicates that the latter might have evolved from the previous one. Sadaquat and Tousif (2021).

However, high-throughput methods for assessing internal anatomy remain elusive, precluding the widespread inclusion of internal anatomy in many modern -omics- level studies (Yadav *et al*., 2021).

Morpho-Anatomical Traits and Soluble Sugar Concentration Largely Explain the Responses of Three Deciduous Tree Species to Progressive Water Stress. (Jonathan *et al*.,2021)

Morpho-Anatomical Feature and Phytochemical Assessments of Lasia spinosa (L)Thwaites. (Arya Lakshmi *et al*.,2021)

Morphological, anatomical and palynological studies of the genus *Dolichandrone* (Bignoniaceae) Thailand were conducted. Three species, *D. columnaris* Santisuk, *D. serrulata* (Wall. ex DC.) Seem., and *D. spathacea* (L. f.)

Seem. were investigated. Morphological descriptions, distributions and ecological information are provided. A key to the species based on morphological characters are leaflet margins, length of lower cylindric tube and upper campanulate tube of corolla, width of upper campanulate tube of corolla, winged seed, shape and characters of fruits, width of septum, characters and width of pseudoseptum. *D. columnaris* occurs in low-lying rice fields and marshlands only in the peninsular region. *D. serrulata* occurs in mixed deciduous forest and low-lying rice fields in the eastern, central and peninsular regions. *D. spathacea* occurs in edges of mangrove forest in the central, south-eastern and peninsular regions. A key to the species based on anatomical characters includes leaf type, number of rows of palisade cells, arrangement of axial parenchyma, and height of ray parenchyma. All pollen grains are similar and do not provide characters for identification within the genus *Dolichandrone*. Weereesa and Chatchai (2021).

Fridericia is a large genus of neotropical lianas with a complicated taxonomy. The genus is monophyletic and well supported by molecular characters, but lacks distinctive morphological synapomorphies, complicating its recognition. Species of *Fridericia* are distributed among six well-supported clades. As part of a series of taxonomic revisions of each clade of *Fridericia*, we present a taxonomic revision of the "Neomacfadya" clade. This group is broadly distributed through the Neotropics and includes species with glandular fruits and long-tubular calyces that are usually laterally split. We recognize 11 species for which we provide an identification key, typifications, morphological descriptions, illustrations, phenology, maps, comments on distribution and habitats, as well as suggest the conservation status for all species. We lectotypify six names (*Arrabidaea ateramnantha*, *A. craterophora* subvar. *glabrescens*, *A. craterophora* subvar. *velutina*, *Bignonia hispida*, *Scobinaria*

amethystina and *S. japurensis*). We correct the typification of *A. lenticellosa* Bureau & K. Schum. and *B. arthrerion*, provide a second-step lectotypification for *B. pearcei* and a neotypification for *Paragonia schumanniana*. We also correct information associated with the type of *A. oligantha* and synonymize this name in *F. japurensis*. We further reject *A. craterophora* var. *obtusifolia* and exclude one name previously treated in *Fridericia (Tecoma moritziana)*. Mirriam and Lucia (2022).

Floristic studies are critical for plant resource conservation and management. The purpose of this study was to investigate and document the members of the Tiliaceae family. During the flowering season, the specimens were collected. For morphological examinations, each species was viewed several times to check whether there were any differences in the specimen. In this study, two species from the Tiliaceae family are reported under one genus. Some of the species are native to the area, while others are cultivated. Updated nomenclature, short descriptions, phenology, range, local and common names, and medicinal uses are supplied for each species. (Beulah Jerlin and Abisha 2022)

Morphological, Phytochemical and FTIR spectroscopy analysis of portulacaceae members were studied. The result of preliminary phytochemical screening indicated that leaf and stem of both species of *Portulacaceae* were free from steroids. Moreover, quantitative estimation of phytochemicals also exhibited that leaf and stem of both species. Secondary metabolites, which are abundant in plants and have fascinating biological activities, are an important source with a variety of structural arrangements and properties. They have a rich source of protein and have high antioxidant (Beulah and Santhiya 2022).

Millingtonia hortensis L.f. of the family Bignoniaceae growing in Mawlamyine, Mon State. The collected specimens were measured and recorded for

taxonomic description. The microscopical studies of specimens were examined by preparing freehand sections and phytochemical test was also studied. The plant was tall tree, flower white. Moe *et al.,* (2020).

Yellow bells is a flowering perennial shrub which belongs to the family Bignoniaceae. It is a native of America, grown as ornamental shrub throughout India. It grows as a weed in India. From the ancient times the parts of the plant like leaves, seeds, pods, roots are used medicinally. Researches on *T.stans* gives the evidence that it contains chemical constituents like alkaloids, aminoacids, phytosterols, monoterpenes, triterpenes, glycosides, phenols, tannins, saponins, quines and flavonoids. However the researches also proves that it is used as anti-diabetic, anti-oxidants, anti- fungal, anti–microbial and antispasmodic. Sushma *et al.,* (2018).

The plant *Tecoma stans* (L.) Kunth, belongs to family Bignoniaceae and commonly known as "Pachagotla" is a dicotyledonous herb popularly grown for its flowers as an ornamental/garden plant in normal gardens and temples. It is also known as *Bignonia stans* L. Almost all parts of the plant is reported for its medicinal use. This plant is considered to be very effective in the treatment of diabetes. The leaves contain the alkaloids tecomine and tecostamine which is potent hypoglycaemic agent when given intravenously. Anthranilic acid is responsible for the antidiabetic activity, roots are powerful diuretic and vermifuge and tonic (Rao *et al.,* 2010).

Tecoma stans Linn is an erect shrub commonly found in India. It is also known as yellow bells, yellow elder, trumpet flower, belonging to the family Bignoniaceae. *Tecoma stans*, showed exhibited antidiabetic, antioxidant, hypoglycemic, antitumor, free radical, anti-inflammatory and antimicrobial, properties (Verma and Sunita 2016).

Tecoma stans (L.) Juss. ex H. B. K., a shrub or small tree in the Bignoniaceae, is a widely distributed polymorphic complex of the Western Hemisphere tropics and subtropics, and is commonly planted as an ornamental throughout the tropical world. Winter temperature limits its poleward distribution. The species is characteristic of rocky slopes, often limestone outcrops, but also alluvial and other substrata as long as drainage is excellent. In arid regions it occurs in climax xerophytic shrub or thorn forest communities, while in humid areas it is common mainly in deforested and other disturbed sites since it behaves as a heliophyte. The large yellow funnel-form flowers are pollinated by hummingbirds and perhaps also by some insects (Pelton 1964).

MATERIALS AND METHODS

(a) *Tecoma stans*

Common name: Trumpet bush, Esperanza (spanish for hope), Yellow elder, Yellow bells or yellow bignonia.

Class : Gamopetalae

Order : Lamiales

Family : Bignoniaceae

Genus : ***Tecoma***

Species : ***stans***

(b) *Tabebuia rosea*

Common name: Trumpet tree, Pink poui, New world trumpet, Rosy trumpet tree, Pink tecoma.

Class : Gamopetalae

Order : Lamiales

Family : Bignoniaceae

Genus : ***Tabebuia***

Spedies : ***rosea***

(c) *Millingtonia hortensis*

Common name : Tree jasmine, Indian cork tree.

Class : Gamopetalae

Order : Lamiales

Family : Bignoniaceae

Genus : ***Millingtonia***

Species : ***hortensis***

and often cultivated in garden. All these taxas are perennial in duration.

Method:

Collection and Identification of Plant Material

The fresh plant materials of *Tecoma stans*, *Tabebuia rosea* and *Millingtonia hortensis* were collected from the St. Mary's college campus, Thoothukudi. The collected plant sample were identified by using Flora of presidency of madras Gamble, (2004). The taxonomic identities of these plants were confirmed by using Flora of Tamil Nadu and Karnatic. Mathew (1983).

Macroscopic Characters

The macroscopical characters of stem, leaf, inflorescence and flowers of the selected taxa were examined by physical observation. Quantitative macro morphological characters viz. plant height, leaf length, leaf width, leaf area, petiole length, calyx length, corolla length, stamen length, pistil length and ovary size were measured. Length and width was measured with the help of scale. Ovary size was measured by the instrument camera lucida and the leaf area was measured with the help of leaf area meter.

Anatomical Identification

The cross section of the stem, petiole and leaf of the selected taxa were taken and stained with safranin and mounted in glycerin. Semi-permanent slides were prepared and observed under compound microscope. Photographs of the sections were taken under trinocular microscope in 45x magnification using T C Capture software.

Pattern and Distribution of Stomata

Pattern and distribution of stomata were studied by stomata peel method. The peels of the epidermis were stained with safranin and mounted in

glycerin. Semi-permanent slides were prepared and observed under compound microscope.

RESULT AND DISCUSSION

Macroscopical characters

The morphological characters of the studied taxa are presented in (Table 1 and Figure 1,2 & 3). *Tecoma stans,* was a perennial shrub, *Tabebuia rosea* and *Millingtonia hortensis* are a perennial tree. The arrangement of the leaf was opposite in all selected taxa. Variations in leaf shape, margin, base and apex of the three taxa are noted. In *Tecoma* leaf was odd pinnately compound, lanceolate with acuminate apex, broad base with serrate margin. In *Tabebuia rosea* leaves were digitately compound, oblong to elliptic, acute apex with smooth margin. In *Millingtonia hortensis* leaf was tripinnately compound, lanceolate with acuminate apex, crenate base with serrate margin. In the selected taxa, the inflorescence position, corolla colour and calyx shape had shown some variation. The corolla colour is yellow in *T. stans*, pink in *T. rosea* and white in *M. hortensis.* The quantitative macromorphological characters were measured and summarized in Table 3. Morphological characters were taxonomically significant for taxa identification and delimitation in Bignoniaceae.

Microscopic Observation:

Tecoma stans Stem T.S

T.S of the stem was slightly quadrangular in shape. The epidermis has a thick cuticle. The outer margin ruptured at places due to the presence of lenticels. The cortex is narrow, consists of 5-8 layered of collenchymatous and parenchymatous cells. The discontinues patches of sclrenchymatous cells beneath the cortex form a sort of pericycle. Endodermis is not clearly visible in the mature stem. Phloem cells are small, compressed and compactly arranged. Primary and secondary vascular elements are normal. The pith is parenchymatous and lack air spaces.

***Tecoma stans* Petiole T.S**

C.S of the petiole was circular with shallow adaxial concavity and two lateral short wings. The epidermis is thin and less conspicuous comprising of narrowly tubular cells. The outer zone of ground tissue is collenchymatous and four or five layered. The remaining part is parenchymatous, the cells are elliptical, compact and thick walled.

***Tecoma stans* Leaf T.S**

The vascular system is quite prominent and occupies entire midrib of the leaf. Adaxial epidermis is apostomatic the epidermal cells are polyhedral and randomly oriented. Their walls are slightly wavy and thick. In the surface view of the epidermis are seen shallow circular cavities in which occur the glandular trichome. The trichomes are randomly distributed in the veinlets. The abaxial epidermis is stomatiferous, the stomata are actinocytic type. Phloem elements are small numerous groups situated along the outer part of the xylem cylinder.

***Tabebuia rosea* Stem T.S**

T.S of the stem is almost circular in outline. Outer margin is ruptured at places due to the presence of lenticels. Cork is a thick walled lignified cells. Cells are squarish to rectangular in shape which are irregularly arranged. Cortex with the presence of well-developed porous tissue (red arrow) that was originated from the division of phellogen cells. Vascular bundles are conjoint, collateral and opened. Phloem cells are compressed. Xylem fibres occupies the major portion. Pith is a wider zone.

***Tabebuia rosea* Petiole T.S**

T.S of petiole is almost similar to the stem. It has a thick cuticle, epidermal cells were parenchymatous, rectangular in shaped compactly arranged one layered thick. 3-4 layered of parenchymatous cells lie beneath the epidermis. Vascular bundles were collateral type. Sclerenchyma patches were present in the cortex.

***Tabebuia rosea* Leaf T.S**

In T.S of leaf, the epidermis is one layered, parenchymatous cell rectangular in shaped and compactly arranged, cuticle thick on both surfaces. The palisade cells were 2 layered on upper surfaces. Vascular bundle were embedded in mesophyll. Parenchyma cells are present in the center of vascular bundle. Vascular bundle was crescent-shaped in structure. The leaves were amphistomatous with lesser number of stomata on dorsal surface but numerous stomata on the ventral surface. Stomata in varying number were observed on both surfaces.

***Millingtonia hortensis* Stem:**

The T.S of mature stem was circular in outline. The phellem 3-4 layered compactly rectangular in shaped. The phellogen 4-5 layered thin wall and rectangular in shaped. The phelloderm 4-5 layered of parenchymatous cells with rounded or polygonal in shaped. The cortex consisted of 10-12 layered. Pith is a wider zone.

***Millingtonia hortensis* Petiole:**

T.S of petiole has a thick cuticle, epidermal cells were parenchymatous, rectangular in shaped compactly arranged one layered thick. 3-4 layered of parenchymatous cells lie beneath the epidermis. Vascular bundles were collateral type. Sclerenchyma patches were present in the cortex. Crystals, oil drops and tannin were present in some parenchyma cells

***Millingtonia hortensis* Leaf:**

In T.S of leaf, the epidermis is one layered, parenchymatous cell rectangular in shaped and compactly arranged, cuticle thick on both surfaces. The palisade cells were 2 layered on upper surfaces. Glandular and non-glandular trichomes were present. The cells were vertically elongated and cylindrical. Spongy mesophyll cells were 5 to 7 layered and loosely arranged. Vascular bundle were embedded in mesophyll. Parenchyma cells are present in the center of vascular bundle. Vascular bundle was crescent-shaped in structure. The leaves were amphistomatous with lesser number of stomata on dorsal surface but numerous stomata on the ventral surface. Stomata in varying number were observed on both surfaces.

In this report, various morphological, microscopical, standards have been developed. These characters are in agreement with those given by Backer (1965), Dassanayake (1985) and Hooker (1885). So this study to serve as a tool for developing standards for identification, quality and purity of the plants.

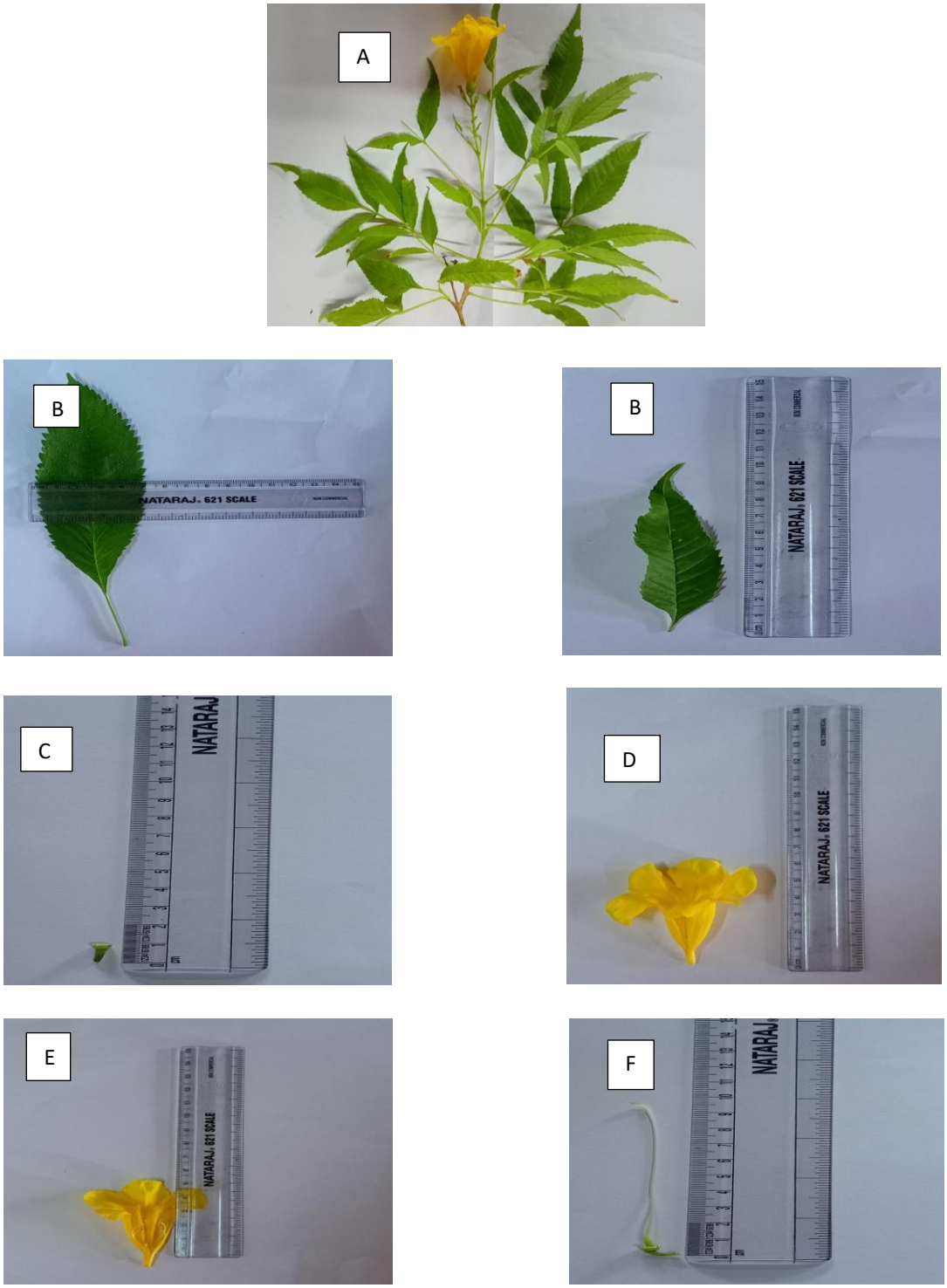


Figure : 1 Morphology of *Tecoma stans* A. A twig with flower, B. Leaf, C. Calyx, D. Corolla, E. Epipetalous stamen, F. Pistil

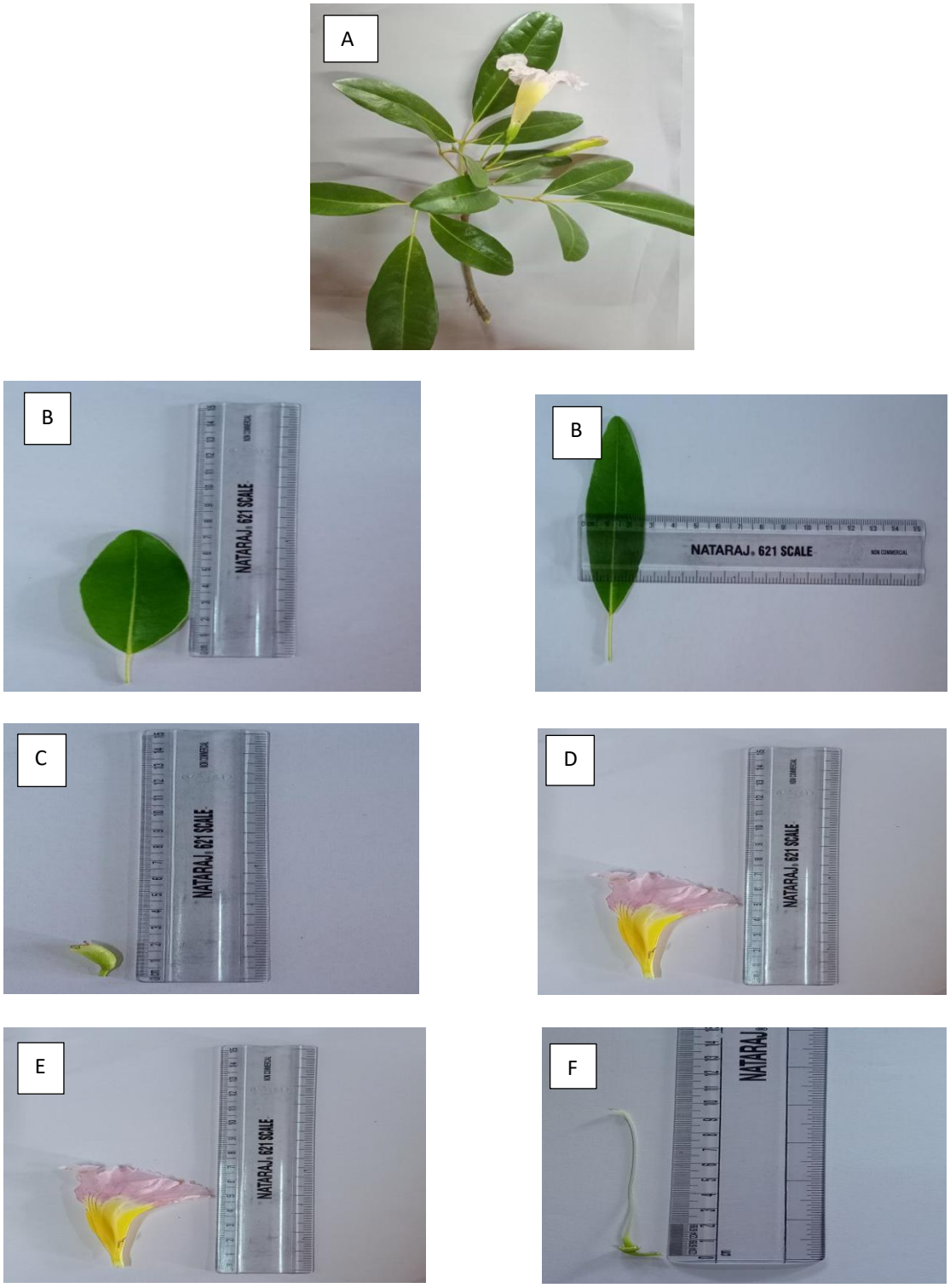


Figure: 2 Morphology of *Tabebuia rosea* A. A twig with flower, B. Leaf, C. Calyx, D. Corolla, E. Epipetalous stamen, F. Pistil

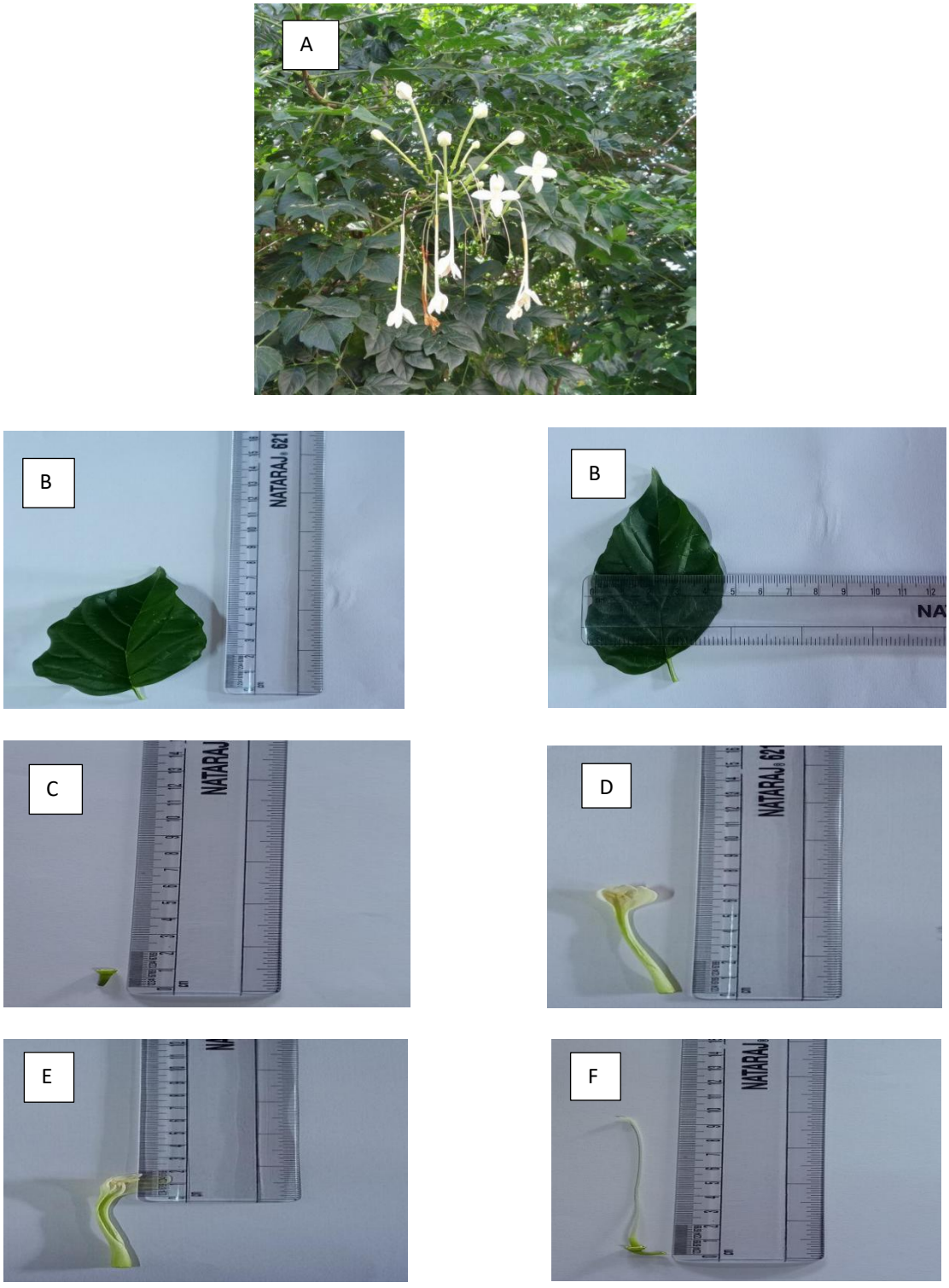


Figure: 3 Morphology of *Millingtonia hortensis* A. A twig with flower, B. Leaf, C. Calyx, D. Corolla, E. Epipetalous stamen, F. Pistil

Table 1: Morphological characters of selected species of Bignoniaceae

Characters		*Tecoma stans*	*Tabebuia rosea*	*Millingtonia hortensis*
Duration		Perennial tree	Perennial tree	Perennial tree
Stem	**Strength**	Woody	Woody	Woody
	Texture	Fissured	Fissured	Fissured
Leaf	**Arrangement**	Opposite	Opposite	Opposite
	Composition	Odd pinnately compound	Digitately compound	Tripinnately compound
	Shape	Lanceolate	Oblong to elliptic	Lanceolate
	Margin	Serrate	Smooth	Serrate
	Apex	Acuminate	Acute	Acuminate
	Base	Broad	Obtuse	Cuneate
Flower	**Inflorescence position**	Terminal panicles/raceme	Terminal panicles	Terminal corymbose panicles
	Bract shape	-	-	Present minute
	Calyx shape	Narrowly bell shape	Cupular shape	Cupular shape
	Corolla colour	Yellow	Lavender to pink	White
	Corolla shape	Bell shape	Funnel shape	Tube shape
	Stamen number	2+2 didynamous	2 + 2 didynamous	2+2 didynamous
	Ovary type	Superior ovary	Superior ovary	Superior ovary
	Locule number	2	2	Many
Fruit	**Type**	Capsule	Loculicide Capsule	Septifragal Capsule
Seed	**Nature**	Oblong seeds	Winged seeds	Discoid seeds

Table 2: Quantitative macromorphological measurements of studied taxa

Parts	Size		
	Tecoma stans	***Tabebuia rosea***	***Millingtonia hortensis***
Plant height	10 - 25 feet	20 - 25 feet	8 - 25 feet
Leaf length	14 ± 2 cm	7.5 ± 0.2 cm	5 ± 2.5 cm
Leaf width	5 ± 2 cm	2.6 ± 0.3 cm	4.5 ± 0.5 cm
Leaf Area	12.1 ±3.2 cm^2	23.5±2.0 cm^2	9.8 ± 3.9 cm^2
Petiole length	3.5 ± 0.2 cm	1.8 ± 0.1 cm	0.7 ± 0.2 cm
Calyx length	1.1 ± 0.2 cm	2.4 ± 0.2 cm	0.9 ± 0.2 cm
Corolla length	6..5 ± 0.5 cm	7.7 ± 0.5 cm	7 ± 3 cm
Stamen length	2.5 ± 0.8 cm	1.5 ± 0.5 cm	1.4 ± 0.3 cm
Pistil length	3.9 ± 0.1 cm	3.3 ± 0.1 cm	10 ± 0.3 cm
Ovary size	0.0455 cm^2	0.0665 cm^2	0.039 cm^2

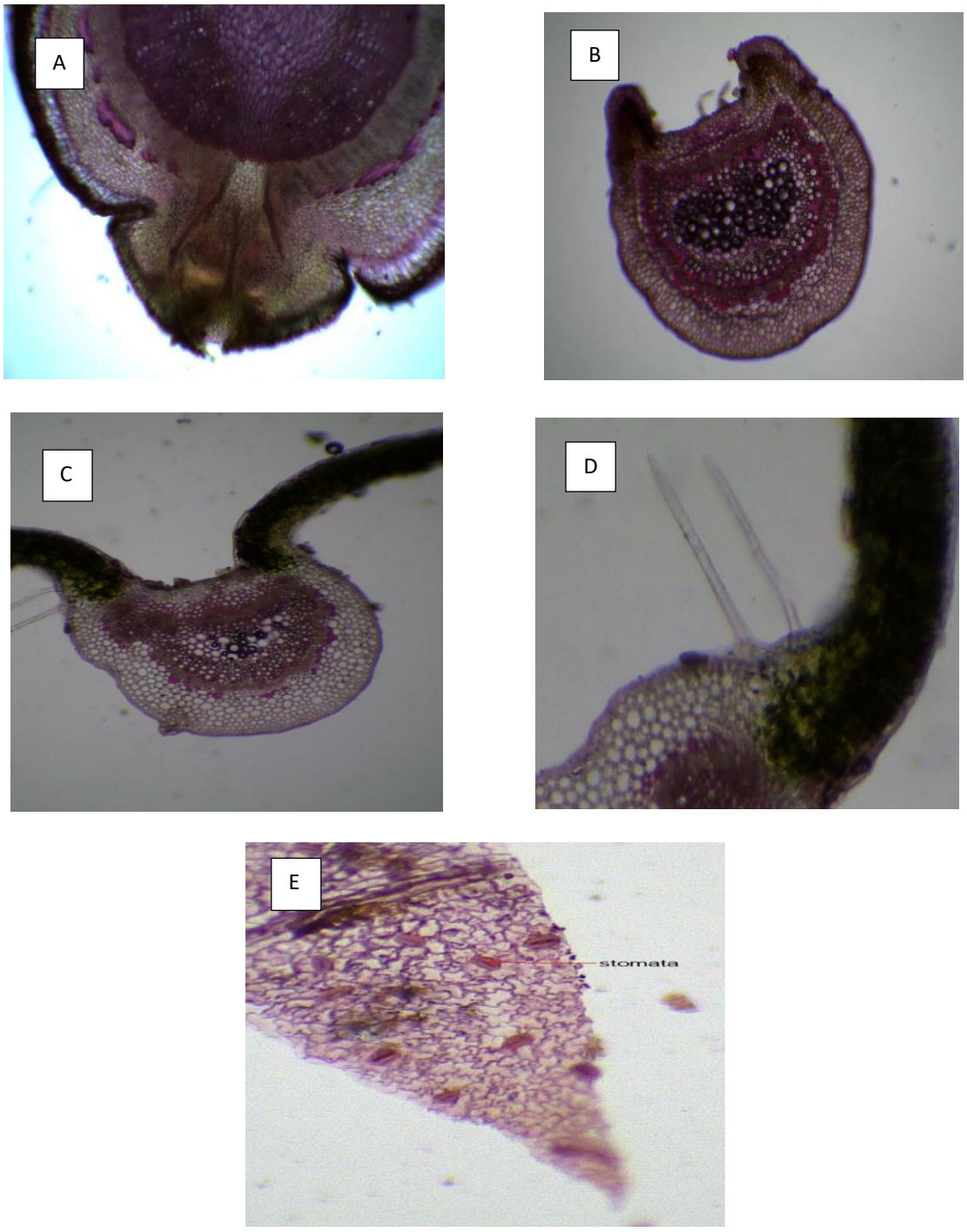


Figure : 4 Microscopic observation of *Tecoma stans* A. T.S of Stem, B. T.S of Petiole, C. L.S of Leaf, D. Trichome, E. Epidermal layer

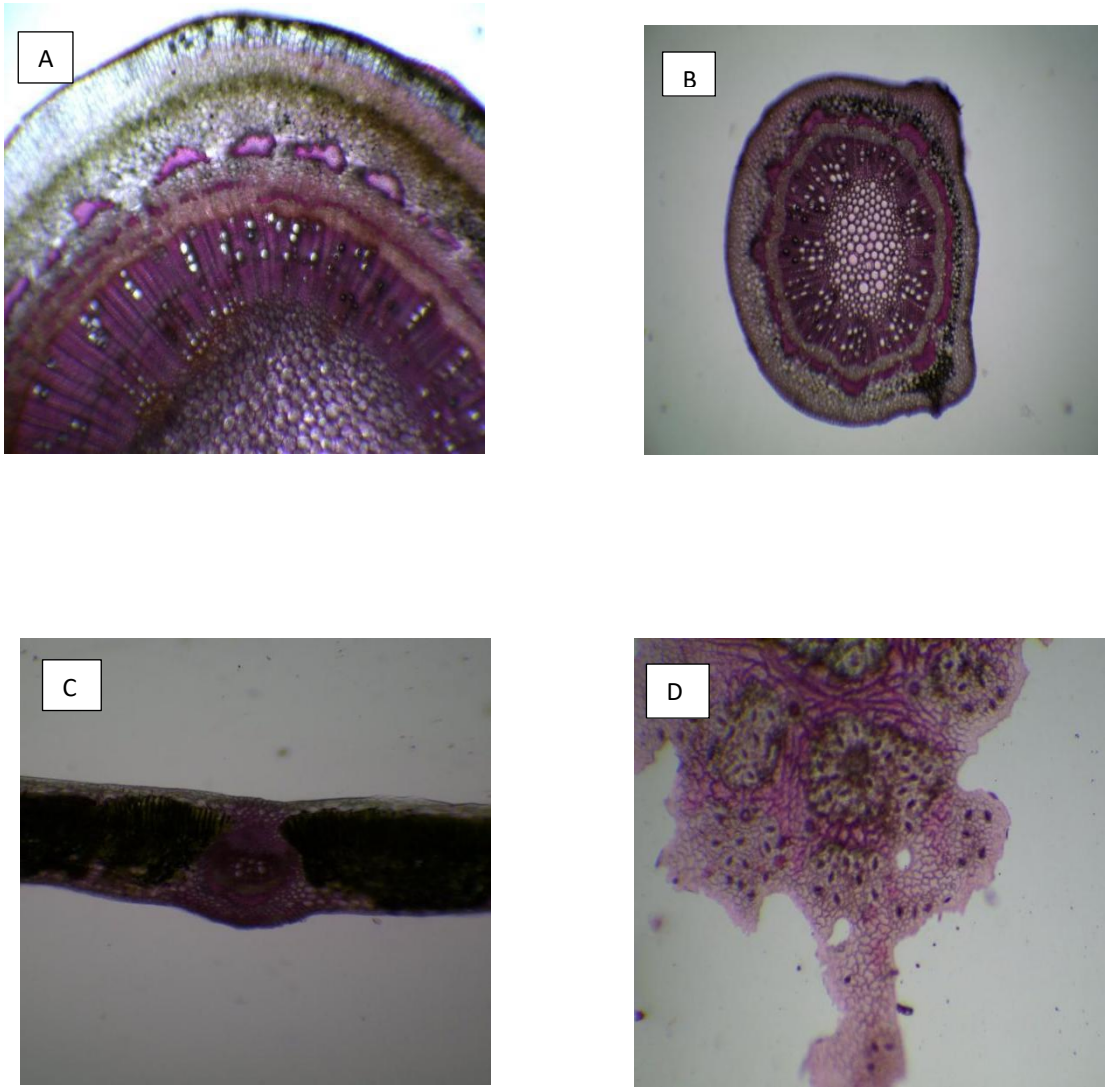
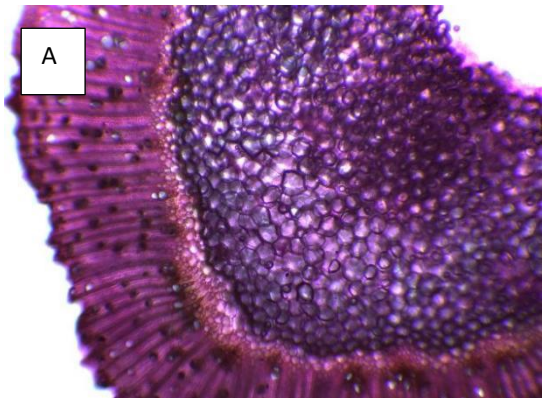
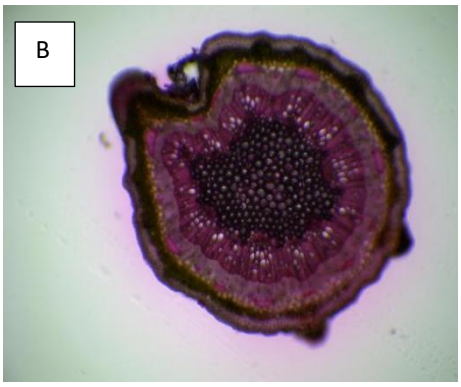
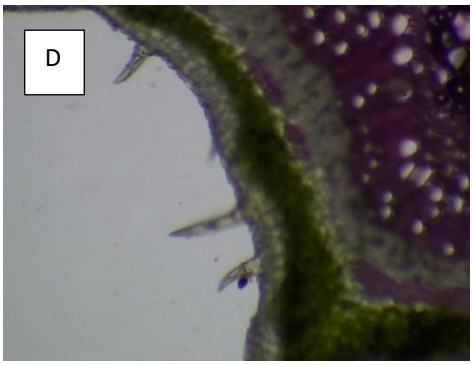


Figure : 5 Microscopic observation of *Tabebuia rosea* A. T.S of Stem, B. T.S of Petiole, C. L.S of Leaf, D. Epidermal layer







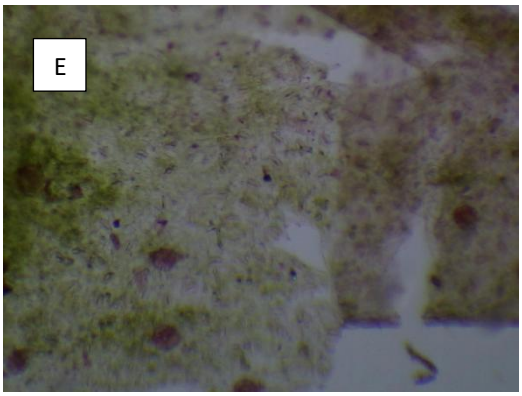


Figure : 6 Microscopic observation of *Millingtonia hortensis* A. T.S of Stem, B. T.S of Petiole, C. L.S of Leaf, D. Trichome, E. Epidermal layer

SUMMARY AND CONCLUSION

The Bignoniaceae family, often known as the Catalpa, Jacaranda, Trumpet Creeper, and Bignonia families, is primarily made up of trees and lianas with a few shrubs and herbs. It contains about 650 species and over 110 genera. Although there are plants in this family all over the planet, tropical and subtropical regions are home to the majority of them.

Bignoniaceae plants are significant and widely utilised by traditional healers in multiple countries, including Bangladesh, who treat a wide range of illnesses with diverse species due to their numerous pharmacological traits and recognised bioactive components.

For this study, the morphological traits of *Tecoma stans*, *Tabebuia rosea*, and *Millingtonia hortensis* were chosen. The stem, leaf and flower of *Tecoma stans, Tabebuia rosea* and *Millingtonia hortensis* were collected from St.Mary's College Campus, Thoothukudi for the current study.

The current investigation concluded that they are morphologically distinct. The size and shape of the leaf and stem also vary. Plant morphological and anatomical characteristics are crucial in identifying species. The findings suggest some pharmacognostic guidelines for future quality control of plant preparations. *Tecoma stans* and *Millingtonia hortensis* both had serrate leaf margins. In all species, the epidermis has anomocytic stomata on both surfaces. *Tecoma stans* and *Millingtonia hortensis* both have trichomes. Secondary growth occurred in all three taxa. Each of the three taxa is appealing and has healing properties. The various physical traits aid in species identification.

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PHYTOCHEMICAL, ANTIOXIDANT, FT-IR, GC-MS ANALYSIS AND ANTIBACTERIAL ACTIVITY OF SYZYGIUM CUMINI (L.) skeels.

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in partial fulfillment of the requirements for the Degree of

Master of Science in Botany.

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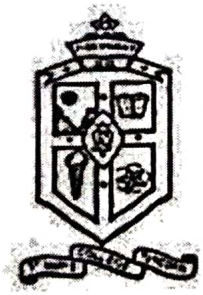

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2022-2023

CERTIFICATE

This is to certified that this dissertation entitled **'PHYTOCHEMICAL, ANTIOXIDANT, FT-IR, GC-MS ANALYSIS AND ANTIBACTERIAL ACTIVITY OF SYZYGIUM CUMINI (L.) skeels.'** Submitted by MARIA PREECIYA.S Reg.No.21APBO09 to St.Mary's College (Autonomous) Thoothukudi – 628001 in partial fulfillment for the award of the degree of **'Master of Science in Botany'** is done by her under my supervision. It is further certified that this dissertation or any pare of this has not been submitted else where for any other degree.

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DECLARATION

I do here by declare that this dissertation entitled **"PHYTOCHEMICAL, ANTIOXIDANT, FT-IR, GC-MS ANALYSIS AND ANTIBACTERIAL ACTIVITY OF SYZYGIUM CUMINI (L.) skeels."** Submitted by me in partial fulfillment for the award of the degree of **'Master of Science in Botany'**,in the result of my original and independent work carried out under theguidance of **Dr. Mrs. B.Maria Sumathi M.Sc, M.Phil., Ph.D.** Assistant Professor. Department of Botany, St. Mary's College (Autonomous) Thoothukudi and it has not been submitted else where for the award of any other degree.

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CONTENTS

INTRODUCTION

India is a tropical country with a rich diversity of medicinal plant resources. Medicinal plants play an important role in supporting health care in India. According to World Health Organisation (WHO) (2002), 80% of the rural population in developing countries rely on locally available medicinal plants for their primary health care. Medicinal plants are important therapeutic aid for the alleviation of ailments of humankind. These plants are used in herbal medicine, cosmetology and nutraceuticals. People are becoming more aware of medicinal plants and many of them utilize these therapeutic interventions and their products for maintaining health and for preventing diseases with an eco-friendly touch. Medicinal plants are potentially renewable. For the conservation and sustainable use of medicinal plants, a long-term integrated, scientific plan needs to be adopted.

Plant derived medicines are believed to be risk-free, milder and superior to chemically synthesized drugs for human health. The human body recognizes components that occur in plants and has a sophisticated mechanism for metabolizing such plant materials. The bioactive compounds naturally available in plants may have lower potency than allopathic (synthesized) drugs. However, as they are traditionally consumed in significant amounts through diet, they may provide long-term physiological benefits without any detrimental side effects (Espin et al., 2007). Consumption of foods rich in phytochemical and other bioactive food components have been clearly linked to the prevention and reduction of cancer (Steinmetz and Potter, 1991), cardiovascular diseases (Duthie and Brown, 1994) and to improvement in the immune system (German and Dillard, 1998)

Plant materials remain an important resource to combat serious diseases in the world. Traditional medicinal methods, especially the use of medicinal plants still play a vital role to

cover the basic health needs in developing countries. The medicinal value of these plants lies in some chemically active substances that produce a definite physiological action on the human body. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids and phenolic compounds. GC-MS and FT-IR have played an important role in pharmaceutical analysis in recent years.

Many infective diseases are treated with a chemotherapeutic agent, such as antibiotics, that selectively inhibit or kill the pathogen with little or no effect on the host. Ideally, antimicrobial agents disrupt microbial processes or structures that differ from those of the host. They may damage pathogens by hampering cell wall synthesis, inhibiting microbial protein and nucleic acid synthesis, disrupting microbial membrane structure and function or blocking metabolic pathways through the inhibition of key enzymes (Prescott *et al.*, 2008). Conventional antimicrobial agents face a lot of resistance problems in recent times as microorganisms are losing sensitivity to some of these drugs. Recently, concern has been expressed about the rising prevalence of pathogenic microorganisms which are resistant to the old generation, and to the newer or modern antibiotics that have been produced in the last decades (Cohen, 1992; Nascimento *et al.*, 2000; Okesola and Makanjuola, 2009). Also, the problem posed by the high cost; adulteration and increasing toxic side effects of these synthetic drugs coupled with their relative inadequacies in disease treatment, especially in developing countries portend serious limitations (Shariff, 2001). Consequently, the continuous acute need for novel and effective antibiotics for antimicrobial chemotherapy is clearly evident. Phytochemicals derived from plants have shown great promise in the treatment of intractable infectious diseases (Nascimento *et al.*, 2000; Rios and Recio, 2005) with lesser side effects compared to the synthetic drug agent (Iwu *et al.*, 1999).

In traditional herbal practice, indigenous medicinal plants have been employed in the treatment of several important infections (Fennell *et al.*, 2004, Taylor *et al.*, 2001). Also,

plant-based extractives have equally served as a source of lead compounds for the further development of future antimicrobial agents. Therefore, the evaluation of a candidate medicinal plant may lead to the identification of very effective herbal antimicrobial treatments or provide leads for further development into novel antimicrobial agents.

Phytochemicals are known to work as immune modulators and may have anti-inflammatory, anticancer and antimicrobial activities. All these properties of the phytochemicals are attributed to its effective antioxidant mechanisms against the endogenously produced harmful free radicals. Our body has effective antioxidant defense systems, which constitute enzymes, such as superoxide dismutase (SOD), catalyze and compounds, such as ascorbic acid, tocopherol, and glutathione. But all these endogenous antioxidants are not sufficient in protecting the body against oxidative stress. Therefore, dietary supplementation through natural antioxidants in place of synthetic antioxidants is necessary for strengthening the antioxidant system of the body by inhibiting free radical generation and thus preventing chronic diseases. Recently, much attention has been directed towards exploring natural antioxidants because they are natural products that are considered to be safe sources (Abdul Sadat *et al.*, 2017).

The Myrtaceae family includes around 150 genera and 3600 species with a cosmopolitan distribution. The genus *Syzygium* is one of the genera of the myrtle family Myrtaceae which is native to the tropics, particularly to tropical America and Australia. It has a worldwide, although highly uneven, distribution in tropical and subtropical regions. The genus comprises about 1 100 species, and has a native range that extends from Africa and Madagascar through southern Asia east through the Pacific (Ayyanar and Pandurangan Subash-Babu, 2012). For the present study, the taxa - *Syzygium cumini* (L.) Skeels. was selected.

Syzygium cumini (L.) Skeels. is one of the best known species. *Syzygium cumini* or *Eugenia jambolana*(Myrtaceae), known as 'Naaval' in Tamil and Jamun, Jambul and Jambool in India and Malaya. For a long period of recorded history, the tree is known to have grown in the Indian sub-continent, and many others adjoin regions of South Asia such as India, Bangladesh, Burma, Nepal, Pakistan, Sri Lanka and Indonesia. It was long ago introduced into and became naturalized in Malaysia. In southern Asia, the tree is venerated by Buddhists, and it is commonly planted near Hindu temples because it is considered sacred to Lord Krishna (Morton, 1987)

All parts of the *Syzygium cumini* can be used medicinally and it has a long tradition in alternative medicine. From all over the world, the fruits have been used for a wide variety of ailments, including cough, diabetes, dysentery, inflammation and ringworm (Reynertson *et al* 2005). It is also an ancient medicinal plant with an illustrious medical history and has been the subject of classical reviews for over 100 years. It is widely distributed throughout India and ayurvedic medicine (Indian folk medicine) mentions its use for the treatment of diabetes mellitus. Various traditional practitioners in India use the different parts of the plant in the treatment of diabetes, blisters in the mouth, cancer, colic, diarrhea, digestive complaints, dysentery, piles, pimples and stomachache (Jain 1991,). In Unani medicine, various parts of jambolan act as a liver tonic, enrich the blood, strengthen teeth and gums and form a good lotion for removing ringworm infection of the head (Sagrawat *et al* 2006).

The current study of phytochemical analysis, antioxidants and antimicrobial analysis will contribute to the knowledge of the medicinal properties of selected plants and it will create a path to travel for the researchers. Through this research, I could share the therapeutic uses of these plants to my society.

SCOPE AND OBJECTIVE

SCOPE AND OBJECTIVES

The aspiration of the current study was to assess the biochemistry and bioactivities of the plant extract (*Syzygium cumini*). In this work, the following objectives are focused.

i. Collection of the leaf and seed of *Syzygium cumini* plants for extract preparation.

ii. To qualitatively screen the presence of phytochemicals by using different solvents (acetone, methanol, ethanol,) and aqueous extracts of leaf and seed of *Syzygium cumini* .

iii. To quantitatively analyze and compare the total phenols, flavonoids, vitamin C and Tannin of the leaf and seed of *Syzigium cumini.*

iv. To identify and compare the functional group of leaf and seed of *Syzigium cumini* by Fourier transform infrared spectroscopy (FTIR) analysis.

v. To identify the bioactive compounds of the leaf and seed extract of *Syzigium cumini* using GC-MS analysis.

vi. To assess the antioxidant potential of leaf and seed aqueous extract of *Syzigium cumini* against DDPH radical scavenging activity.

vii. To evaluate the anti-bacterial potential of acetone, methanol, ethanol and aqueous extracts of leaf and seed of *Syzigium cumini*

REVIEW OF LITERATURE

Phytochemicals are chemical compounds formed during the plants normal metabolic processes. These chemicals are often referred to as "Secondary metabolites" of which there are several classes including alkaloids, flavonoids, coumarins, glycosides, polysaccharides, phenols, tannins, terpenes and terpenoids (Okwu, 2004). In addition to these substances, plants contain other chemical compounds. These can act as agents to prevent unconsiderable side effects of the main active substances or to assist in the assimilation of the main substances. Many herbaceous and medicinal plants contain important photochemical and vitamins such as alkaloids, flavonoids, tannins, cyanogenic glycosides, phenolic compounds, saponins, lignins, vitamin C, vitamin E and carotenoids, which are utilized both by humans and animals as important components of diets (Hussain *et al.,* 2011). The medicinal effects of plants are considered to be due to metabolites, especially secondary compounds, produced by plant species. Phytochemical analysis suggests that the presence of various biologically active compounds [alkaloids, phenols, flavanoids, proteins-lectin, carbohydrates, indigo, steroids etc.] and could be correlated to various therapeutic purposes (Vinoth *et al.,* 2011). Plants have an almost limitless ability to synthesize aromatic substances, mainly secondary metabolites of which 12,000 have been isolated, a number estimated to be less than 10% of the total (Mallikharjuna *et al.,* 2007).

The phytochemical screening of different parts of the *Jatropha curcus* revealed the presence of tannins, saponins, carbohydrates, sterols, diterpenes, alkaloids, flavanoids and various enzymes. Root contains di-terpenoid, Jatrophol and Jatropholones A and B, taraxerol b-sito-sterol. The bark contains tannins, resins, saponins, reducing sugar and traces of a volatile oil. Leaves contain Steroid, alkaloids triterpene (Rajore and Batra, 2004). Musa *et al.*,

(2000) studied the phytochemistry of powdered leaves of *Acalypha recemosa* (Euphorbiaceae). This study revealed the presence of alkaloid, tannin, flavanoid and terpenes.

Sivaraj *et al.*, (2011) conducted preliminary phytochemical screening using five different solvents extracts of *Aegle marmelos, Ruta graveolens, Opuntia dillenii, Euphorbia royleana*and *Euphorbia antiquorum*. Phytochemical profiling of *Mimosa pudica*was carried out by Sriram *et al.*, (2011). Sukumaran *et al.*, (2011) identified the phytochemical constituents of methanol extract of flower of *Peltophorumpterocarpum*. Phytoconstituents found in *Tridax procumbens* were isolated and characterized by Surendra and Talele (2011).

Nwokocha *et al.,* (2011) studied the comparative phytochemical screening of *Jatropha curcus*, *Jatrophagossypifolia, Jatropha multifida* and *Jatropha podagrica* on leaf, stem root and seeds and the results revealed that tannins were found to be the most abundant followed by saponins and flavanoids and phenols. Vindhya K *et al.*, (2014) conducted the preliminary phytochemical study in *Gardenia latifolia* and *Gardenia gummifera*, using different solvents. The petroleum ether extract of both the plants were found to contain glycosides, phytosterols, fats and oils, resins, phenols and triterpenes. Flavonoid was found to be present in *Gardenia latifolia* and not in *Gardenia gummifera*. Alkoloids, carbohydrates, saponins, tannins, proteins, amino acids and diterpenes were absent in both the plants. Ethyl acetate extracts of the plant was found to contain glycosides, phytosterols, resins, phenols, flavonoids and triterpenes. Alkoloids, carbohydrates, saponins, fats, oils, tannins, proteins, amino acids and diterpenes were absent in both the plants.

Ved Prakash *et al,* (2015) investigated phytochemical screening and antioxidant activity of *Adina cordifolia* leaf. The plant extracts were screened for presence of flavonoids, carbohydrate, alkaloid, saponin, phenol, tannins, phlobatannins, terpenoids, and cardiac glycosides. Total flavonoid content, phenols content was estimated. Antioxidant activity was

determined using nitric oxide scavenging assay, DPPH assay, hydrogen peroxide scavenging and ferric reducing methods, also MIC was calculated against a set of bacteria (*S. aureus, B. subtilis, E. coli, V. cholarae*). Ravindranath (2003) has been isolated a novel macrocyclic diterpene–Jatrophenone from the whole plant of *Jatropha gossypifolia*. This compound possesses significant antibacterial activity.

FTIR

A large number of medicinal plants are used as alternate medicine for diseases of man and other animal since most of them are without side effects when compared with synthetic drugs. Identification of the chemical nature of phytochemical compounds present in the medicinal plant will provide some information on the different functional groups responsible for their medicinal properties. Iqbal Ahamed *et al.*, (2006) detected major groups of compounds as the most active fraction of four plants extract by infrared spectroscopy.

Ramamoorthi and Kannan (2007) screened the bioactive group of chemicals in the dry leaf powder of *Calotropis gigantea* by FTIR analysis Kareru *et al.*, (2008) detected saponins in crude dry powder of 11 plants using FTIR spectroscopy.

Muruganantham *et al.*, (2009) carried out the FTIR spectroscopic analysis in the powder samples of leaf, stem and root of *Eclipta alba* and *Ecliptaprostratea*. The FTIR analysis of aqueous methanolic leaf extracts of *Bauhinia racemosa* for phytochemical compounds was done by Gauravkumar *et al.*, (2010). Ragavendran *et al.*, (2011) detected the functional groups in various extracts of *Aervalanata* using spectroscopic method .

Thangarajan Starlin *et al.*, (2012), analyzed the ethanolic extracts of *Ichnocarpus frutescens*, by FTIR, revealed the presence of functional group components of amino acids, amides, amines, carboxylic acid, carbonyl compounds, organic hydrocarbons and halogens. Parag A. Petnekar and Bhanu Raman (2013) carried out the FTIR

spectroscopic analysis of methanolic leaf extract of *Ampelo cissuslantifolia* for antimicrobial compounds.

FTIR analysis for five selected green leafy vegetables (GLVs) viz., *Hibiscus cannabinus* , *H. sabdariffa* , *Basella alba* , *B. rubra* L. and *Rumex vesicarius* confirmed the presence of free alcohol, intermolecular bonded alcohol, intramolecular bonded alcohol, alkane, aromatic compounds, imine or oxime or ketone or alkene, phenol and amine stretching (Sravan Kumar and Manoj., 2015).

The functional group identification is made by FTIR analysis and the active components based on the peak value in the region of infrared radiation. The ethanolic flower extract of *Erythrina variegata* L. is passed into the FTIR spectroscopy and the functional groups of the components are separated based on the peak ratio. The results of FTIR analysis confirm the presence of functional groups such as non-bonded, O-H stretch, carboxylic group, acidic, H bonded, C-H stretch, asymmetric stretching of –CH (CH2) vibration, C=N (stretch), carbon-carbon triple bond, multiple bonding, carbonyl compound frequency, C=O stretch, C=C stretch, O-H bend, alcoholic group, C-N stretch, C-O stretch, PO3 stretch, =C-H bending and C-Cl (Priyanga S *et al.,* (2017).

GC-MS

The chemical composition of the essential oils from leaves and wood of *Ocotea brenesii* growing wild in Costa Rica was determined by capillary GC/FID and GC-MS. From the leaves, 64 compounds were identified, corresponding to 85.9% of the oil, and from the wood 57 compounds were identified corresponding to 69.0% of the oil (Carlos and Jose, 2005). The chemical compositions of the essential oils of *Ocimum basilicum* L. *cv.* purple and *Ocimum basilicum* L. *cv.* green cultivated in Iran were investigated by GC-MS (Seyed, 2006).

GC-MS analysis of *Jatropha curcas* leaves revealed the presence of 16 compounds. The most abundant components were 22, 23-dihydro-stigmasterol (16.14%) alpha-tocopherol (15.18%), beta amylin (7.73%) and dotriacontanol (7.02%) The content of gamma tocopherol reached 2.88% and Vitamin E reached 18.06% in the extract (Wang *et al.*, 2009). The GC-MS analysis of *Strobilanthes crispus* oil revealed the presence of 28 components. The main constituents were found to be phytol, α-cadinol, Megastigmatrienone, 2,3-dihydrobenzofuranand eugenol (Asmah*et et al.*, 2006).

Nithya Narayanaswamy and Balakrishnan (2011) evaluated the antioxidant properties of 13 important medicinal plants and it showed that *Ocimum basilicum* leaf, *Alpinia calcarata* leaf, *Jatropha mulitifida* flower, *Hyptissua veolens* leaf, *Solanum indicum* leaf and *Clitoria ternatea* leaf and flower possessed higher DPPH scavenging activity. Moussa *et al.*, (2011). The aqueous leaf extracts of 124 Egyptian plant species belonging to 56 families were investigated and compared for their antioxidant activity by DPPH scavenging assay. Safi *et al.* (2012) studied the biological activities of aqueous extract of the root of *Jatropha curcas* like antimicrobial and free radical scavenging activities. In the evaluation of DPPH free radical scavenging activity. Olabinri *et al.,* (2013) investigated *in vitro* antioxidant and nitric oxide radical scavenging capabilities of *Jatropha gossypifolia* extract.

Sermakkani M. And V. Thangapandian (2012) evaluated GC-MS analysis of *C. italica* leaves revealed the presence of seventeen compounds. The identified compounds possess many biological properties. For instance, 9,12,15-Octadecatrienoic acid, (Z,Z,Z)-Linolenic acid (R/T 20.06) possesses anti-inflammatory, insectifuge, hypocholesterolemic, cancer preventive, nematicide, hepatoprotective, antihistaminic, antieczemic, antiacne, 5-alpha reductase inhibitor, antiandrogenic, antiarthritic and anticoronary properties. n-

Hexadecanoic acid - palmitic acid (R/T 17.25) can be an antioxidant, hypocholesterolemic, nematicide, pesticide, lubricant activities.

Fenghuan Wei *et al.*, (2015) identified thirty compounds in *Jasminum grandiflorum* by using GCMS. The major volatile components of the flower were 3,7,11,15- tetramethyl-2-hexadecen-1-o (phytol) (25.77 %), 3,7,11- trimethyldodeca -1,6,10-trien-3-ol (12.54 %) and 3,7,11,15- tetramethyl -1-Hexadecen-3-ol (12.42 %). The results show that phytol is the major volatile component of *Jasminum grandiflorum.*

Praveen Kumar P *et al.,* (2018) studied the identification of bioactive compounds from the Neem sap by Gas chromatography and Mass spectroscopy (GC-MS). The GC-MS analysis of the Neem sap revealed the presence of 30 volatile compounds. Among the 30 compounds, the most predominant compounds are fatty acids like Hexadecanoic acid and Pentadecanoic acid. Hence, this current attempt forms a basis for the biological characterization and importance of the compounds which could be exploited for future development of drugs.

Seventy six kinds of chemical compounds were found in methanol extract of *E.cephalotes* including aldehydes (7.9%), phenols (7.5%), fatty acids (5.8%) and furfural (5.4%) and 86 kinds of chemical compounds found in *M.anisodan* extract. Furfural, steroids, vitamin B and flavonoids are the main compounds of *M.anisodan* by S. Mohammadi*et al.*, (2019).

Antioxidant activity

Antioxidant compounds in food play an important role as a health protecting factor. Primary sources of naturally occurring antioxidants are whole grains, fruits and vegetables. Natural antioxidants can also be replaced by commercially available, synthetic antioxidants such as butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT), which are quite unsafe to use and is restricted due to their carcinogenic effect (Velioglu*et al.*, 1998).

Natural antioxidants or phytochemical antioxidants are the secondary metabolites of plants (Walton and Brown, 1999). Carotenoids, flavonoids, cinnamic acids, folic acid, ascorbic acid, tocopherols, tocotrienols *etc.*, are some of the antioxidants produced by this plant for their sustenance. Beta-carotene, ascorbic acid and alpha tocopherol are the widely used as antioxidants (McCall and Frei, 1999).

Flavonoids are polyphenolic compounds, which are ingredients of many vegetables and fruits. They are classified into flavanols, flavanones, flavones, iso-flavones, catechins, anthocyanins, proanthocyanidins, etc. (Huy*et et al.*, 2008). They are among the most bioactive plant secondary metabolites which outperform well-known antioxidants.

Natural antioxidants are known to exhibit a wide range of biological effects including antibacterial, antiviral, anti-inflammatory, anti-allergic, anti-thrombic and vasodilatory activities. Antioxidant activity gives rise to anti-carcinogenicity, anti- immunogenicity and anti-aging activity (Gulcin*et al.*, 2010).

Flavonoids serve as ROS scavengers by locating and neutralizing radicals (Gill and Tuteja, 2010). Bioactive properties such as free radical scavenging, inhibition of hydrolytic and oxidative enzymes and anti-inflammatory action of flavonoids is known (Njoku *et al.*, 2011). The antioxidant activity of the dietary phenolics considered to be superior to that of the essential vitamins and is ascribed to their high redox potential, which allows them to interrupt free radical mediated reactions by donating hydrogen from the phenolic hydroxyl groups (Beevi*et al.*, 2010).

Phenolics are secondary metabolities that behave as antioxidants due to the reactivity of the phenol moiety (hydroxyl substituent on the aromatic ring). The antioxidant activities of phenolic compounds are also attributed to their ability to chelate transition metal ions, such as those of iron and copper, which have been proposed as the catalyst for the initial formation of ROS (Knezevic*et al.*, 2011).

Ascorbic acid (vitamin C) is a vital component in human diet with the highest concentrations in animal organs like the liver, leukocytes, and anterior pituitary. It is used for its antioxidant effect (Ensafi*et al.*, 2010). Vitamin C is a major ubiquitous non-enzymatic, water soluble antioxidant (Ueta*et al.*, 2003). It acts as ROS scavenger, thus potentially protecting cells from harmful oxidative products (Fossati *et al.*, 2011). Vitamin C functions in enzyme activation, oxidative stress reduction, and immune function. There is considerable evidence that vitamin C protects against respiratory tract infections and reduces risk for cardiovascular disease and some cancers (Schlueter and Johnston, 2011).

Tannins are group of polymeric phenolic substances. Consumption of tannin containing beverages, especially green teas and red wines can cure or prevent a variety of illness including heart related diseases (Van-Burden and Robinson, 1981).

Swamy *et al.*, (2004) tested the leaf extracts of medicinal plant, *Leptadenia reticulata* for AgNPs production and antioxidant activity studies. He observed that, 500 µg/ml of green synthesized silver nanoparticles showed maximum (64.81 %) radical scavenging activity. The silver nanoparticles were synthesized using aqueous *Piper longum* fruit extract and the aqueous *P. longum* fruit extract and the green synthesized silver nanoparticles showed powerful antioxidant properties *in vitro* antioxidant assays. Haes *et al.*, (2002).

Pourmorad *et al.*, (2006) carried out a comparative study on the antioxidant potentials of some selected Iranian medicinal plant extracts. The antioxidant properties of 25 edible tropical plants were studied by Wong *et al.*, (2006). Badami and Channabasavaraj (2007) studied the *in vitro* antioxidant activities of thirteen medicinal plants collected from Western Ghats of India.

Ademiluyi and Oboh (2008) studied the antioxidant activity of methanol leaf extract of *Viscum album* by using linolenic acid peroxidation and DPPH methods. Effat *et al.*,(2008) screened thirteen medicinal plant extracts for antioxidant activity.

MoniRani *et al.*,(2008) evaluated antioxidant activities of methanol extract of *Ixora coccinea* by DPPH free radical scavenging activity, reducing power and total antioxidant activity assays.

Gayatri *et al.*, (2011) observed that the piperine, an alkaloid found naturally in *Piper nigrum* and *Piper cubeba*. It is widely used in various herbal cough syrups and anti-inflammatory, antimalarial, anti-leukemiatreatement. Ethanol extract of *Piper cubeba* showed high antioxidant activity.

Inbathamizh *et al.,* (2013) studied in vitro evaluation of antioxidant and anticancer potential of *Morindapubescens* synthesized silver nanoparticles. The decolorization from purple DPPH radical to yellow DPPH molecule by the sample in a dose-dependent manner with an IC50 value of 84±0.25 µg/ml indicated the sample's high radical scavenging activity, which was closer to that of the standard whose IC50 value was found to be 80±0.69 µg/ml.

Niraimathi *et al.,* (2013) investigated on biosynthesis of silver nanoparticles using *Alternanthera sessilis* (Linn.) leaf extract and determined antioxidant activities. Free radical scavenging activity of the AgNPs on DPPH radical was found to increase with increase in concentration, showing a maximum of 62% at 500 µg/ml. The standard gallic acid, however, at this concentration exhibited 80% inhibition. The IC50 value was found to be 300.6 µg/ml.

The silver nitrate extract of *Annona squamosa* and *Sapium macrocarpum* showed two times more DPPH scavenging activity than the commercial antioxidant butylated hydroxyl anisole. (Ruiz *et al.*, 2008). The silver nitrate extracts of *Melissa officinalis*, *Matricaria recuttia* and *Cymbopogan citrates* were found to possess DPPH scavenging activity. (Pereira *et al.*, (2009). Sowndharajan *et al.*, (2010) studied the antioxidant capacity and total phenolic contents present in the silver nitrate extracts of leaves, stem, and roots of *Melothria maderaspatana* were evaluated. Sathisha *et al.*, (2011) determined antioxidant potentials in

silver nitrate extract of some plants, *Curcuma longa, Coffea Arabica, Tribulus terrestris, Bacopa monnieri* and *Trigonella foenumgraceum* using various *in vitro* assays.

Iwalewa *et al.*, (2005) studied the pro and antioxidant effects of silver nitrate extracts of nine edible vegetables in southwest Nigeria using 1, 1-diphenyl-2-picrylhydrazyl free radical assay. The silver nitrate extract of *Helichrysum plicatum* had been reported to have antioxidant activity using two *in vitro* methods, namely DPPH and -carotene linoleic acid assays . (Tepe *et al.*,(2005)

The silver nitrate extracts of *Chlorophytum borivilianum* had been shown to scavenge DPPH radical and decrease TBRAS (Thiobarbituric Acid Reactive Substances), revealing that it is a promising anti-stress agent as well as a potential antioxidant. (Kenjale *et al.*, 2007).

Antibacterial Activity

Musa *et al.*, (2000) studied the phytochemistry of powdered leaves of *Acalypha recemosa* (Euphorbiaceae). This study revealed the presence of alkaloid, tannin, flavanoid and terpenes. Antimicrobial activities of cold water, hot water and methanolic extracts were studies against *Staphylococcus aureus* was more than *Escherichia coli* but *Candida albicans* was completely resistant to the extracts. The cold water extracts showed activity with MIC range from 3.0 mg/ml (against *S. aureus*) to 4.0 mg/ml *Escherichia coli* for cold water and 7.0 mg/ml for the two isolates (methanolic extract). The MBC of cold water extract (6.0 mg/ml) was able causes 2 log cycle reduction of cell population in 90 minutes. Prema (2004) studied the antibacterial activity in eleven medicinal plants. The acetone extract of *Acalypha indica* was more effective against *Staphylococcus aureus*. Ethanol extract of *A. indica* and *Eucalyptus globulus* were highly sensitive to *S. aureus* and *P. Aeruginosa*.

Poonkothai *et al.*, (2005) worked on antibacterial activity of chloroform, ethanol and aqueous extracts of the leaves of *Gymnema sylvestre* on *Bacillus subtilis,*

Staphylococcus aureus, Klebsiella pneumoniae, Pseudomonas aeruginosa, Escherichia coli and *Salmonella typhi* on Muller Hindon agar plates. Commercially available chloramphenicol disc (30 mg) was used as control and discs impregnated with DMSO were also used in this technique. *Klebsiella pneumoniae* was resistant to both chloroform and ethanol extracts exhibiting a zone of inhibition of 12 and 11 mm respectively. *Pseudomonas aeruginosa* (16 and 21mm) and *Salmonella typhi* (17 and 19mm) were found to be sensitive to both the extracts. This indicates that gymnemic acid, an active component of *Gymnemasylvestre* double in both chloroform and ethanol was found to have a strong antibacterial activity. There was no significant effect of aqueous extract because there was no zone of inhibition.

Akinpelu *et al.*, (2009) studied the medicinal plants *Jatropha curcas*and *Newboulda laevis*. Methanolic leaf extract of *J. curcas, N. laevis* exhibited antibacterial activity against 8 of the thirteen tested bacterial isolates at a concentration of 20 mg/ml. The zones of inhibition exhibited by *J. curcas*ranged between 18 and 17mm. *N. laevis* varies between 10 and 23 mm.

Dhale and Birari (2010) studied the antimicrobial effect of *Jatropha gossypifolia* leaf extracts on gram positive species *Staphylococcus spp.* and *Bacillus spp.* and gram negative species like *Escherichia spp.* and *Pseudomonas spp.,* in solvents like petroleum ether, alcohol and chloroform. The method employed was disc diffusion method, standard was Amphicillin, the alcoholic extract of leaves showed maximum antibacterial activity.

Dipankar Choudhury *et al.*, (2011) studied phytochemical screening and antimicrobial activity of extracts from leaves and stem of *Ecbolium linnean* .The bacterial pathogens were strongly inhibited by leaf extracts but acetone extracts of stem have failed to inhibit the growth of *Staphylococcus aureus* and *Pseudomonas aeruginosa* even at the highest concentration. The results revealed that leaf extracts were found to be more effective than

stem extracts. *E. linneanum* possesses antimicrobial activity against most commonly encountered human pathogens.

Yusha'u, *et al.*, (2011) studied antibacterial activites of ethanolic extracts of *Annona squamosal (L.)* leaves were studied against clinical respiratory tract isolates of *Klebsiella pnemoniae, Proteus species, Pseudomonas species, Staphylococcus aureus, Streptococcus pnemoniae* and α- haemolytic *Streptococci* using disc diffusion and micro broth dilution techniques. Sensitively test results showed that water fraction of the plant was active on *Stephylococcus aures and Streptococcus pnemoniae* (10mm) at 50µg/disc concentration while ethanolic extract of the plant was active , *Streptococcus pnemoniae*and *Proteus species* at 200µg/disc concentration with zone diameter formed by *Klebsiella pnemoniae*(11mm) being wider than that formed in response to standard Augmentin disc (06mm).

Nidhi uttamkumar and sumitkumar (2013) evaluated antibacterial activity of rhizome of *Barleria prionitis*. The methanol extract showed antibacterial activity against two Gram's positive *(S. aureus and B. cereus*) and two Gram's negative (*E. coli and S. typhi*) bacteria. The antibacterial potential was measured by agar disc plate method. The active phytocomponents of *Barleria prionitis* were revealed using Gas chromatography with mass spectrophotometric detector and 27 constituents identified, Phthalazine was the most abundant phytocompound in methanol extract. All the results supported that the extract can be used to prevention of bacterial infection and may have role in pharmaceutical medicine evolution.

Nayan R. Bhalodia and V.J.Shukla (2014) reported extracts obtained from *Cassia fistula* show strong activity against most of the tested bacterial and fungal strains. The results were compared with standard antibiotic drugs. The results show that the activity of hydroalcohol extracts of *Cassia fistula* shows significant antibacterial and antifungal activities.

Niveditapatel *et al.*, (2014) reported phytochemical analysis and antibacterial activity of *Moringa oleifera*. The result showed that the plant leaves are very good nutrient supplement for malnutrition and also used as an antibiotic. To evaluate the antibacterial activity of *Moringa oleifera* leaf extracts, *Escherichia coli, Pseudomonas aeroginosa, Staphylococcus aureus, Proteus vulgaris, Streptococcus mutans, Bacillus subtilus,* and *Staphylococcus epidermidis bacteria* were used. Phytochemical analysis of the leaf in solvents of varying polarity; viz., aqueous, ethanol were also carried out. The phytochemical screening indicated the presence of flavonoids, tannins, steroid, alkaloid, saponins etc. in the both extracts. Well diffusion method was used to assess the antibacterial effect of the extracts on micro-organisms. The ethanolic and aqueous extract were active against all strains but the ethanol leaf extract showed maximum activity against *Streptococcus mutant* and aqueous extract shows maximum activity against *Proteus vulgaris*.

Hassan Waseem *et al.*, (2016) detected the antimicrobial activity of *C. tamala* against a number of organisms. The Plant extract from *C. tamala* was found to have antimicrobial activity against only one tested bacterium, *S. aureus* (ATCC 25293). They found different degrees of antimicrobial activity against all tested gram positive and gram negative bacteria contrary to our result where only *S. aureus* was found to be effective.

MATERIALS AND METHODS

Plant material

Botanical Name - *Syzygium cumini* (L) Skeels

CLASSIFICATION :

Kingdom : Plantae

Division : Tracheophyta

Sub – Division : Spermatophytina

Class : Magnoliopsida

Sub – Class : Rosidae

Order : Myrtales

Family : Myrtaceae

Distribution :

The original home of *Syzygium cumini* (L) Skeels is India or the East Indies. It is found in Thailand, Philippines, Madagascar and some other countries. The plant has been successfully introduced into many other tropical countries such as the West Indies, East and West Africa and some sub-tropical regions including Florida, California, Algeria and Israel.

Botanical Description :

Syzygium cumini (L) Skeels - A smooth tree of the Myrtaceae family, It is also known as black plum, jambolan, Java plum, purple plum, Malabar plum, Portuguese plum, Damson plum, and Duhat plum. It is 4-15 meters in height. Leaves leathery oblong-ovate to elliptical and 6-12 cm long, the tip is broad and shortly pointed. The panicles are borne mostly from the branchlets below the leaves, often being axillary or terminal and 4-6 cm long. Flowers are characterized by the presence of a prominent cup-shaped receptacle and an inferior ovary. The flowers are numerous, pink or nearly white, without stalks, and borne in crowed fascicles on the ends of the branchlets. The calyx is funnel-shaped, about 4 mm long, and 4-toothed. The petals cohere and fall together as a small disk. Stamens are arranged on the receptacle whereas solitary style is placed in the middle of the floral axis. The stamens are very numerous and as long as the calyx. The fruit is oval to elliptic; 1.5-3.5 cm long, dark purple or nearly black, luscious, fleshy and edible; it contains a single large seed.

COLLECTION AND IDENTIFICATION OF PLANT MATERIALS

The fresh plant materials of leaf and seed of *Syzygium cumini* were collected from Thoothukudi, Tamil Nadu. The collected samples were cut into small fragments and shade dried until the fracture is uniform and smooth. The dried plant material was granulated or powdered by using a blender, and sieved to get uniform particles by using sieve No. 60. The final uniform powder was used for extraction of active constituents of the plant materials.

QUALITATIVE ANALYSIS

Water soluble extractive

Two grams of the shade-dried powder of *Syzygium cumini* was macerated with 50 ml water in a closed flask for 24 hours. Shaking frequently during the first 6 hours and being allowed to stand for 18 hours. It was filtered using a muslin cloth and used for phytochemical analysis.

Methanol soluble extractive

Two grams of the shade-dried powder of *Syzygium cumini* was macerated with 50 ml methanol in a closed flask for 24 hours. Shaking frequently during the first 6 hours and being allowed to stand for 18 hours. It was filtered using a muslin cloth and used for phytochemical analysis.

Acetone soluble extractive

Two grams of the shade dried powder of *Syzygium cumini* was macerated with 50 ml acetone in a closed flask for 24 hours. Shaking frequently during first the 6 hours and allowed to stand for 18 hours. It was filtered using a muslin cloth and used for phytochemical analysis.

Ethanol soluble extractive

Two grams of the shade-dried powder of *Syzygium cumini* was macerated with 50 ml ethanol in a closed flask for 24 hours. Shaking frequently during first the 6 hours and allowed to stand for 18 hours. It was filtered using a muslin cloth and used for phytochemical analysis.

Test for tannins (Ciulei I.)

To 1 ml of the extract, 2 ml of 5% $FeCl_3$ was added. A dark blue or green-black indicates the presence of tannins.

Test for saponins (Harbronejb)

Foam test

The crude extract is mixed with 5 ml of distilled water and shaken vigorously, resulting in the formation of stable foam which is a positive indication for saponins.

Test for Flavonoids (Savithramma*et al* and selvaraj *et al.*,)

For the identification of flavonoids, 2ml of plant extract, 1ml of 2N sodium hydroxide (NaOH) were added. The Formation of yellow colour indicates the presence of flavonoids.

Test for Coumarins (Harbrone JB)

For identification of coumarins, 1ml of plant extract, 1ml of 10% NaOH were added. The Formation of yellow colour indicates the presence of coumarins.

Test for Terpenoids (Harbrone JB)

For identification of terpenoids, 0.5 ml of the plant extract, 2ml of chloroform along with concentrated Sulphuric acid. The Formation of red brown colour at the interface indicates the presence of Terpenoids.

Test for Quinines (P. D. Egwaikhide and C. E. Gimba)

A small amount of extract was treated with concentrated HCl and observed for the formation of a yellow colour precipitate.

Test for Alkaloids (E. C. G. Clarke)

Wagner's test

A fraction of the extract was treated with Wagner's reagent (1.27 g of iodine and 2 g of potassium iodide in 100 ml water) and observed for the formation of reddish-brown colour precipitate. There was a formation of reddish brown colour confirming the presence of alkaloid.

Test for Sterols (P. D. Egwaikhide and C. E. Gimba)

Extract (1 ml) was treated with chloroform, acetic anhydride and drops of H_2SO_4 were added and observed for the formation of dark pink or red colour. No dark pink or red colour precipitate, absence of sterols.

Test for Carbohydrate (Harbrone JB)

Fehling's test

5 ml of Fehling's solution was added to 0.5 mg of extract and boiled in a water bath. The formation of a yellow or red precipitate indicates the presence of reducing sugars.

Test for Glycosides (E. C. G. Clarke)

0.5 mg of extract was dissolved in 1 ml of water and then an aqueous NaOH solution was added. The Formation of yellow colour indicates the presence of glycosides.

Test for Protein (Harbrone JB)

Ninhydrin test:

0.5 mg of extract was taken and 2 drops of freshly prepared 0.2% ninhydrin reagent were added and heated. The appearance of pink or purple colour indicates the presence of proteins, peptides or amino acids.

Test for phenol (Harbrone JB)

To 1 ml of the extract, 2 ml of distilled water was added and followed bya few drops of 10% aqueous ferric chloride. The Appearance of blue or green colour indicates the presence of phenols.

Quantitative analysis of antioxidant

Total phenolic content :(Duan *et al.*, 2006)

Reagents

- 50%Folin – ciocalteau reagent
- 20%sodium – carbonate
- Gallic acid – standard

Procedure

100mg of samples were homogenated with 10 ml of distilled water and filtered through a muslin cloth. 1 ml of the filtrate was added to 1.5 ml of deionized water and 0.5 ml of 50% folinciocalteau reagent and the contents were mixed thoroughly. After 1min, 1 ml of 20% sodium carbonate solution was added and mixed the control contained all the reagents except the sample. After 30 minutes of incubation at 37°c, the absorbance was measured at 750nm. Total phenolics were calculated as Gallic acid equivalent (GAE) per gram fresh weight.

Total flavonoid content (Zhinshen *et al.*, 1999)

Reagents

- 5% sodium nitrate (NaNo2)
- 10% Aluminium chloride (Alcl3, H_2O)
- 1N sodium hydroxide (NaOH)
- Quercetin standard

Procedure

100mg of plant material was homogenized with 10ml of distilled water and filtered through a muslin cloth. 0.5 ml of the extract was added with 2.5 ml distilled water and mixed. After 6 minutes 0.15 ml NaOH, was added and again after 6min 0.3 ml of 10% $Alcl^3$ was added. After 5 minutes 1ml of 1M NaOH and 0.5 ml of water were added. Following through mixing of the solution the absorbance against blank were recorded at 510nm. Quercetin was used as standard and the results were expressed as my quercetin equivalents (QE) 1g fresh weight.

Vitamin C [Ascorbic acid] (Baker and Frank, 1968)

Reagents

- 5% of TCA
- Indophenols reagent
- 20mg of dichlorophenol indophenols was dissolved in 10ml of warm distilled water
- DT reagent 2g of 2, 4 dinitraphenyl hydrazine and 1g of thiourea were dissolved.
- 85% sulphuric acid
- L-ascorbic acid - standard

Procedure

100 mg of plant material was homogenized with 10ml of 5% Trichloro acetic acid (TCA). The homogenate was centrifuged. To 2 ml of indophenols reagent and 0.5ml of DT reagent was added and incubated at 10c for 1hour and then cooled in ice bath and 2.5 ml of 85% sulphuric acid was added and shaken well for 30 minutes (until) red colour appeared. The absorbance was measured at 540nm. 1-ascorbic acid was used as standard and the results were expressed as mg/1g/FW.

Estimation of Tannin (Julkunen-Titto, 1985)

Procedure

100 mg of sample homogenized with 10 ml of distilled water and filtrated through a muslin cloth. 1ml of aliquot of aqueous extract was mixed with 1.5ml of 4% vanillin (prepared with methanol) and 750 µl of concentrated HCL was added the solution was shaken vigorously and left to stand at room temperature for 20 minutes in darkness the absorbance against blank was read at 500nm using UV-Visible spectrophotometer. Results were expressed as mg catechin equivalent (CE) 1g tissue.

FT-IR analysis

A little powder of plant specimen was mixed with KBr salt, using a mortar and pestle, and compressed into a thin pellet. Infra -red spectra were recorded as KBr pellets on a Thermo Scientific NicotiS5ID1 transmission, between 4000-400 cm^{-1} (Kareru *et al.*, 2008).

GC-MS Analysis:

Extract Preparation

The 50g tuber powder of *Syzygium cumini* was serially extracted with 250 ml of Methanol with the help of Soxhlet apparatus. The extraction procedures were continued for 3-4 hours at 60°C -80°C15. These extracts were concentration under reduced pressure evaporator and stored in air tight vials at 4°C for further study.

Phytochemical analysis by GC-MS (Hema *et al.*, 2011)

Gas chromatography-Mass spectrometry (GC-MS) analysis of the ethanolic extracts was performed by using a GC-MS (Model; QP2010 series, Shimadzu, Tokyo, Japan) equipped with a VF-5ms fused silica capillary column of 30 m length, 0.25 mm dia.and0.25µm film thickness. For GC-MS detection, an electron ionization system with ionization energy of 70 eV was used. Helium gas (99.99%) was used as a carrier gas at a

constant flow rate of 1.51 ml/min. Injector and mass transfer line temperature was set at 200 and 240°C respectively. The oven temperature was programmedfrom70to220°Cat10°C/min, held isothermal for 1 min and finally raised to 300°C at 10°C/min. 2 µl of respective diluted samples was manually injected in the split less mode, with split ratio of 1:40 and with mass 18 scan of 50-600 amu. Total running time of GC-MS is 35min. The relative percentage of the each extract constituents was expressed as percentage with peak area normalization.

Identification of phytochemical components

The identity of the components in the extracts was assigned by the comparison of their retention indices and mass spectra fragmentation patterns with those stored on the computer library and also with published literatures. NIST08s. LIB and WILEY 8. LIB library sources were used for matching the identified components from the plant material.

ANTIOXIDENT ACTIVITY

Crude samples extracts were prepared by pouring 100ml of distilled water in a conical flask containing 10g of each samples separately in the ratio of 10:1 (V/W). After 24 hours, the mixture was filtrated through whatman no: 1 filter paper and the filtrate were evaporated to dryness. Crude (aqueous) extracts of all samples (1mg/ml) were used for the determination of free radical scavenging activity.

Free radical scavenging assays (Hatano *et al.*, 1998).

Free radical scavenging assay was measured by 2-2 Diphenyl, 1-picryl hydrazine (DPPH) method proposed by with slight modifications. 1ml of aliquot of test sample was added to 3ml of 0.004% DPPH solution prepared in methanol. The mixture was vortexes for 1min and kept at room temperature for 30 minutes in darkness the absorbance was read at 517 nm. Allow absorbance of the reaction mixture indicated a high free radical scavenging activity. Ascorbic acid was used as standard.

DPPH scavenging activity (%)

A control –A test / A control * 100

Where, a control is the absorbance of the DPPH solution without test solution. A test is the absorbance of DPPH with the test solution. Aqueous extract was used as blank.

Antibacterial studies

Extraction of plant materials

The plant powder was extracted with methanol, ethanol, acetone and water.25 gms of plant powder was extracted with methanol, acetone and water solution individually in soxhlet apparatus continuously for about 4-6 hours, which was again concentrated till it become semi- solid. It was evaporated to dryness and stored at 0 C, until the time of the experiment.

Bacterial strains used

The test organisms were obtained from the Department of Microbiology; St. Mary's College (Autonomous), Thoothukudi. One gram-positive bacteria viz; *Bacillus subtilis* G-ve MTCC 1133 and four gram-negative bacteria *Escherichia coli*, G-ve, MTCC 50, *Staphylococcus* G-ve, 737were used in the present study.

Broth Medium:

- Nutrient broth Himedia MOO1
- Nutrient broth 1.3 gm
- Distilled water 100 ml

2-3 ml of sterilized broth medium was taken in the culture tube. The inoculating loop was flamed and after a few minutes a loopful bacterial colony was transferred to the broth medium. This microbe culture was incubated at room temperature for 24 hours.

Agar medium:

- Nutrient broth Himedia MOO1
- Nutrient broth 1.3 gm
- Distilled water 100 ml

To prepare the agar medium all the above ingredients were dissolved and sterilized.

Disc diffusion method

Anti- bacterial activity was evaluated by agar disc diffusion method (Kirby – Bauer *et al.*, 1986). Test solution was prepared with known weight of methanol, ethanol, acetone and water extracts dissolved in 5% dimethyl sulphoxide (DMSO). What man No.1 filter paper disc (5mm) was impregnated with 20 of these extracts and allowed to dry at room temperature. The spread plates were prepared by proper concentration of inoculate. Each sample loaded discs was placed in the seeded agar plate. 24-48 hours of + 37^0c incubation, the diameter of the inhibition zone was for positive control, amoxicillin discs (100g/ml) was used, whereas for negative control; respective solvents loaded on the sterile discs.

RESULT AND DISCUSSION

The plant kingdom is a treasure house of potential drugs used for the prevention and treatment of ailments. Plants have been major sources of bioactive principles employed in drug formulations both modern and traditional medicine. According to the World Health Organization, 80% of the people living in rural areas depend on medicinal herbs as a primary health care system. (Sakarkar and Deshmukh, 2011)

Syzygium cumini (L.) Skeels. is an important medicinal plant in the family Myrtaceae was selected for the present study. Various traditional practitioners in India use the different parts of the plant in the treatment of diabetes, blisters in the mouth, cancer, colic, diarrhea, digestive complaints, dysentery, piles, pimples and stomach ache (Jain ,1991). In Unani medicine, various parts of jambolan act as a liver tonic, enrich the blood, strengthen teeth and gums and form a good lotion for removing ringworm infection of the head (Sagrawat *et al* 2006). In the present study, active constituents of the plant was analyzed and evaluated.

QUALITATIVE ANALYSIS

Preliminary phytochemical analysis of the various solvent by the leaf and seed of *Syzygium cumini* showed different results. The alkaloids, phenols, tannins, saponins, glycosides, quinones, flavonoids, terpenoids and coumarins were predominantly present in the solvent extracts. Tables (1&2)

Johnson *et al.,* (2012) reported the methanol extracts of some medicinal plants to contain tannin, saponin, flavonoids, phenol, betacyanin and coumarin. Sukumaran *et al.,* (2011) reported the presence of alkaloids, flavonoids, tannins, saponins, phenol and terpenoids in *Peltrophorum pterocarpum* flowers.

Table –1: Preliminary phytochemical screening and distribution of secondary constituents in leaves of *Syzygium cumini*

Phytochemical test	Solvent extract				
	Water	Acetone	Methanol	Ethanol	Hexane
Alkaloids	+	+	+	+	+
flavonoids	+	+	+	+	+
Sterols	+	+	+	+	-
Carbohydrates	+	+	+	+	-
Glycosides	+	+	+	+	+
Saponim	+	+	+	+	+
Protein	+	-	+	+	+
Quinone	+	+	+	+	+
Phenol	+	+	+	+	-
Coumarin	+	+	+	+	+
Tannin	+	-	+	+	-
Terpenold	+	+	+	+	+

Table – 2: Preliminary phytochemical screening and distribution of secondary constituents in seeds of *Syzygium cumini*

Phytochemical test	Solvent extract				
	Water	Acetone	Methanol	Ethanol	Hexane
Alkaloids	+	+	+	+	+
Flavonoids	+	+	+	+	+
Sterols	+	+	+	+	+
Carbohydrates	+	+	+	+	-
Glycosides	+	+	+	+	+
Saponim	-	+	+	+	+
Protein	+	+	+	+	+
Quinone	+	+	+	+	+
Tannin	+	+	+	+	-
Terpenold	+	+	+	+	+
Phenol	+	+	+	+	+
Coumarin	-	+	+	+	+

TOTAL PHENOL

Phenolics are the most widespread secondary metabolites and are believed to be responsible for antioxidant activity. The phenol contents of the seed extract of *Syzygium cumini* (3.453±0.451mg GAE/g) were higher than that of the leaf extract of *Syzygium cumini* (2.052±0.014mg GAE/g). (Table-3; fig:1). Phenolic compounds are a class of antioxidant agents that act as free Terminators (Shahidi and Wanasundra, 1992). Phenolic compounds have a variety of beneficial activities. They have potent antioxidants and free radical scavengers. (Meenakshi *et al.*, 2010) the antimicrobials (most of the phenolics) may provide a microbe-free environment within the body.

TOTAL FLAVONOIDS

Flavonoids are secondary metabolites and have responsible for antioxidant activity in the medicinal field. The total flavonoid contents of the seed extract of *Syzygium cumini* (3.675±1.009 mg (GAE) -g were higher than that of the leaf extract of *Syzygium cumini* (2.828±0.045 mg (GAE) – g)(Table-4; fig:2). Flavonoids are potent antioxidants and epidermic studies indicate that high flavonoids in taking are correlated with decreased risk of lifestyle diseases like diabetes and cardiovascular diseases (Kareru *et al.*,2008). Flavonoids are potent water-soluble antioxidants and free radical scavengers, which prevent oxidative cell damage and have strong anti-cancer activity (Havsteen,2012)

TOTAL VITAMIN-C

Table-5; figure:3 shows Seed extract of *Syzygium cumini* (4.389±0.004) and leaf extract of *Syzygium cumini* (3.654±0.032) contains a significant amount of vitamin C. vitamin C is a vital component in the human diet with the highest concentration in animal organs. Vitamin C is a non-enzymatic, water-soluble antioxidant (Ueta *et al.*, 2003). Vitamin C functions in enzyme activation, oxidative stress reduction, and immune function.

TABLE – 3

TOTAL PHENOL CONTENT OF SYZYGIUM CUMINI LEAF AND SEED	
SAMPLE	**AMOUNT OF PHENOLS mg (GAE)/g**
Syzygium cumini Leaf	2.052±0.014
Syzygium cumini Seed	3.453±0.451

TABLE – 4

TOTAL FLAVANOID CONTENT OF SYZYGIUM CUMINI LEAF AND SEED	
SAMPLE	**AMOUNT OF FLAVANOIDS mg (GAE)/g**
Syzygium cumini Leaf	2.828±0.045
Syzygium cumini Seed	3.675±1.009

TABLE – 5

TOTAL VITAMIN C CONTENT OF SYZYGIUM CUMINI LEAF AND SEED	
SAMPLE	**AMOUNT VITAMIN C OF mg (GAE)/g**
Syzygium cumini Leaf	3.654±0.032
Syzygium cumini Seed	4.389±0.004

TABLE – 6

TOTAL TANNIN CONTENT OF SYZYGIUM CUMINI LEAF AND SEED	
SAMPLE	**AMOUNT OF TANNINS mg (GAE)/g**
Syzygium cumini Leaf	1.119±0.239
Syzygium cumini Seed	1.389±0.004

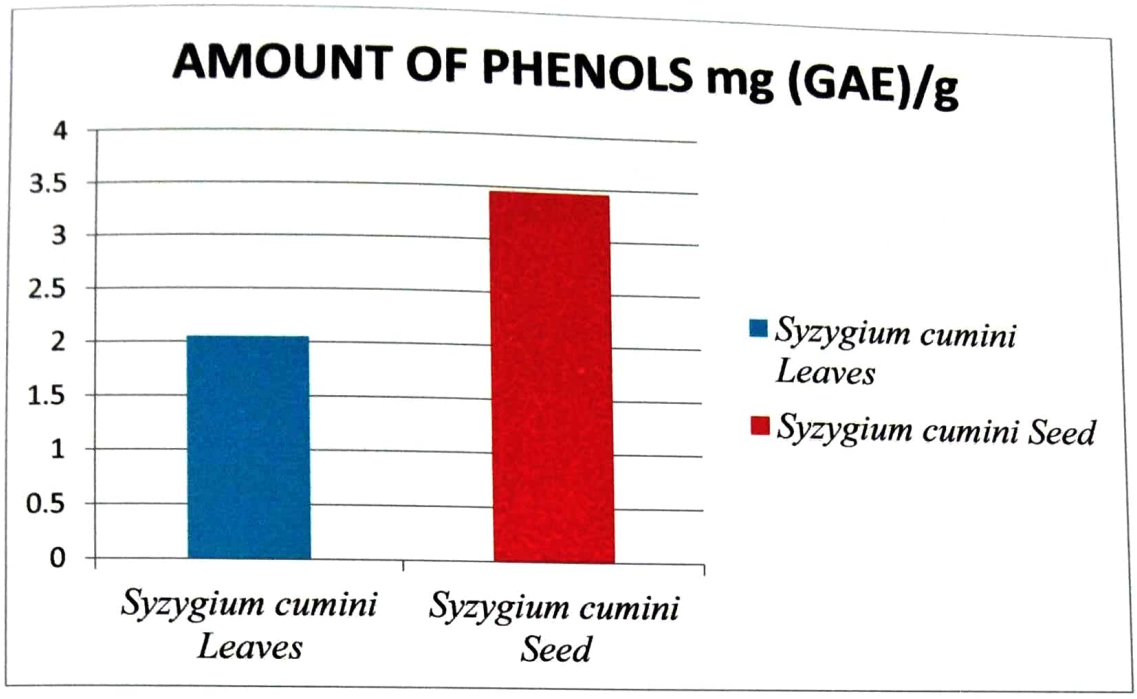


Figure : 1 TOTAL PHENOL CONTENT OF LEAF AND SEED EXTRACT OF SYZYGIUM CUMINI

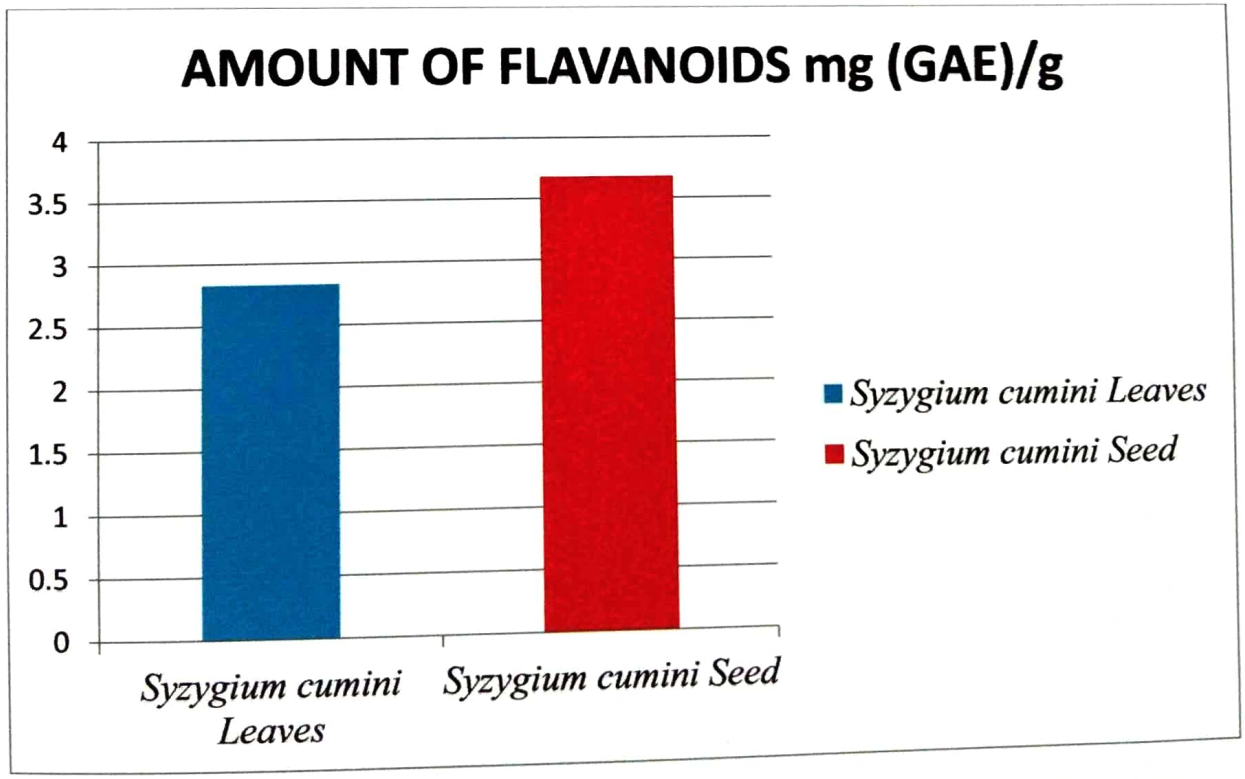


Figure : 2 TOTAL FLAVANOID CONTENT OF LEAF AND SEED EXTRACT OF SYZYGIUM CUMINI

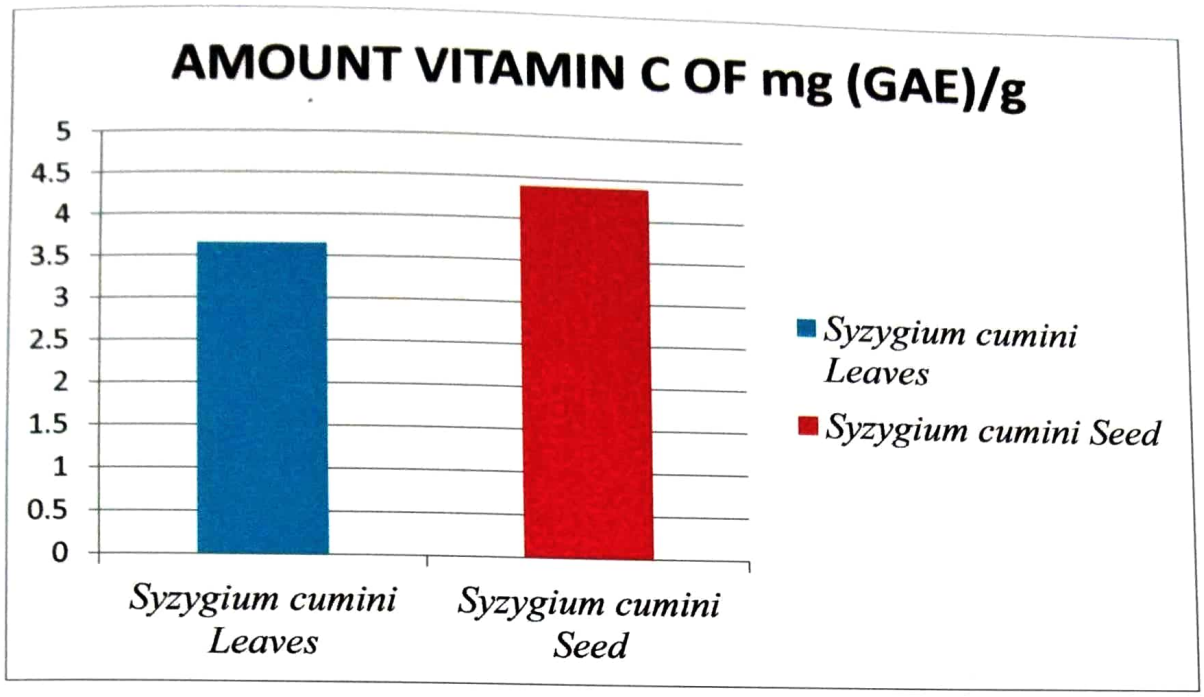


Figure : 3 TOTAL VITAMIN –C CONTENT OF LEAF AND SEED EXTRACT OF SYZYGIUM CUMINI

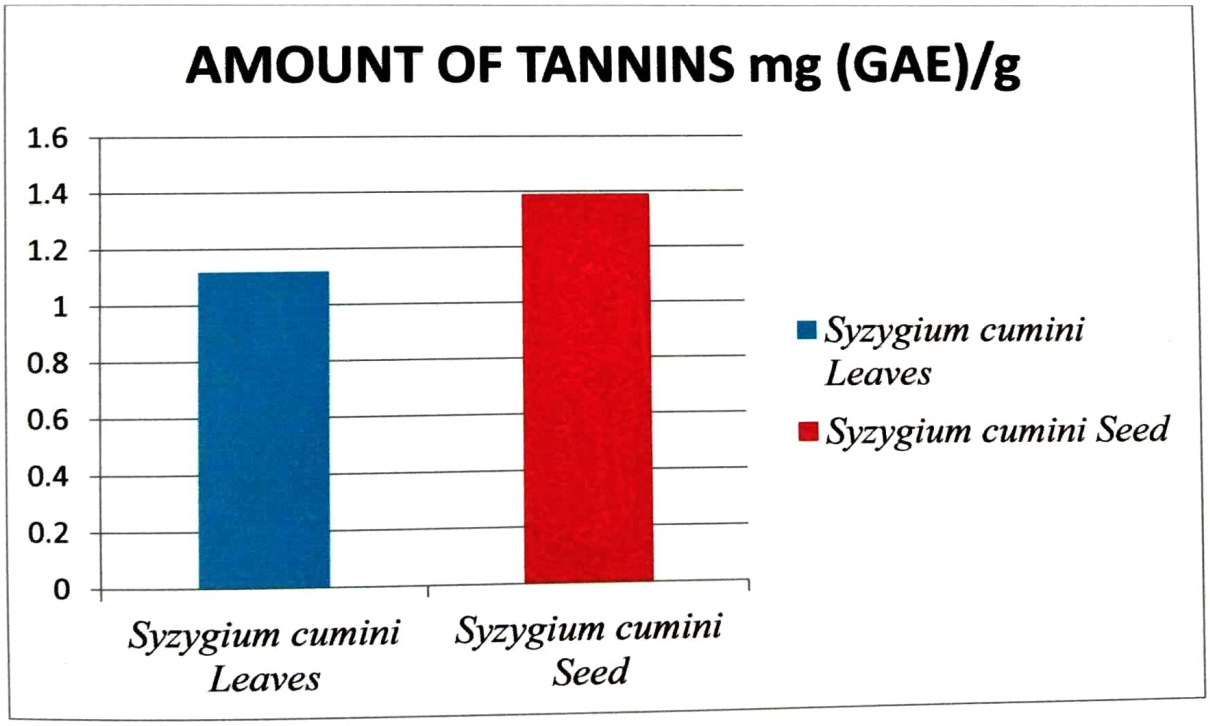


Figure : 4 TOTAL TANNINS CONTENT OF LEAF AND SEED EXTRACT OF SYZYGIUM CUMINI

It protects against respiratory tract infection and reduces the risk of cardiovascular disease and cancer.

TOTAL TANNINS:

Table 6; fig:4 shows seed extract of *Syzygium cumini* (1.124±0.025mg/g) and leaf extract of *Syzygium cumini* (1.119±0.239mg/g) contain a significant amount of tannin (Table-6). Tannins are present primarily in the leaves of trees growing in stressful conditions. They are accumulated in the vacuoles, especially those of the epidermal layer and the palisade layer and the palisade mesophyll. Tannins are useful in treating inflammation, ulcers and remarkable activity in cancer prevention and anticancer activities (Li *et al.*, 2023; Akinpelu et *al.*, 2009).

FT-IR

Fourier Transform Infrared Spectroscopy was to analzse the functional group present in *Syzygium cumini*. The FTIR spectroscopy analysis of *Syzygium cumini* seed obtained peaks at 522.67cm-1, 655.75 cm-1, 709.76 cm-1,761.83 cm-1,861.16 cm-1, 1081.99 cm-1, 1157.21 cm-1, 1456.16cm -1, 1736.78cm-1,2360.71 cm-1, 3383.87 cm-1. These absorption peaks are known to be associated with the stretching vibration for C-Br in strong aromatic, N-H in strong primary amines, N-O in weak nitrate group, C-H in strong hydrogen atom,S-O in strong sulphinic acid group, C-O in very strong ring stretch, C-O-C in strong symmetric stretch, N=O in strong amines, C=O in strong esters, H-C-H in medium asymmetric stretch, O-H in Carboxylic Acid Group (Fig-5 table-7).

The FTIR spectroscopy analysis of *Syzygium cumini* leaf obtained peaks at 599.82cm-1, 656.72cm-1, 755.8cm-1, 1100.31cm-1, 1317.29cm-1, 1400.22cm1, 3163.04cm-1. These absorption peaks are known to be associated with the stretching vibration for C-Br in Aromatic, N-H in Amines, C-H in strong hydrogen atom, C-O-C in symmetric stretch, C-N

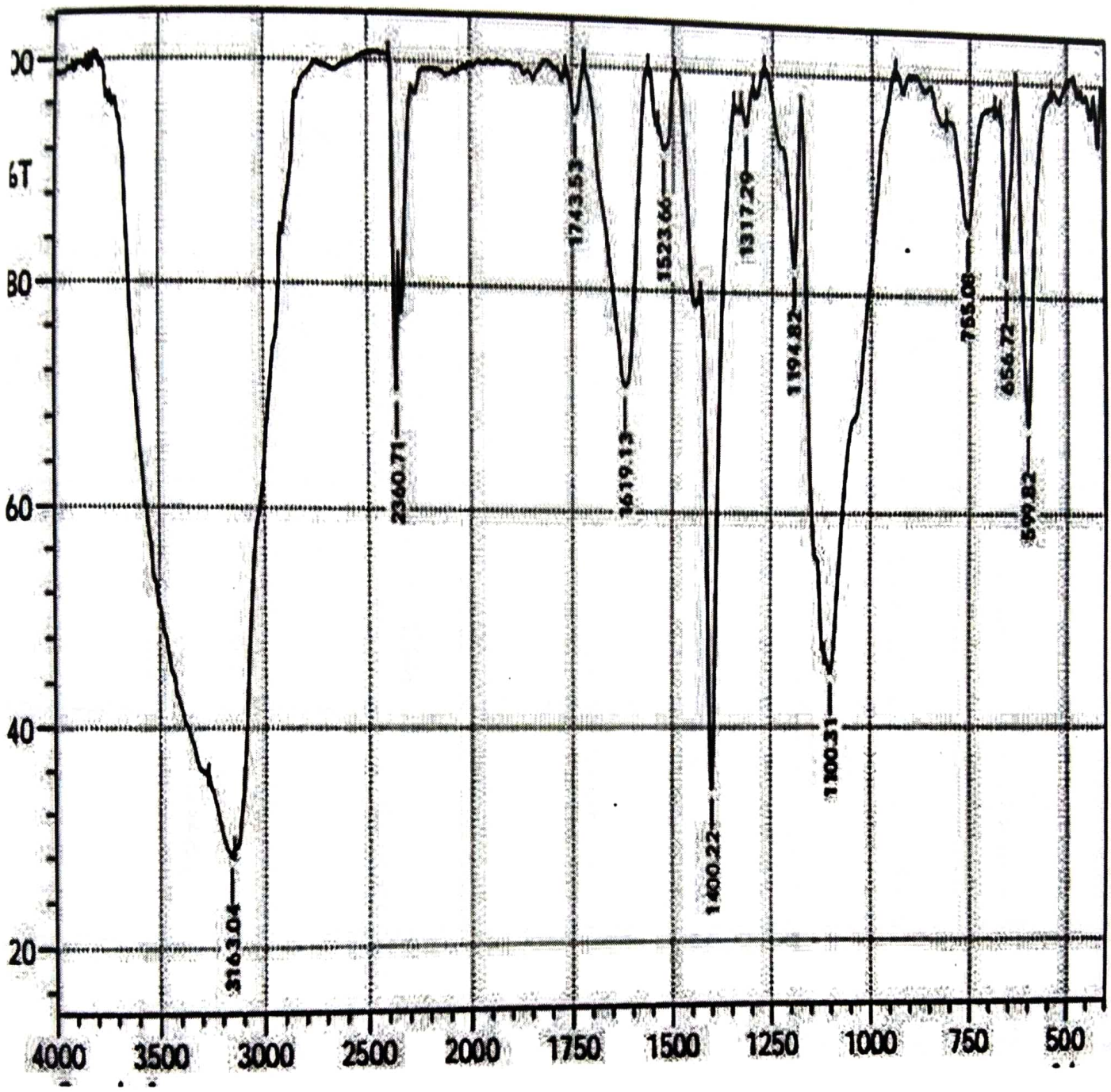


Figure 5: FT-IR Spectroscopy Analysis of *Syzygium cumini* leaf

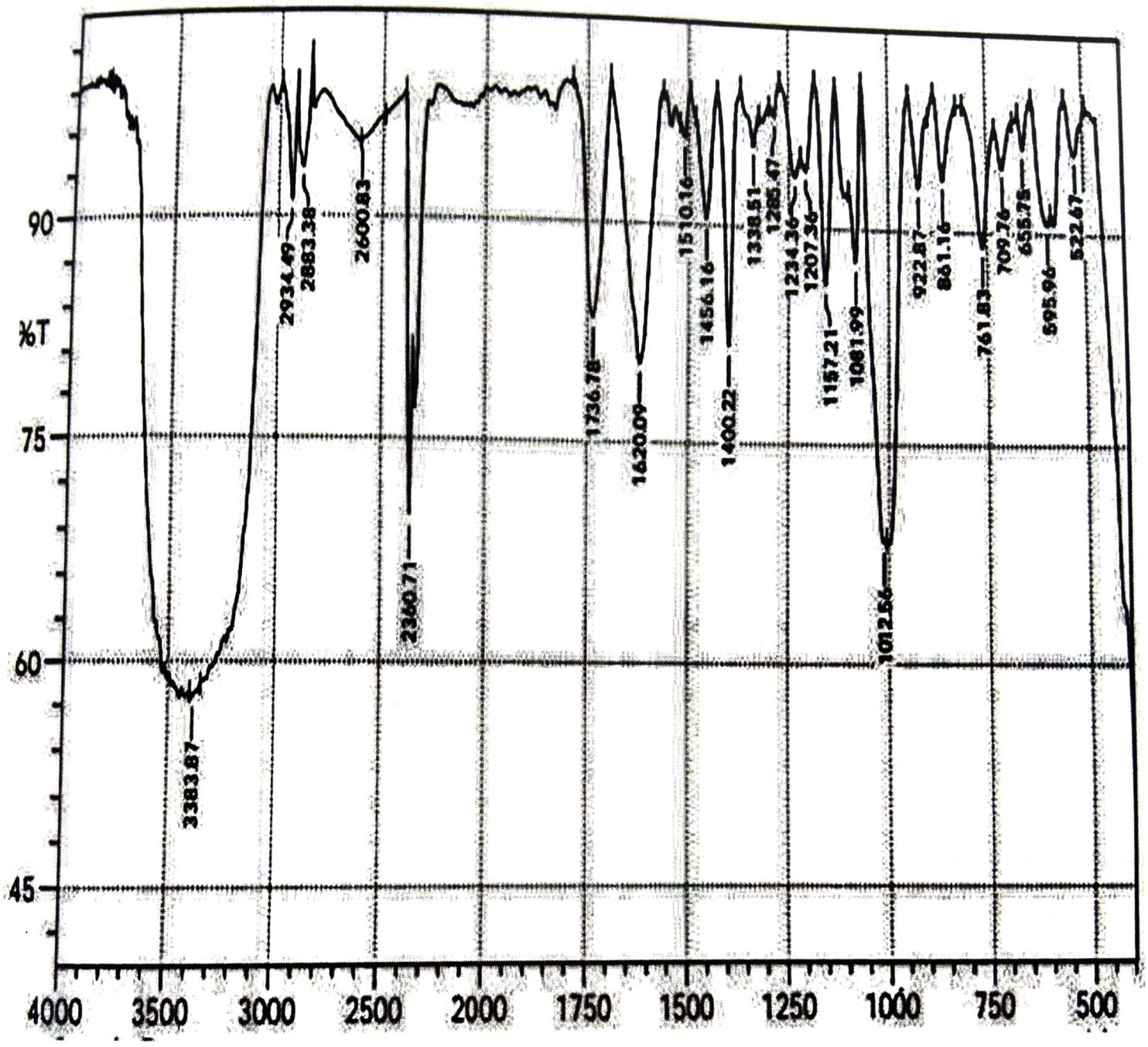


Figure 6: FT-IR Spectroscopy analysis of *Syzygium cumini* Seed

Table 7: FT-IR Spectroscopy Analysis of *Syzygium cumini* leaf

S.NO	PEAK VALUE	BOND	FUNCTIONAL GROUP
1	599.82	STRONG, AROMATIC	C-Br
2	656.72	STRONG, PRIMARY AMINES	N-H
3	755.08	STRONG, HYDROGEN ATOMS	C-H
4	1100.31	SYMMETRIC, STRETCH	C-O-C
5	1194.82	SYMMETRIC, STRETCH	C-O-C
6	1317.29	STRONG, ARYL TERTIARY AMINE	C-N
7	1400.22	STRONG AMINE	N=O
8	1523.66	AROMATIC, ASYMMETRIC	N=O
9	1619.13	MEDIUM, PRIMARY AMIDE	N-H
10	1743.53	STRONG, STRETCH	C-O-C
11	2360.71	MEDIUM, STRETCH ASYMMETRIC	H-C-H
12	3163.04	CARBOXYLIC ACID GROUP	O-H

Table 8 : FT-IR Spectroscopy Analysis of *Syzygium cumini* Seed

S.NO	PEAK	BOND	FUNCTIONAL GROUP
1	522.67	STRONG, AROMATIC	C-Br
2	595.96	STRONG, AROMATIC	C-Br
3	655.75	STRONG, PRIMARY AMINES	N-H
4	709.76	WEAK, NITRATE GROUP	N-O
5	761.83	STRONG, HYDROGEN ATOMS	C-H
6	861.16	STRONG, SULPHINIC ACID GROUP	S-O
7	922.87	STRONG HYDRO COMPOUND	C-H
8	1012.56	STRONG, ETHER	C-O
9	1081.99	VERY STRONG RING STRETCH	C-O
10	1157.21	STRONG, SYMMETRIC STRETCH	C-O-C
11	1207.36	STRONG, ARALKYL, ASYMMETRIC	C-O-C
12	1234.36	STRONG, ACETATES	C-O-C
13	1285.47	STRONG, ASYMMETRIC	C-O-C
14	1338.51	STRONG, HYDROXYL GROUP	C-O
15	1400.22	MEDIUM	C-H

16	1456.16	STRONG, AMINES	N=O
17	1510.16	WEAK, SECONDARY AMIDE	N-H
18	1620.09	MEDIUM PRIMARY AMIDES	N-H
19	1736.78	STRONG, ESTERS	C=O
20	2360.71	MEDIUM ASYMMETRIC STRETCH	H-C-H
21	2600.83	MEDIUM	H-C-H
22	2883.38	MEDIUM, ASYMMETRIC	O-H
23	2934.49	MEDIUM, CARBOXYLIC ACID GROUP	O-H
24	3383.87	CARBOXYLIC ACID GROUP	O-H

in strong aryl tertiary amine, N=O in strong amines, O-H in Carboxylic Acid Group (Fig-6 table-8).

From the spectral data presence of N=O, C-BR, N-O, C-O-C, C=O, N-H, N-O, , H-C-H, N=O, C-N and O-H were identified. These bonding are responsible for the presence of amines, Aromatic, nitrate group, symmetric stretch, Ester, amines and aryl tertiary amine Medium. The carboxylic acid present in the medicinal plant serves as the main pharmaceutical product in curing ulcers, jaundice, headache, stomatitis, hemicranias, fever, pain in the liver and treatment of rheumatic joint pain. Amides, amines and amino acids are the main groups, involved in protein synthesis (Da-young lee and Eun-Hee kim,2019). The study revealed that the seed and leaf of *Syzygium cumini* contain a considerable amount of secondary metabolites and it may be considered in the future to be used in human disease management.

GC-MS Analysis:

The GC-MS analysis of the methanolic leaf extract of *Syzygium cumini* confirmed the presence of 20 compounds with retention time. Interpretation of the mass spectrum of GC-MS was conducted using the database of NIST and WILEY libraries. Out of these 20 compounds, 7 compounds are majorly present in the leaf extract of *Syzygium cumini* respectively Dodecanoic acid, 1,2,3-propanetriyl ester (31.38%), Dodecanoic acid, 1,2,3-propanetriyl ester (31.38%), 3,5-Dimethyl-4-phenylpyridine (31.38%), 1,2,3-propanetriyl ester (18.93%), Dodecanoic acid, 1,2,3-propanetriyl ester (13.49%), Quinoline-3-carbonitrile, 2-amino-4-methyl- (13.49%), Phen-1,2-diol, 4-fluoro-5-aminoacetyl-, dimethyl ether (13.49%).

The 13 minor compounds such as 8H-Thiazolo[5,4-c]azepin-8-one, 2-amino-4,5,6,7-tetrahydro- (1.60%), 5,8-Methano-4H-3,1-benzoxazine-2-thione, 1,2,4a-rel,5-trans,6,7,8-trans, 8a-cis-octahydro- (1.60%), 3-Dibenzofuranamine (1.60%), Dodecanoic

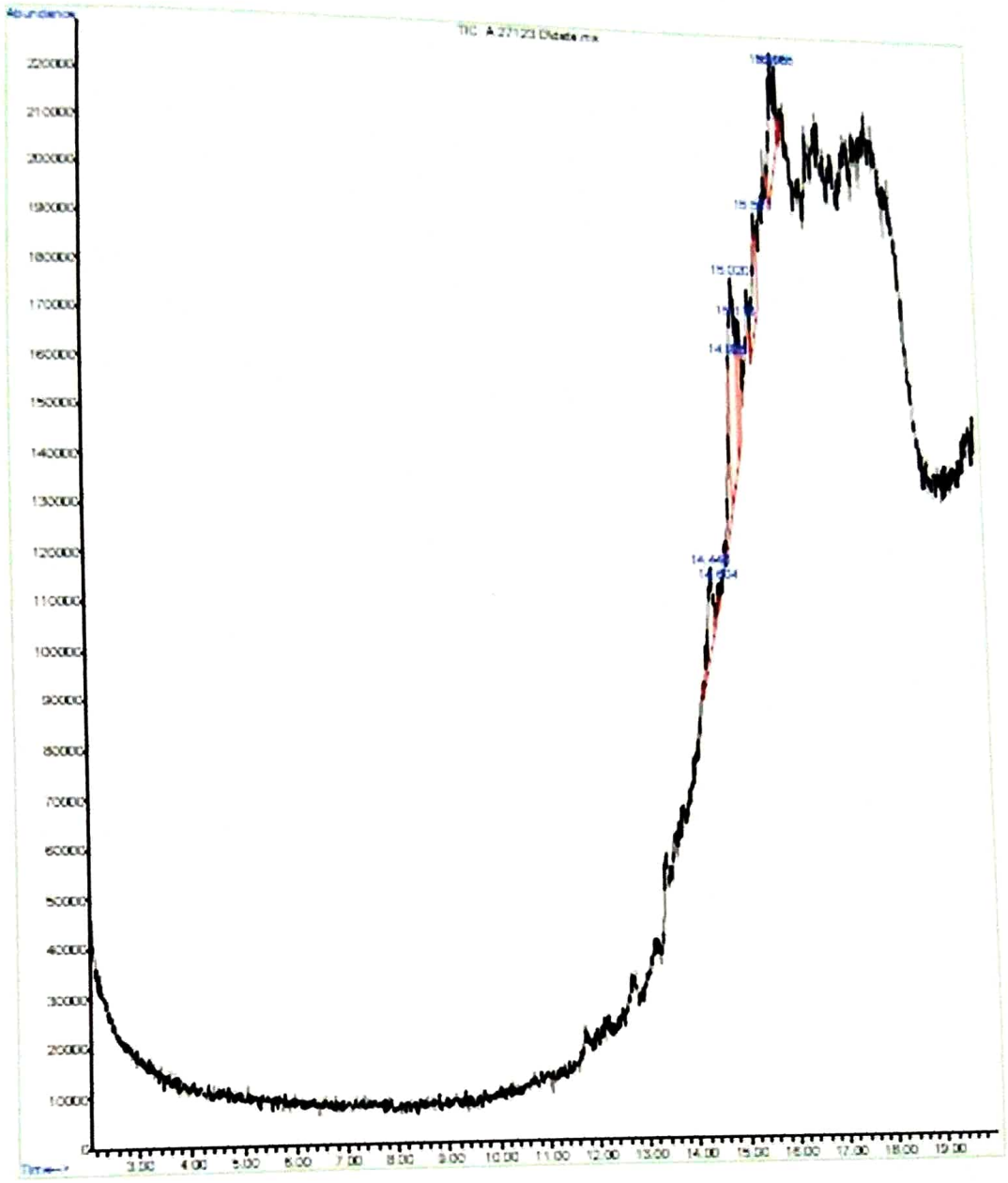
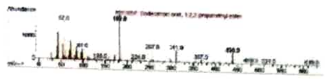
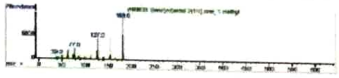
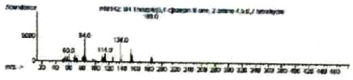
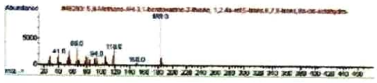
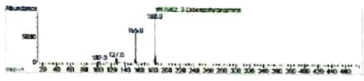
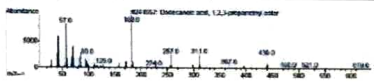
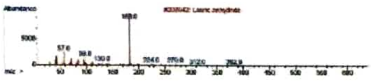
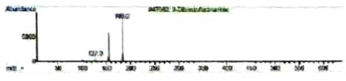
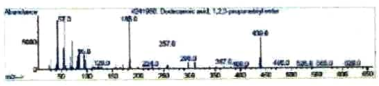
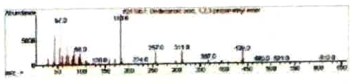
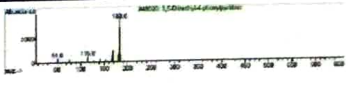
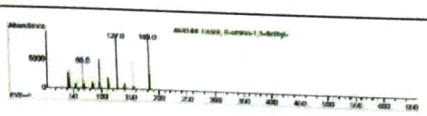
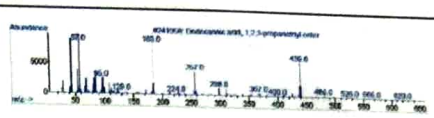
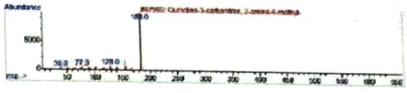
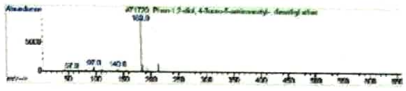
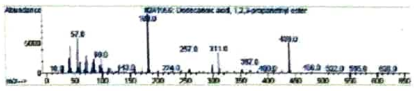
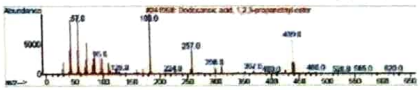
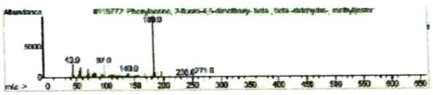


Figure: 7 GC -MS Chromatogram of methanolic leaf extract of *Syzygium cumini*

Table 9:Mass spectrum of *Syzygium cumini* leaf

S.No	R.T	Name of the compound	Area %	Mass spectrum
1.	14.445	Dodecanoic acid, 1,2,3-propanetriyl ester	12.24	
2.	14.445	Dodecanoic acid, 1,2,3-propanetriyl ester	12.24	
3.	14.445	Benz(cd)indol-2(1H)-one, 1-methyl-	12.24	
4.	14.606	8H-Thiazolo[5,4-c]azepin-8-one, 2-amino-4,5,6,7-tetrahydro-	1.60	
5.	14.606	5,8-Methano-4H-3,1-benzoxazine-2-thione, 1,2,4a-rel,5-trans,6,7,8-trans,8a-cis-octahydro-	1.60	
6.	14.606	3-Dibenzofuranamine	1.60	
7.	14.927	Dodecanoic acid, 1,2,3-propanetriyl ester	12.83	
8.	14.927	Lauric anhydride	12.83	
9.	14.927	3-Dibenzofuranamine	12.83	
10.	15.022	Dodecanoic acid, 1,2,3-propanetriyl ester	31.38	
11.	15.022	Dodecanoic acid, 1,2,3-propanetriyl ester	31.38	
12.	15.022	3,5-Dimethyl-4-phenylpyridine	31.38	
13.	15.116	Dodecanoic acid, 1,2,3-propanetriyl ester	7.45	

14.	15.116	Uracil, 6-amino-1,3-diethyl-	7.45	
15.	15.551	Dodecanoic acid, 1,2,3-propanetriyl ester	13.49	
16.	15.551	Quinoline-3-carbonitrile,2-amino-4-methyl-	13.49	
17.	15.551	Phen-1,2-diol,4-fluoro-5-aminoacetyl-, dimethyl ether	13.49	
18.	15.996	Dodecanoic acid, 1,2,3-propanetriyl este	18.93	
19.	16.119	Dodecanoic acid, 1,2,3-propanetriyl ester	2.08	
20.	16.119	Phenylserine, 2-fluoro-4,5-dimethoxy-.beta.,.beta.-didehydro-, methyl(ester	2.08	

LIST OF CHEMICAL COMPOUND IDENTIFIED FROM METHANOLIC LEAF EXTRACTOOF SYZYGIUM CUMINI THROUGH GC-MS ANALYSIS

S.No	R.T	Name of the compound	Area %	Biological
1.	14.445	Dodecanoic acid, 1,2,3-propanetriyl ester	12.24	Antithrombotic, Respiratory analeptic, Antiviral, Antiinflammatory, Anticonvulsant, Antitoxic, Antipyretic, Antifungal, Antioxidant,
2.	14.445	Dodecanoic acid, 1,2,3-propanetriyl ester	12.24	Antiinfective, Antiviral , Antiinflammatory, Antitoxic, Antipyretic, Antifungal, Antioxidant, Antidiabetic symptomatic, Antimyopathies,
3.	14.445	Benz(cd)indol-2(1H)-one, 1-methyl-	12.24	Antihypoxic, Antiviral, Antineurotic, Antiviral
4.	14.606	8H-Thiazolo[5,4-c]azepin-8-one, 2-amino-4,5,6,7-tetrahydro-	1.60	Antiischemic,Antiulcerative, Kidney function stimulant,Antiinflammatory, Anticonvulsant
5.	14.606	5,8-Methano-4H-3,1-benzoxazine-2-thione, 1,2,4a-rel,5-trans,6,7,8-trans,8a-cis-octahydro-	1.60	No active found
6.	14.606	3-Dibenzofuranamine	1.60	HIF1A expression inhibitor,Antiviral,Antiseborrheic,Antiinflammatory,Antitu berculosic Antiviral,Antiparasitic
7.	14.927	Dodecanoic acid, 1,2,3-propanetriyl ester	12.83	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective, Antiviral
8.	14.927	Lauric anhydride	12.83	Antieczematic, Antiviral, Antiinflammatory, Antipruritic, Antimutagenic
9.	14.927	3-Dibenzofuranamine	12.83	HIF1A expression inhibitor,Antiviral,Antineuroti c,Antischistosomal,Antihypox ic,Antipyretic
10.	15.022	Dodecanoic acid, 1,2,3-propanetriyl ester	31.38	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective, Antiviral
11.	15.022	Dodecanoic acid, 1,2,3-propanetriyl	31.38	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective,

		ester		Antiviral
12.	15.022	3,5-Dimethyl-4-phenylpyridine	31.38	Antiseborrheic, Antineurotic, Antinociceptive, Antiviral, Diabetic neuropathy treatment
13.	15.116	Dodecanoic acid, 1,2,3-propanetriyl ester	7.45	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective, Antiviral
14.	15.116	Uracil, 6-amino-1,3-diethyl-	7.45	Kidney function stimulant, Cardiovascular analeptic, Antiviral, Antidiarrheal, Antitoxic, Antihelmintic
15.	15.551	Dodecanoic acid, 1,2,3-propanetriyl ester	13.49	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective, Antiviral
16.	15.551	Quinoline-3-carbonitrile, 2-amino-4-methyl-	13.49	Antineoplastic, Antiviral, Antiinfective, Antineoplastic (brain cancer), Antiinflammatory, Immunomodulator, Antiuremic
17.	15.551	Phen-1,2-diol, 4-fluoro-5-aminoacetyl-, dimethyl ether	13.49	Antidyskinetic, Antineurotic, Antimyopathies, Antineoplastic, Antinociceptive,
18.	15.996	Dodecanoic acid, 1,2,3-propanetriyl este	18.93	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective, Antiviral
19.	16.119	Dodecanoic acid, 1,2,3-propanetriyl ester	2.08	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective, Antiviral
20.	16.119	Phenylserine, 2-fluoro-4,5-dimethoxy-.beta.,.beta.-didehydro-, methyl(ester	2.08	Antinociceptive, Antianorexic, Antimyopathies, Antineurotic, Antiinflammatory, Cancer associated disorders treatment

acid, 1,2,3-propanetriyl ester (2.08%), Phenylserine, 2-fluoro-4,5-dimethoxy-.beta.,.beta.-didehydro-, methyl(ester (2.08%), Dodecanoic acid, 1,2,3-propanetriyl ester (7.45%), Uracil, 6-amino-1,3-diethyl- (7.45%), Dodecanoic acid, 1,2,3-propanetriyl ester (12.83%), Lauric anhydride (12.83%), 3-Dibenzofuranamine (12.83%), Dodecanoic acid, 1,2,3-propanetriyl ester (12.24%), Dodecanoic acid, 1,2,3-propanetriyl ester (12.24%), Benz(cd)indol-2(1H)-one, 1-methyl- (12.24%) were also reported from the leaf extract of *syzygium cumini* . The chemical constituents analysis result of *syzygium cumini* leaf was reported in table-8 and their GC-MS chromatogram is presented in Fig-7; table-9.

The first compound identified with less retention (14.445 min) was, Dodecanoic acid, 1,2,3-propanetriyl ester and Benz(cd)indol-2(1H)-one, 1-methyl- whereas Dodecanoic acid, 1,2,3-propanetriyl ester and Phenylserine, 2-fluoro-4,5-dimethoxy-.beta.,.beta.-didehydro-, methyl(ester) was the last compound which took the longest retention time (16.119 min) to identify. At (15.022 min) retention time Dodecanoic acid, 1,2,3-propanetriyl ester and 3,5-Dimethyl-4-phenylpyridine were found to be high (31.38%) and the lowest percentage (1.60%) was found to be 8H-Thiazolo[5,4-c]azepin-8-one, 2-amino-4,5,6,7-tetrahydro- and 5,8-Methano-4H-3,1-benzoxazine-2-thione, 1,2,4a-rel,5-trans,6,7,8-trans, 8a-cis-octahydro- and 3-Dibenzofuranamine.

The GC-MS analysis of methanolic seed extract of *syzygium cumini* confirmed the presence of 55 compounds with retention time. Interpretation of the mass spectrum of GC-MS was conducted using the database of NIST and WILEY libraries. Out of these 55 compounds 13 compounds were majority present in the seed extract of *syzygium cumini* respectively Dodecanoic acid, 1,2,3-propanetriyl ester (21.21%), Octadecane, 3-ethyl-5-(2-ethylbutyl)- (21.21%), Dodecanoic acid, 1,2,3-propanetriyl ester (12.16%), Acetamide, 2-(4-iodopyrazol-1-yl)-N-(2-trifluoromethylphenyl)- (12.16%), Dodecanoic acid, 1,2,3-propanetriyl ester (12.16%), 1H-1,3-Benzimidazole-1-acetonitrile, 2-(difluoromethyl)- (

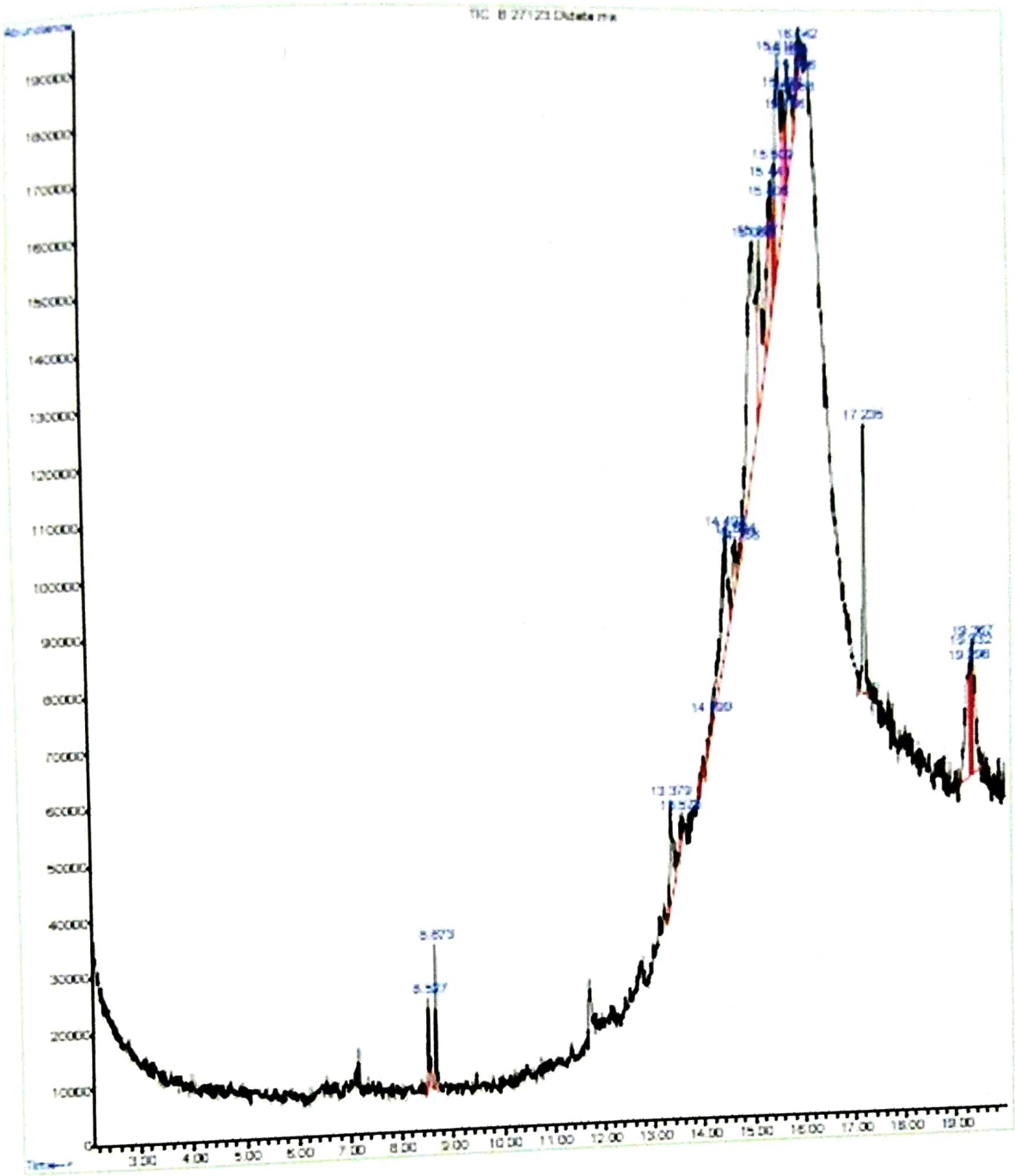
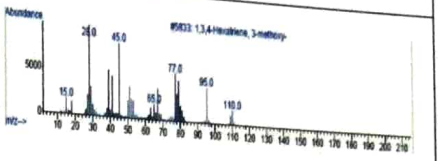
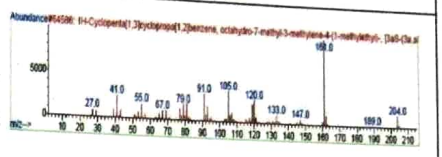
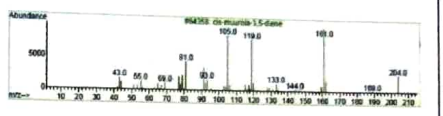
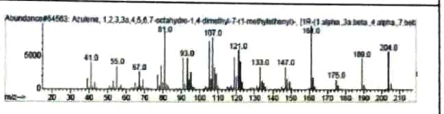
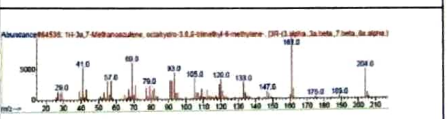
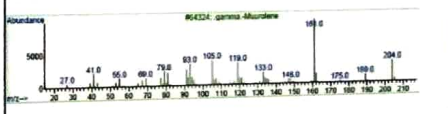
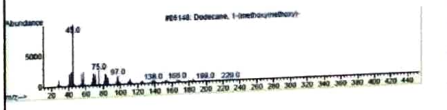
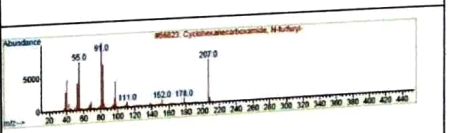
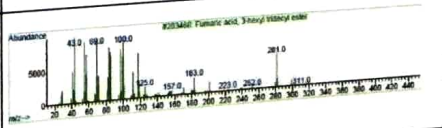
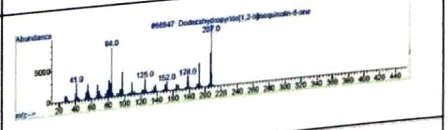
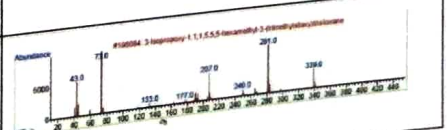
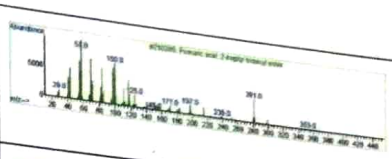
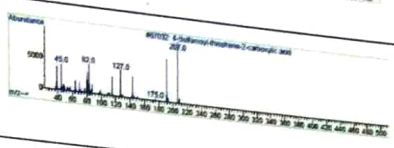
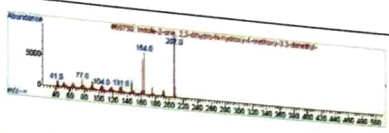
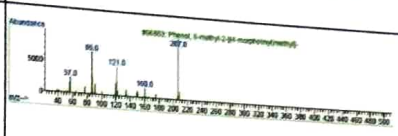
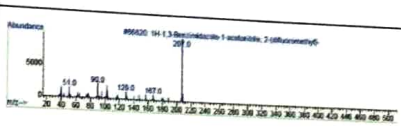
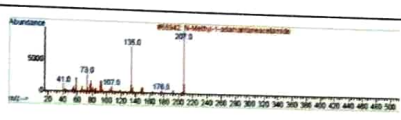
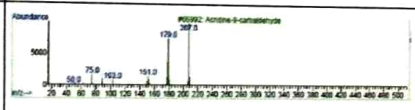
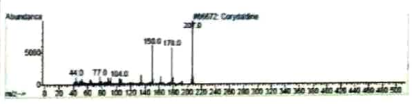
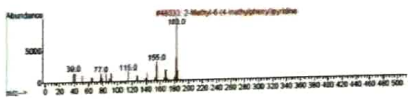
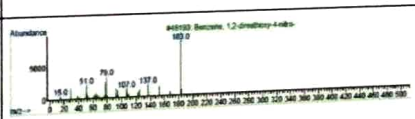
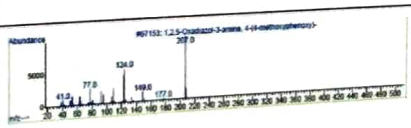
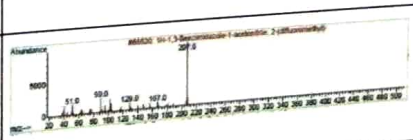
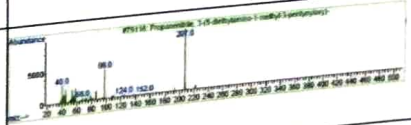
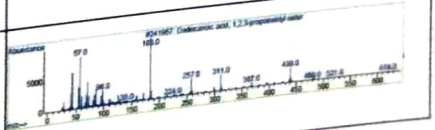
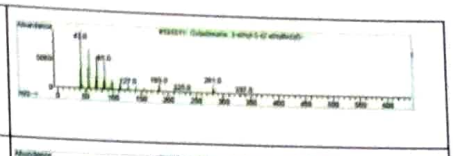
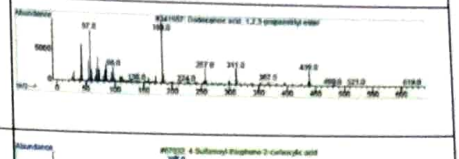
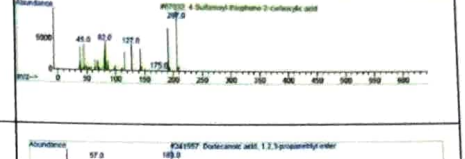
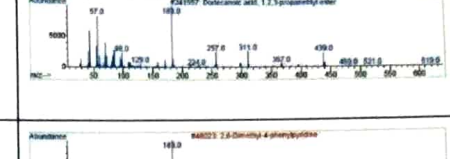
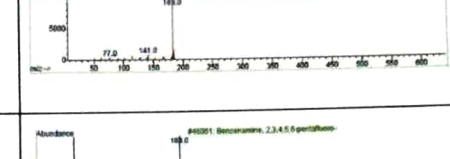
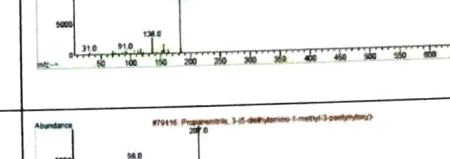
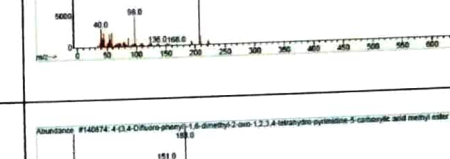
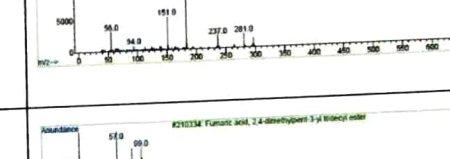
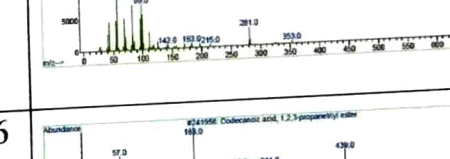
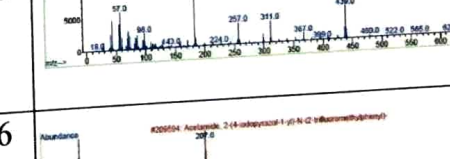
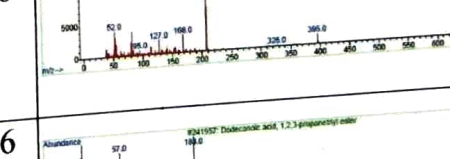
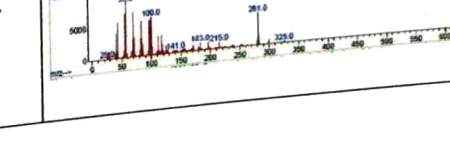


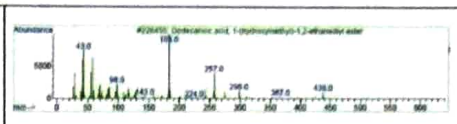
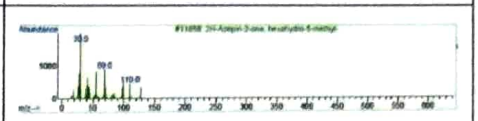
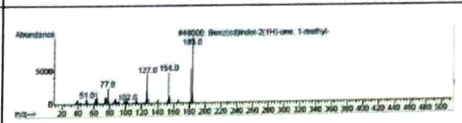
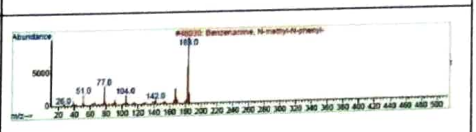
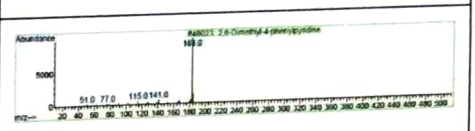
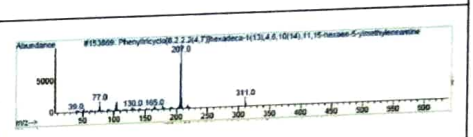
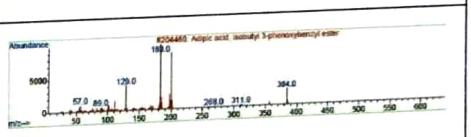
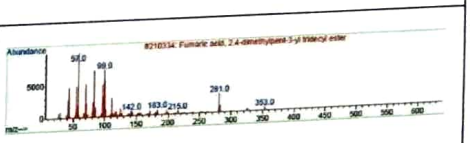
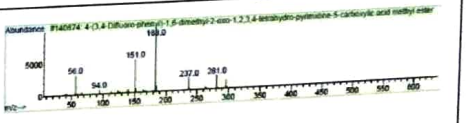
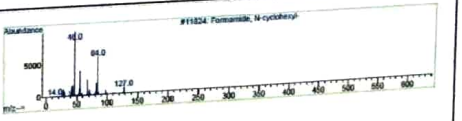
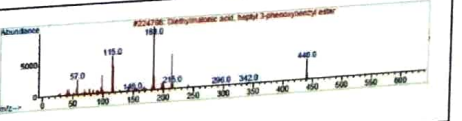
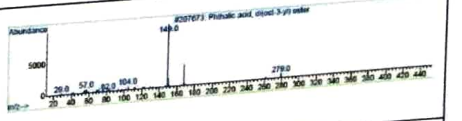
Figure 8: GC-MS Chromatogram of methanolic seed extract of *Syzygium cumini*

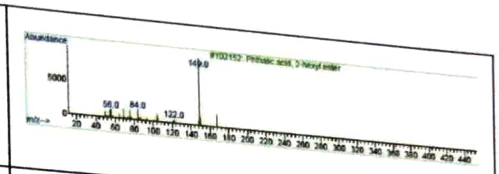
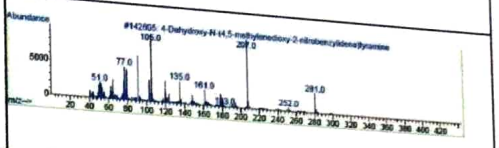
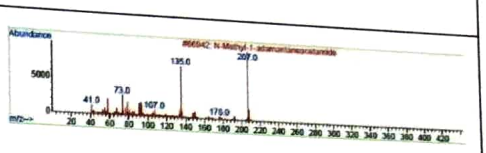
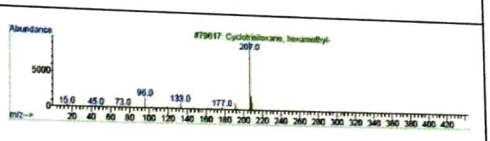
Table:10 Mass spectrum of *Syzygium cumini* seed

S.No		Name of the compound	Area %	Mass spectrum
1.	8.526	1,3,4-Hexatriene, 3-methoxy-	1.32	
2.	8.526	1H-Cyclopenta[1,3]cyclopropa[1,2]benzene, octahydro-7-methyl-3-methylene-4-(1-methylethyl)-, [3aS-(3a.al	1.32	
3.	8.526	cis-muurola-3,5-diene	1.32	
4.	8.677	Azulene, 1,2,3,3a,4,5,6,7-octahydro-1,4-dimethyl-7-(1-methylethenyl)-, [1R-(1.alpha.,3a.beta.,4.alpha.,7.beta	1.95	
5.	8.677	1H-3a,7-Methanoazulene, octahydro-3,8,8-trimethyl-6-methylene-, [3R-(3.alpha.,3a.beta.,7.beta.,8a.alpha.)]	1.95	
6.	8.677	gamma.-Muurolene	1.95	
7.	13.377	Dodecane, 1-(methoxymethoxy)-	3.66	
8.	13.377	Cyclohexanecarboxamide, N-furfuryl-	3.66	
9.	13.377	Fumaric acid, 3-hexyl tridecyl ester	3.66	
10.	13.575	Dodecahydropyrido[1,2-b]isoquinolin-6-one	0.70	
11.	13.575	3-Isopropoxy-1,1,1,5,5,5-hexamethyl-3-(trimethylsiloxy)trisiloxane	0.70	

12.	13.575	Fumaric acid, 2-heptyl tridecyl ester	0.70	
13.	14.199	4-Sulfamoyl-thiophene-2-carboxylic acid	0.72	
14.	14.199	Indole-2-one, 2,3-dihydro-N-hydroxy-4-methoxy-3,3-dimethyl-	0.72	
15.	14.199	Phenol, 6-methyl-2-[(4-morpholinyl)methyl]-	0.72	
16.	14.492	1H-1,3-Benzimidazole-1-acetonitrile, 2-(difluoromethyl)-	9.31	
17.	14.492	N-Methyl-1-adamantaneacetamide	9.31	
18.	14.492	Acridine-9-carbaldehyde	9.31	
19.	14.681	Corydaldine	2.19	
20.	14.681	2-Methyl-6-(4-methylphenyl)pyridine	2.19	
21.	14.681	Benzene, 1,2-dimethoxy-4-nitro-	2.19	
22.	14.766	1,2,5-Oxadiazol-3-amine, 4-(4-methoxyphenoxy)-	0.66	
23.	14.766	1H-1,3-Benzimidazole-1-acetonitrile, 2-(difluoromethyl)-	0.66	
24.	14.766	Propanenitrile, 3-(5-diethylamino-1-methyl-3-pentynyloxy)-	0.66	
25.	15.079	Dodecanoic acid, 1,2,3-propanetriyl ester	21.21	

26.	15.079	Octadecane, 3-ethyl-5-(2-ethylbutyl)-	21.21	
27.	15.201	Dodecanoic acid, 1,2,3-propanetriyl ester	7.34	
28.	15.201	4-Sulfamoyl-thiophene-2-carboxylic acid	7.34	
29.	15.409	Dodecanoic acid, 1,2,3-propanetriyl ester	4.18	
30.	15.409	2,6-Dimethyl-4-phenylpyridine	4.18	
31.	15.409	Benzenamine, 2,3,4,5,6-pentafluoro-	4.18	
32.	15.438	Propanenitrile,3(5-dithylamino-1-methyl-3-pentynnyloxy	3.27	
33.	15.513	4-(3,4-Difluoro-phenyl)-1,6-dimethyl-2-oxo-1,2,3,4-tetrahydro-pyrimidine-5-carboxylic acid methyl ester	1.59	
34.	15.513	Fumaric acid, 2,4-dimethylpent-3-yl tridecyl ester	1.59	
35.	15.617	Dodecanoic acid, 1,2,3-propanetriyl ester	12.16	
36.	15.617	Acetamide, 2-(4-iodopyrazol-1-yl)-N-(2-trifluoromethylphenyl)-	12.16	
37.	15.617	Dodecanoic acid, 1,2,3-propanetriyl ester	12.16	
38.	15.731	Fumaric acid, 4-heptyl tridecyl ester	2.54	

39.	15.731	Dodecanoic acid, 1-(hydroxymethyl)-1,2-ethanediyl ester	2.54	
40.	15.769	2H-Azepin-2-one, hexahydro-5-methyl-	1.41	
41.	15.835	Benz(cd)indol-2(1H)-one, 1-methyl-	5.03	
42.	15.835	Benzenamine, N-methyl-N-phenyl-	5.03	
43.	15.835	2,6-Dimethyl-4-phenylpyridine	5.03	
44.	15.958	Phenyltricyclo[8.2.2.2(4,7)]hexadeca-1(13),4,6,10(14),11,15-hexaen-5-ylmethyleneamine	0.23	
45.	15.958	Adipic acid, isobutyl 3-phenoxybenzyl ester	0.23	
46.	15.996	Fumaric acid, 2,4-dimethylpent-3-yl tridecyl ester	0.29	
47.	15.996	4-(3,4-Difluoro-phenyl)-1,6-dimethyl-2-oxo-1,2,3,4-tetrahydro-pyrimidine-5-carboxylic acid methyl ester	0.29	
48.	16.043	Formamide, N-cyclohexyl-	0.99	
49.	16.043	Diethylmalonic acid, heptyl 3-phenoxybenzyl ester	0.99	
50.	17.234	Phthalic acid, di(oct-3-yl) ester	6.52	
51.	17.234	2-(Decyloxycarbonyl)benzoic acid	6.52	

52.	17.234	Phthalic acid, 2-hexyl ester	6.52	
53.	19.296	4-Dehydroxy-N-(4,5-methylenedioxy-2-nitrobenzylidene)tyramine	5.48	
54.	19.296	N-Methyl-1-adamantaneacetamide	5.48	
55.	19.296	Cyclotrisiloxane, hexamethyl-	5.48	

LIST OF CHEMICAL COMPOUND IDENTIFIED FROM METHANOLIC SEED EXTRACTOOF SYZYGIUM CUMINI THROUGH GC-MS ANALYSIS

S.No	R.T	Name of the compound	Area %	Biological activity
1.	8.526	1,3,4-Hexatriene, 3-methoxy-	1.32	Antieczematic, Respiratory analeptic, Antiinflammatory, Antifungal
2.	8.526	1H-Cyclopenta[1,3]cyclopropa[1,2]benzene, octahydro-7-methyl-3-methylene-4-(1-methylethyl)-, [3aS-(3a.al	1.32	No activity found
3.	8.526	cis-muurola-3,5-diene	1.32	Antieczematic, Antinociceptive, Antiinfertility, Antiinflammatory, Antineurotic
4.	8.677	Azulene, 1,2,3,3a,4,5,6,7-octahydro-1,4-dimethyl-7-(1-methylethenyl)-, [1R-(1.alpha.,3a.beta.,4.alpha.,7.beta	1.95	Carminative, Antieczematic, Antineoplastic (thyroid cancer), Antifungal, Antipruritic
5.	8.677	1H-3a,7-Methanoazulene, octahydro-3,8,8-trimethyl-6-methylene-, [3R-(3.alpha.,3a.beta.,7.beta.,8a.alpha.)]	1.95	Dermatologic, Antipsoriatic, Antiinflammatory, Antineoplastic
6.	8.677	gamma.-Muurolene	1.95	Antineoplastic, Antiinflammatory, Antipruritic, Antifungal, Antibacterial
7.	13.377	Dodecane, 1-(methoxymethoxy)-	3.66	Antineurotic, Antiinflammatory, Antiparasitic, Antiviral, Antipruritic
8.	13.377	Cyclohexanecarboxamide, N-furfuryl-	3.66	Antiviral, Antiinfective, HIV , Anti-Helicobacter pylori, Antineurogenic pain
9.	13.377	Fumaric acid, 3-hexyl tridecyl ester	3.66	Respiratory analeptic, Antiviral, Antisecretoric, Antipruritic, Antithrombotic
10.	13.575	Dodecahydropyrido[1,2-b]isoquinolin-6-one	0.70	Cardiovascular analeptic, Antineurotic, Antipsychotic, Antinociceptive, Antipruritic

11.	13.575	3-Isopropoxy-1,1,1,5,5,5-hexamethyl-3-(trimethylsiloxy)trisiloxane	0.70	Antineoplastic, HIV-1 reverse transcriptase, Antiinfective, Antiviral (HIV), Antiseborrheic
12.	13.575	Fumaric acid, 2-heptyl tridecyl ester	0.70	Antieczematic, Antisecretoric, Antihelmintic, Antiviral, Antiinflammatory
13.	14.199	4-Sulfamoyl-thiophene-2-carboxylic acid	0.72	Antiglaucomic, Antiinflammatory, Antidiabetic, Antiarthritic, Antianginal
14.	14.199	Indole-2-one, 2,3-dihydro-N-hydroxy-4-methoxy-3,3-dimethyl-	0.72	Antiseborrheic, Antidiabetic, Antiviral, Antinephritic, Antiinflammatory
15.	14.199	Phenol, 6-methyl-2-[(4-morpholinyl)methyl]-	0.72	Cardiovascular analeptic, Antiischemic, Antidyskinetic, Antiseborrheic, Antiulcerative
16.	14.492	1H-1,3-Benzimidazole-1-acetonitrile, 2-(difluoromethyl)-	9.31	Antineoplastic, Antiinflammatory, Antiinflammatory, Antineoplastic, Antiarthritic
17.	14.492	N-Methyl-1-adamantaneacetamide	9.31	Antiarthritic, Antiviral, Cardiovascular analeptic, Antiparkinsonian, Antimyopathies
18.	14.492	Acridine-9-carbaldehyde	9.31	Antineoplastic, Antituberculosic, Prostate cancer treatment, Antiseborrheic
19.	14.681	Corydaldine	2.19	Antidyskinetic, Antineurotic, Antiischemic, Antinociceptive
20.	14.681	2-Methyl-6-(4-methylphenyl)pyridine	2.19	Antiseborrheic, Antiviral, Antihelmintic, Antinociceptive, Antihypoxic
21.	14.681	Benzene, 1,2-dimethoxy-4-nitro-	2.19	Antiprotozoal, Antianginal, Antiviral, Antiseptic, Antimycobacterial
22.	14.766	1,2,5-Oxadiazol-3-amine, 4-(4-methoxyphenoxy)-	0.66	Antiischemic, Antineoplastic, Antiviral, Antiinflammatory, Antimyopathies

23.	14.766	1H-1,3-Benzimidazole-1-acetonitrile, 2-(difluoromethyl)-	0.66	Antineoplastic, Antiinflammatory, Antiinflammatory, Antineoplastic, Antiarthritic
24.	14.766	Propanenitrile, 3-(5-diethylamino-1-methyl-3-pentynyloxy)-	0.66	Antihypoxic, Antisecretoric, Antianginal, Antihypertensive, Cardiotonic
25.	15.079	Dodecanoic acid, 1,2,3-propanetriyl ester	21.21	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective, Antiviral
26.	15.079	Octadecane, 3-ethyl-5-(2-ethylbutyl)-	21.21	Antiseborrheic, Antieczematic, Antineurotic Anticonvulsant, Cardiovascular analeptic
27.	15.201	Dodecanoic acid, 1,2,3-propanetriyl ester	7.34	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective, Antiviral
2 8.	15.201	4-Sulfamoyl-thiophene-2-carboxylic acid	7.34	Antiglaucomic, Antiinflammatory, Antidiabetic, Antiarthritic, Antianginal
29.	15.409	Dodecanoic acid, 1,2,3-propanetriyl ester	4.18	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective, Antiviral
30.	15.409	2,6-Dimethyl-4-phenylpyridine	4.18	Antiseborrheic, Antiviral, Antineurotic, Antidyskinetic, Antinociceptive
31.	15.409	Benzenamine, 2,3,4,5,6-pentafluoro-	4.18	Antineurotic, Antineoplastic, Antiviral, Antiseborrheic, Cardiovascular analeptic
32.	15.438	Propanenitrile, 3-(5-diethylamino-1-methyl-3-pentynyloxy)-	3.27	Antihypoxic, Antisecretoric, Antianginal, Cardiotonic, Antiglaucomic
33.	15.513	4-(3,4-Difluoro-phenyl)-1,6-dimethyl-2-oxo-1,2,3,4-tetrahydro-pyrimidine-5-carboxylic acid methyl ester	1.59	Antiadrenergic, Antianginal, Antieczematic, Antieczematic atopic, Antihypertensive
34.	15.513	Fumaric acid, 2,4-dimethylpent-3-yl tridecyl ester	1.59	Antihypoxic, Antieczematic, Antiviral,

35.	15.617	Dodecanoic acid, 1,2,3-propanetriyl ester	12.16	Antipruritic, Antisecretoric
36.	15.617	Acetamide, 2-(4-iodopyrazol-1-yl)-N-(2-trifluoromethylphenyl)-	12.16	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective, Antiviral
37.	15.617	Dodecanoic acid, 1,2,3-propanetriyl ester	12.16	Antipsychotic, Antineoplastic, Antifibrinolytic, Antitreponemal, Antipyretic
38.	15.731	Fumaric acid, 4-heptyl tridecyl ester	2.54	Antieczematic, Antihypoxic, Antithrombotic, Antiinfective, Antiviral
39.	15.731	Dodecanoic acid, 1-(hydroxymethyl)-1,2-ethanediyl ester	2.54	Antieczematic, Antifungal, Antihypercholesterolemic, Antiparasitic, Antiinflammatory
40.	15.769	2H-Azepin-2-one, hexahydro-5-methyl-	1.41	Antitoxic, Antipruritic, Anticonvulsant, Antiglaucomic, Antipyretic
41.	15.835	Benz(cd)indol-2(1H)-one, 1-methyl-	5.03	Cardiovascular analeptic, Antieczematic, Antidyskinetic, Antiviral, Antithrombotic
42.	15.835	Benzenamine, N-methyl-N-phenyl-	5.03	Antinociceptive, Antisecretoric, Antialcoholic, Antialcoholic, Anxiolytic
43.	15.835	2,6-Dimethyl-4-phenylpyridine	5.03	Antiseborrheic, Antiviral, Antipsoriatic, Cardiovascular analeptic, Antihypoxic
44.	15.958	Phenyltricyclo[8.2.2.2(4,7)]hexadeca-1(13),4,6,10(14),11,15-hexaen-5-ylmethyleneamine	0.23	Antiseborrheic, Antiviral, Antineurotic, Antidyskinetic, Antinociceptive
45.	15.958	Adipic acid, isobutyl 3-phenoxybenzyl ester	0.23	Antituberculosic, Antiseborrheic, Antimycobacterial, Antiinflammatory, Antineoplastic
				Antieczematic, Antihypercholesterolemic, Antipruritic, Antihypoxic, Antimetastatic

46.	15.996	Fumaric acid, 2,4-dimethylpent-3-yl tridecyl ester	0.29	Antihypoxic, Antieczematic, Antiviral, Antipruritic, Antisecretoric
47.	15.996	4-(3,4-Difluoro-phenyl)-1,6-dimethyl-2-oxo-1,2,3,4-tetrahydro-pyrimidine-5-carboxylic acid methyl ester	0.29	Antiadrenergic, Antiadrenergic, Antiviral, Antiamyloidogenic, Anticonvulsant
48.	16.043	Formamide, N-cyclohexyl-	0.99	Antibacterial, Antibacterial, Antibacterial, Antimycobacterial, Cardioprotectant
49.	16.043	Diethylmalonic acid, heptyl 3-phenoxybenzyl ester	0.99	Antihypercholesterolemic , Antiviral, Antiseptic, Antihelmintic, Antipruritic
50.	17.234	Phthalic acid, di(oct-3-yl) ester	6.52	Antiseborrheic, Antiviral, Antiischemic, Antisecretoric, Antifungal
51.	17.234	2-(Decyloxycarbonyl)benzoic acid	6.52	Antiseborrheic, Antihypercholesterolemic , Antiseptic, Antisecretoric, Antihypoxic
52.	17.234	Phthalic acid, 2-hexyl ester	6.52	Antiviral , Carnitinamidase, Antiseborrheic, Antiinfective, Antihypoxic
53.	19.296	4-Dehydroxy-N-(4,5-methylenedioxy-2-nitrobenzylidene)tyramine	5.48	Antineoplastic, Antihypertensive, Antidyskinetic, Antiparasitic, Antiseptic
54.	19.296	N-Methyl-1-adamantaneacetamide	5.48	Antiarthritic, Antiviral, Cardiovascular analeptic, Antimyopathies, Antiperistaltic
55.	19.296	Cyclotrisiloxane, hexamethyl-	5.48	Antineoplastic, Antiseborrheic, Antimyopathies, Antiviral, Antiinflammatory

9.31%), N-Methyl-1-adamantaneacetamide (9.31%), Acridine-9-carbaldehyde (9.31%), Dodecanoic acid, 1,2,3-propanetriyl ester (7.34%), 4-Sulfamoyl-thiophene-2-carboxylic acid (7.34%), Phthalic acid, di(oct-3-yl) ester (6.52%), 2-(Decyloxycarbonyl)benzoic acid (6.52%), Phthalic acid, 2-hexyl ester (6.52%).

The 42 minor compounds such as Phenyltricyclo [8.2.2.2 (4,7)] hexadeca-1(13),4,6,10(14),11,15-hexaen-5-ylmethyleneamine (0.23%),Adipic acid, isobutyl 3-phenoxybenzyl ester (0.23%) , Fumaric acid, 2,4-dimethylpent-3-yl tridecyl ester (0.29%), 4-(3,4-Difluoro-phenyl)-1,6-dimethyl-2-oxo-1,2,3,4-tetrahydro-pyrimidine-5-carboxylic acid methyl ester (0.29%), 1,2,5-Oxadiazol-3-amine, 4-(4-methoxyphenoxy)- (0.66%), 1H-1,3-Benzimidazole-1-acetonitrile, 2-(difluoromethyl)- (0.66%), Propanenitrile, 3-(5-diethylamino-1-methyl-3-pentynyloxy)- (0.66%), Dodecahydropyrido[1,2-b]isoquinolin-6-one (0.70%),3-Isopropoxy-1,1,1,5,5,5-hexamethyl-3-(trimethylsiloxy)trisiloxane (0.70%), Fumaric acid, 2-heptyl tridecyl ester (0.70%), 4-Sulfamoyl-thiophene-2-carboxylic acid (0.72%), Indole-2-one, 2,3-dihydro-N-hydroxy-4-methoxy-3,3-dimethyl- (0.72%), Phenol, 6-methyl-2-[(4-morpholinyl)methyl]- (0.72%), Formamide, N-cyclohexyl- (0.99%), Diethylmalonic acid, heptyl 3-phenoxybenzyl ester (0.99%), 1,3,4-Hexatriene, 3-methoxy- (1.32%), 1H-Cyclopenta[1,3]cyclopropa[1,2]benzene, octahydro-7-methyl-3-methylene-4-(1-methylethyl)-, [3aS-(3a.al (1.32%),cis-muurola-3,5-diene (1.32%), 2H-Azepin-2-one, hexahydro-5-methyl- (1.41%),4-(3,4-Difluoro-phenyl)-1,6-dimethyl-2-oxo-1,2,3,4-tetrahydro-pyrimidine-5-carboxylic acid methyl ester (1.59%), Fumaric acid, 2,4-dimethylpent-3-yl tridecyl ester (1.59%), Azulene, 1,2,3,3a,4,5,6,7-octahydro-1,4-dimethyl-7-(1-methylethenyl)-, [1R-(1.alpha., 3a.beta., 4.alpha.,7.beta (1.95%), 1H-3a,7-Methanoazulene, octahydro-3,8,8-trimethyl-6-methylene-, [3R-(3.alpha.,3a.beta.,7.beta.,8a.alpha.)] (1.95%), gamma.-Muurolene (1.95%), Corydaldine (2.19%), 2-Methyl-6-(4-methylphenyl)pyridine (2.19%), Benzene, 1,2-dimethoxy-4-nitro-

(2.19%), Fumaric acid, 4-heptyl tridecyl ester (2.54%), Dodecanoic acid, 1-(hydroxymethyl)-1,2-ethanediyl ester (2.54%), Propanenitrile, 3-(5-diethylamino-1-methyl-3-pentynyloxy)- (3.27%), Dodecane, 1-(methoxymethoxy)- (3.66%), Cyclohexanecarboxamide, N-furfuryl- (3.66%), Fumaric acid, 3-hexyl tridecyl ester (3.66%), Dodecanoic acid, 1,2,3-propanetriyl ester (4.18%), 2,6-Dimethyl-4-phenylpyridine (4.18%), Benzenamine, 2,3,4,5,6-pentafluoro- (4.18%) Benz(cd)indol-2(1H)-one, 1-methyl- (5.03%), Benzenamine, N-methyl-N-phenyl- (5.03%), 2,6-Dimethyl-4-phenylpyridine (5.03%), 4-Dehydroxy-N-(4,5-methylenedioxy-2-nitrobenzylidene)tyramine (5.48%), N-Methyl-1-adamantaneacetamide (5.48%), Cyclotrisiloxane, hexamethyl- (5.48%)were also reported from the seed extract of *syzygium cumini*. The chemical constituents analysis results of *Syzygium cumin*i seed were reported in Table 8 and their GC-MS chromatogram is presented in Fig : 8, Table-10

The first compound identified with less retention (8.526 min) was 1,3,4-Hexatriene, 3-methoxy- and 1H-Cyclopenta[1,3]cyclopropa[1,2] benzene, octahydro-7-methyl-3-methylene-4-(1-methylethyl)-, [3aS-(3a.al and cis-muurola-3,5-diene whereas 4-Dehydroxy-N-(4,5-methylenedioxy-2-nitrobenzylidene)tyramine and N-Methyl-1-adamantaneacetamide and Cyclotrisiloxane, hexamethyl- was the last compound which took longest retention time (19.296 min) to identify. At (15.079 min) Dodecanoic acid, 1,2,3-propanetriyl ester , Octadecane, 3-ethyl-5-(2-ethylbutyl)- was found to be high (21.21%) and the lowest percentage (0.23%) was found to be Phenyltricyclo [8.2.2.2 (4,7)] hexadeca-1(13),4,6,10(14),11,15-hexaen-5-ylmethyleneamine and Adipic acid, isobutyl 3-phenoxybenzyl ester. The above mentioned isolated compounds from the leaf and seed extract of *Syzygium cumini* have a medicinal importannce.

Quinolone, 2,4-dimethyl in the leaf extract of *Syzygium cumini* is a main antiviral compound (WWW.Pharmaexpert.ru/pass online predict.php). Quinolines are important compounds because of their bioactive properties and medicinal uses such as antimalarial

(Larsen *et al.*, 1996), anti-inflammatory (Chen *et al.*, 2001), antiasthmatic (Roma *et al.*, 2000), antibacterial (Dube *et al.*, 1998) and tyrosine kinase inhibiting agents (Billker *et al.*, 1998). Cyclotrisiloxane and hexamethyl found in the seed extract of *Syzygium cumini* are the main antioxidant compounds that help remove harmful toxins and free radicals in the body. (Anju Krishna *et al.*, 2015). The presence of the identified bioactive components present in leaf and seed extracts of *Syzygium cumini* could be responsible for the antioxidant and antimicrobial effects of the plants. Identification of these compounds in the plants that serve as the basis for determining the possible health benefits of the plant leads to further biological and pharmacological studies.

ANTIOXIDENT ACTIVITY

An antioxidant is a molecule capable of showing or preventing the oxidant of other molecules. In a biological system, they protect cells from the damage caused by unstable molecules known as free radicals. Antioxidants terminate the chain reaction by removing free radical intermediates, and inhibit other oxidant reactions by being oxidized themselves. They are believed to play a role in preventing the development of chronic diseases like cancer, heart diseases, stroke, AD, RA and cataracts (chakraborty *et al.*, 2010).

Antioxidant chemicals found in nature inhibit or prevent the oxidation of substrate leading to the formation of reactive oxygen species and reactive nitrogen species and thus protect the biological system (Hwang *et al.*, 2007).

Fruits and vegetables are endowed with antioxidants and consumption of these, prevent and protects from oxidative stress-related diseases, inflammatory diseases viz. , arthritis, autoimmune disease, carcinogenesis, neurodegenerative diseases, inflammatory diseases, cardiovascular disorders etc. several food industries use butylated hydroxyl, butylated hydroxynol toluene and tertiary butyl hydroquinone, the common synthetic antioxidants for preventing lipid oxidation in food products while processing and storage.

DPPH FREE RADICAL SCAVENGING ACTIVITY

Table: 11 Anti – oxidant activity in aqueous extract of *Syzygium cumini*

S. No	Aqueous Extract	DPPH free radical assay %
1	*Syzygium cumini* Seed	27.40
2	*Syzygium cumini* leave	21.30

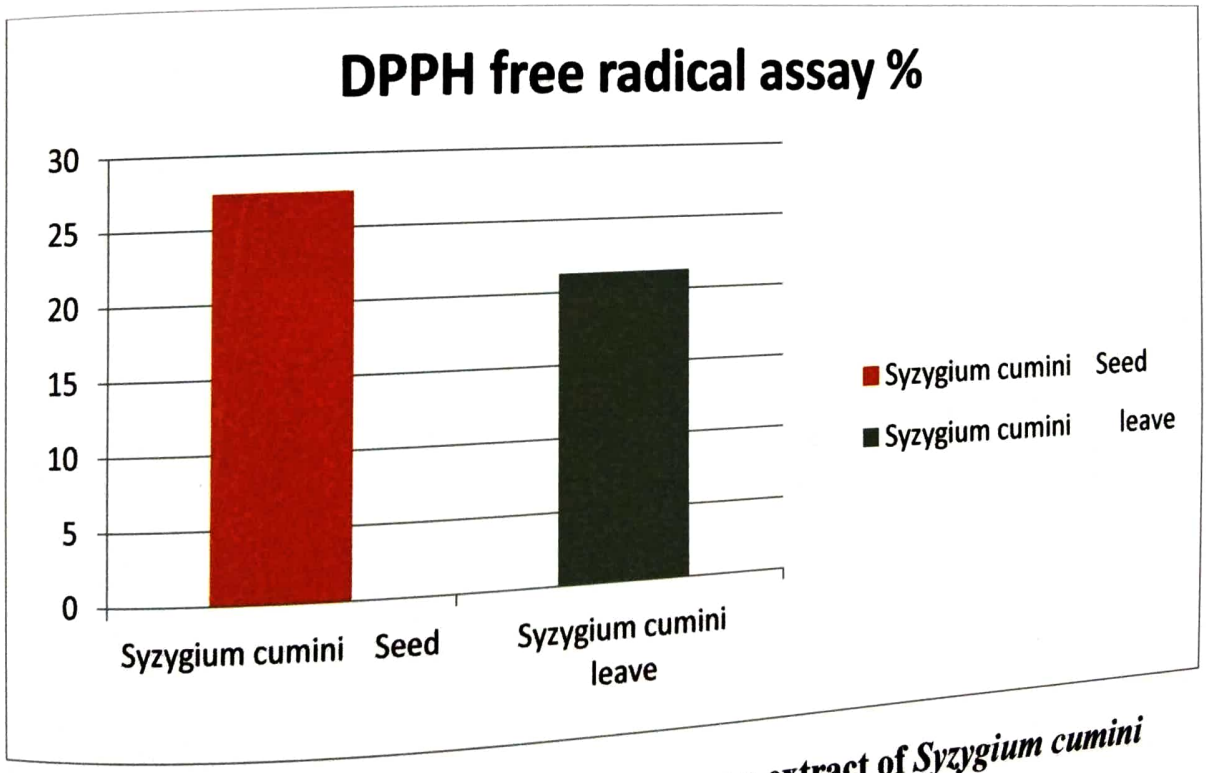


Figure: 9 Anti – oxidant activity in aqueous extract of *Syzygium cumini*

These synthetic antioxidants have been suspected to be carcinogenic and hence their use as food ingredients has been prohibited (hung and Wang, 2004). Natural antioxidants comprised non –detrimental decimal combinations are considered to be rather safer for use in food products. Further, uncared wastes if exploited as a resource of antioxidants, will be more beneficial to humankind and protect the environment. Flavonoids are water soluble polyphenolic molecules with antioxidant activity which has many beneficial effects on the cardiovascular system (Evans ,1989). Vitamin C acts as a ROS scavenger, thus potentially protecting cells from harmful oxidative products (Fossati *et al.*,). Vitamin E supplement elevates the activities of antioxidant enzymes(Kiron *et al.*, 2004).

DPPH FREE RADICAL SCAVENGING ACTIVITY

DPPH is a stable free radical and is widely used to assess the radical scavenging activity of a specific compound or plants extracts (Wei *et al.*, 2010) DPHH solution has a strong absorption band at 517nm appearing as a deep violet colour, The absorption vanishes and the resulting decolourization is stoichiometric with respect to the degree of reduction. The leaf and seed extract of *Syzygium cumini* was able to reduce stable DPPH radical to yellow colour diphenyl picryl hydrazine. The degree of reduction in absorbance is the reflection of the radical scavenging power of the compound.

The antioxidant activity of the aqueous leaf and seed extract of *Syzygium cumini* was evaluated by using a DPHH scavenging assay. Aqueous extract using *Syzygium cumini* leaf has higher scavenging activity (27.40%) followed by *Syzygium cumini* seed (21.30%), as shown in figure (9) and Table (11)

This result indicated aqueous leaf extract of the *Syzygium cumini* shows higher scavenging activities. It has been reported that leaf and seed extract of *Syzygium cumini* was due to the presence of phenolics and it is responsible for redox properties, which allow them

to act as reducing agents, hydrogen donors and singlet oxygen quenchers. (Arasali and kadimi 2009)

ANTIBACTERIAL ACTIVITY

In the present study, the antibacterial activity of different solvents (Hexane, acetone, ethanol, methanol and water) using leaf and seed of *Syzygium cumini* were tested against four human pathogenic bacteria (*Bacillus substilis*, *Escherichia coli*, *Staphylococcus sp,)* presented in table (12 & 13). The diameter of the inhibition zones against these species ranged from (1 to 14mm)

The different solvents (acetone, ethanol, methanol and water) extract of *Syzygium cumini* seed exhibited maximum activity against different bacterial species, *E. Coli* (3-13mm), *Bacillus substilis* (1-10mm), *staphylococcus sp* (3-14mm), .(plate:1),(Fig 10).

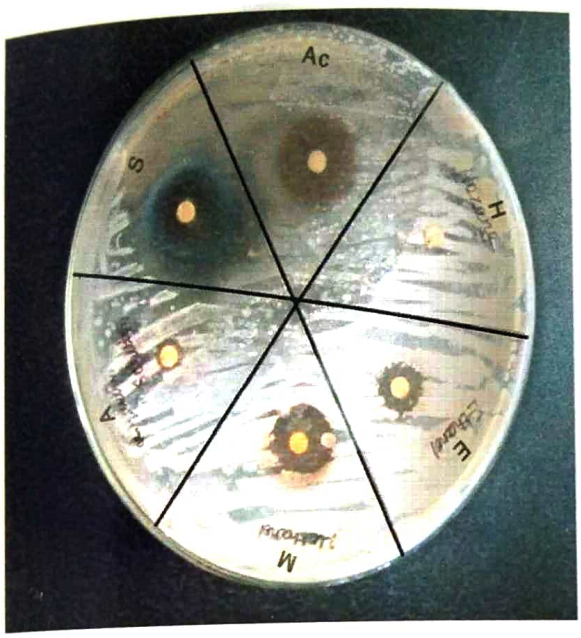
The different solvents (Hexane, acetone, ethanol and water) extracts of *Syzygium cumini* leaf exhibited maximum activity against different bacterial species, *E.coli* (3-10mm), *Bacillus substills* (4-11mm), *Staphylococcus sp* (1-11mm) inhibition zone .(plate:2), (Fig 11).

The maximum activity was found to be a 14mm zone of inhibition obtained by acetone seed extract of *Syzygium cumini* against *Staphylococcus sp*. The acetone extract of the seed of *Syzygium cumini* exhibited high antibacterial activity against *staphylococcus sp*, The diameter of the inhibition zone was 14mm. The acetone extract of *Syzygium cumini* seed exhibited more or less the same zone of inhibition compared to standard antibiotics streptomycin. The maximum bacterial effect was found in *Staphylococcus sp*, for acetone seed extracts of *Syzygium cumini.*

The maximum activity was found to be an 11mm zone of inhibition obtained by acetone leaf extract of *Syzygium cumini* against *Bacillus substilis*. The acetone extract of *Syzygium cumini* exhibited high antibacterial activity against *Bacillus substilis*, The diameter

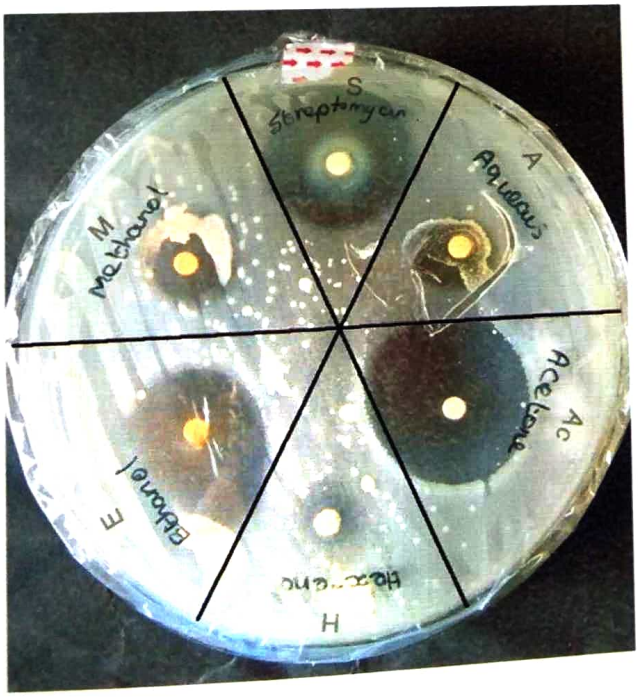
Plate- 1

Antibacterial activity of different solvent extracts of *Syzygium cumini* seed



Staphylococcus aureus

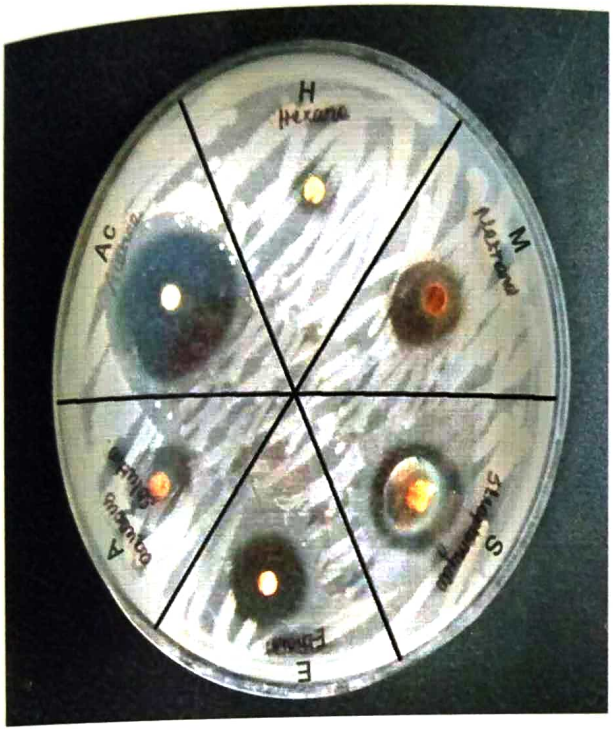
Bacillus subtilis



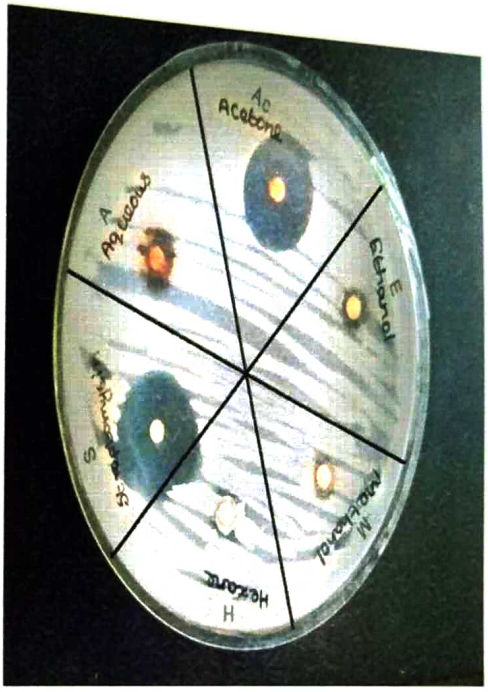
E.coli

Plate - 2

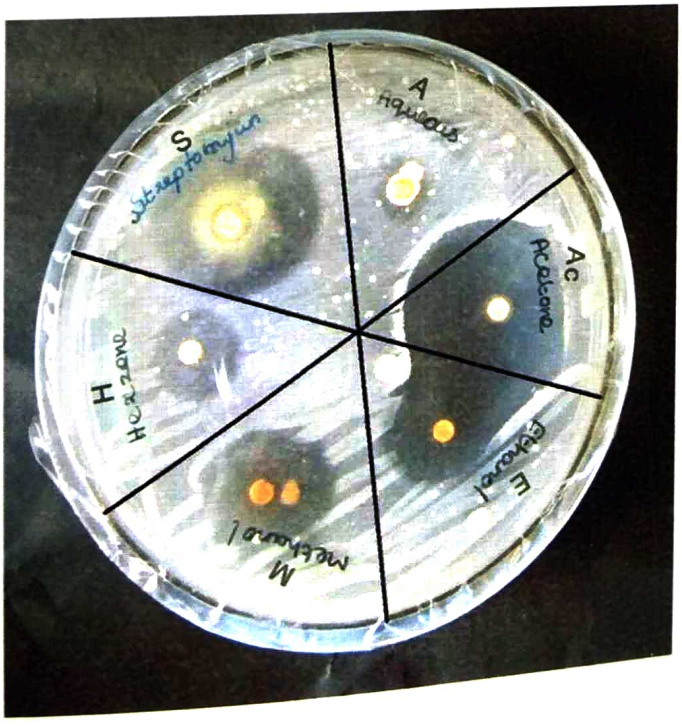
Antibacterial activity of different solvent extracts of *Syzygium cumini* leaf



Staphylococcus aureus



Bacillus subtilis



E.coli

Table - 12: Antibacterial activity of seed extract of *Syzygium cumini* with different solvents against human pathogen

samples	*Syzygium cumini* seed					
Microorganisms	Hexane	Ethanol	Methanol	Acetone	Aqueous	Streptomycin
Bacillus subtilis	1mm	5mm	8mm	10mm	3mm	15mm
Staphylococcus	5mm	5mm	3mm	14mm	8mm	14mm
Ecoli	3mm	8mm	7mm	13mm	3mm	13mm

Table - 13 : Antibacterial activity of leaf extract of *Syzygium cumini* with different solvents against human pathogen

Samples	*Syzygium cumini* leaf					
Microorganisms	Hexane	Ethanol	Methanol	Acetone	Aqueous	Streptomycin
Bacillus subtilis	2mm	8mm	8mm	11mm	4mm	13mm
Staphylococcus	1mm	7mm	4mm	11mm	4mm	12mm
Ecoli	3mm	5mm	4mm	10mm	1mm	11mm

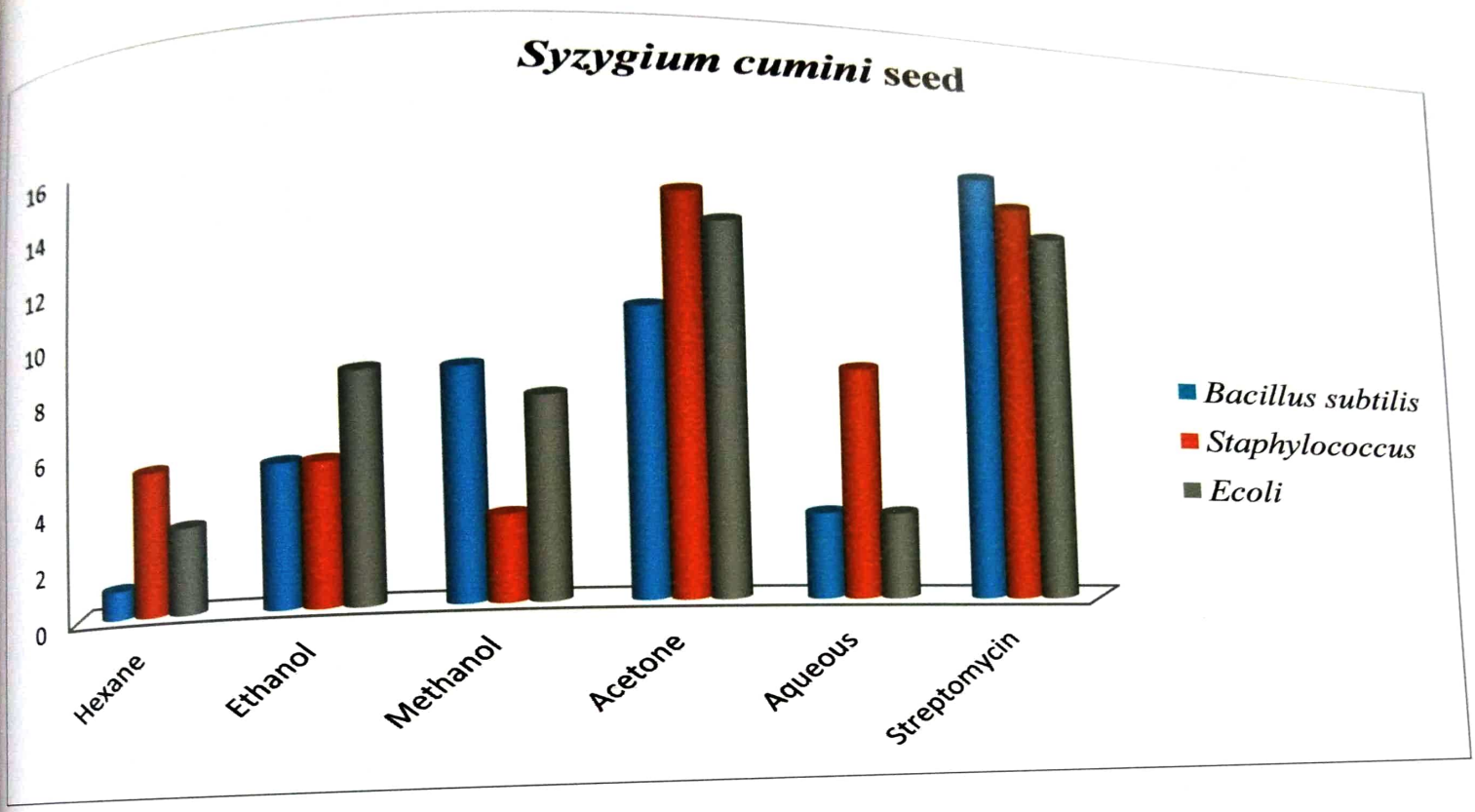


Figure: 10 Antibacterial activity of different solvent seed extracts of *Syzygium cumini* with against human pathogen

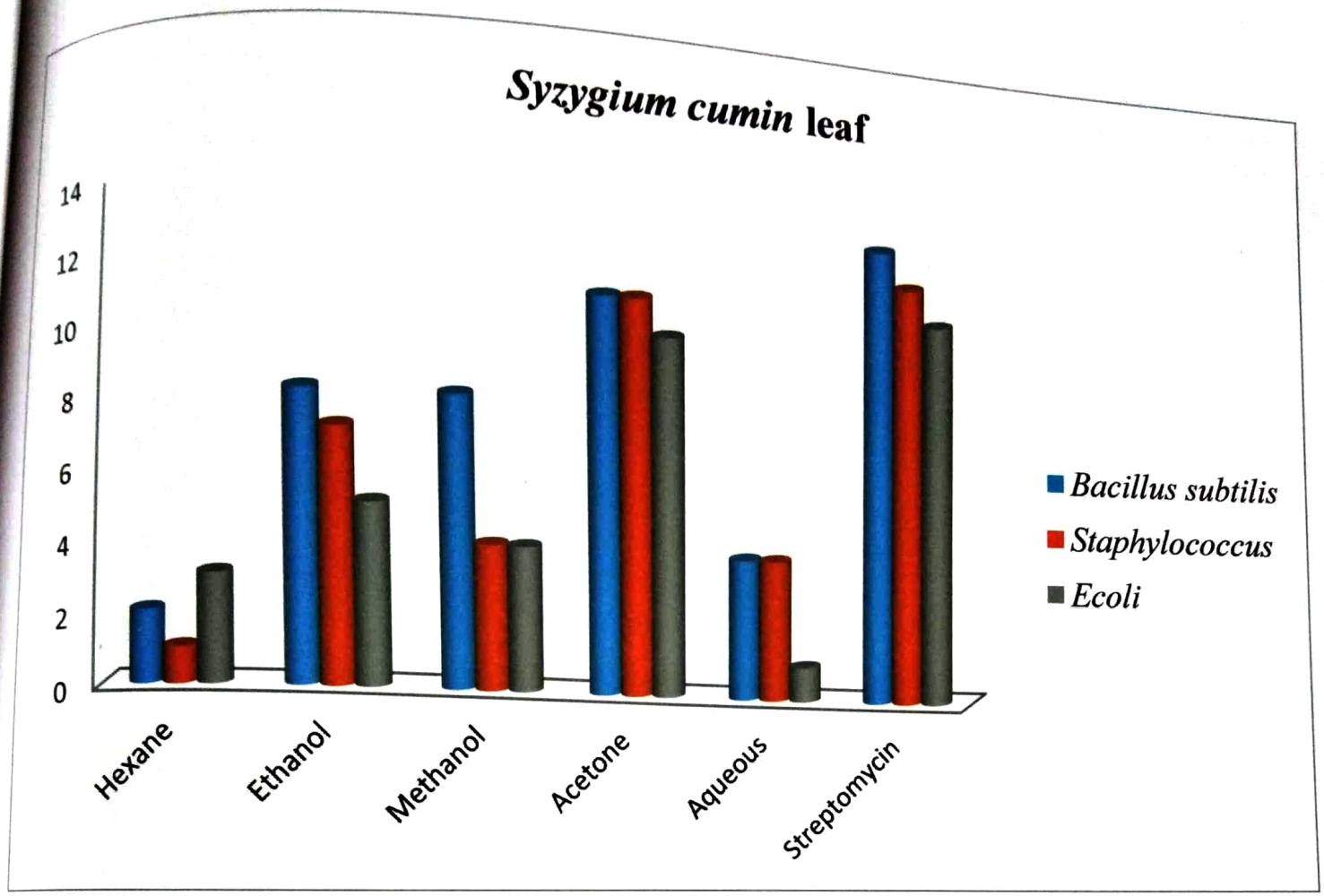


Figure: 11 Antibacterial activity of different solvent leaf extracts of *Syzygium cumini* with against human pathogen

of the inhibition zone was 11mm. The acetone extract of *Syzygium cumini* exhibited more or less the same zone of inhibition compared to standard antibiotics streptomycin..Maximum bacterial effects were found in *Bacillus substilis* for acetone extracts of *Syzygium cumini*

The antibacterial activity of seed and leaf extract of *Syzygium cumini* was nearly similar to streptomycin.. Maximum bacterial effects were found in *Staphylococcus sp* in *Syzygium cumini* seed and *Bacillus substilis* in *Syzygium cumini* leaf extract. The effects were significant in the seed and leaf of *Syzygium cumini*. The antibacterial activities of the seed and leaf of *Syzygium cumini* may be due to the presence of various phytochemicals which are known to be synthesized by plants in response to microbial infection (Cowan,1999). The mechanism of action of saponins as antimicrobial agents may be due to mebranolytic properties, rather than simply altering the surface tension of the extracellular medium(Killeen,1998). In our study seeds and leaves of *Syzygium cumini* showed extracellular saponins. The presence of tannins was also reported in seeds and leaves of *Syzygium cumini*

The antibacterial activity of tannins may be due to their intercalation with enzymes, cell envelope transport proteins and also complex with cell wall polysaccharides (Ya *et al.*,1998). Hence these plants stand as a potential candidate as a source of ingredients in drug formulation for the treatment of bacterial infection.

SUMMARY AND CONCLUSION

SUMMARY AND CONCLUSION

The plant kingdom is a treasure house of potential drugs used for the prevention and treatment of ailments. The plant *Syzygium cumini* was collected from Thoothukudi. Tamil Nadu, for the current study. *Syzygium cumini* was a known plant of the family Myrtaceae. All parts of the *Syzygium cumini* can be used medicinally and it has a long tradition in alternative medicine. From all over the world, the fruits have been used for a wide variety of ailments, including cough, diabetes, dysentery, inflammation and ringworm (Reynertson et al 2005). Various traditional practitioners in India use the different parts of the plant in the treatment of diabetes, blisters in the mouth, cancer, colic, diarrhea, digestive complaints, dysentery, piles, pimples and stomachache (Jain 1991,). In Unani medicine, various parts of jambolan act as a liver tonic, enrich the blood, strengthen teeth and gums and form a good lotion for removing ringworm infection of the head (Sagrawat et al 2006). The medicinal effects of plants are considered to be due to metabolites, especially secondary compounds, produced by plants. The phytochemical study revealed the presence of steroids, flavonoids, alkaloids, saponins, terpenoids, phenol and tannins. The preliminary phytochemical tests are helpful in finding chemical constituents in the plants materials that may lead to their quantitative estimation and also in locating the source of pharmacologically active chemical compounds. The information obtained from the preliminary phytochemical screening will be finding out the genuinity of the drug.

In this study, total phenol, flavonoid, tannin and vitamin C content were quantitatively analyzed in the leaf and seed of *Syzygium cumini* using spectrophotometric methods. The results of this study showed that the *Syzygium cumini* seed has a significant amount of phenol, flavonoids, tannins and vitamin C compared to *Syzygium cumini* leaf.

The FTIR spectroscopy analysis of *Syzygium cumini* seed obtained peaks at 522.67cm-1, 655.75 cm-1, 709.76 cm-1,761.83 cm-1,861.16 cm-1, 1081.99 cm-1, 1157.21 cm-1, 1456.16cm -1 1736.78cm-1,2360.71 cm-1,. These absorption peaks are known to be associated with the stretching vibration for C-Br in strong aromatic, N-H in strong primary amines, N-O in weak nitrate group, C-H in strong hydrogen atom,S-O in strong sulphinic acid group, C-O in very strong ring stretch, C-O-C in strong symmetric stretch, N=O in strong amines, C=O in strong esters, H-C-H in medium asymmetric stretch.

The FTIR spectroscopy analysis of *Syzygium cumini* leaf obtained peaks at 599.82cm-1, 656.72cm-1, 755.8cm-1, 1100.31cm-1, 1317.29cm-1, 1400.22cm1, 3163.04cm-1. These absorption peaks are known to be associated with the stretching vibration for C-Br in Aromatic, N-H in Amines, C-H in strong hydrogen atom, C-O-C in symmetric stretch, C-N in strong aryl tertiary amine, N=O in strong amines, O-H in medium.

From the spectral data presence of N=O, C-BR, N-O, C-O-C, C=O, N-H, N-O, , H-C-H, N=O, C-N and O-H were identified. These bonding are responsible for the presence of amines, Aromatic, nitrate group, symmetric stretch, Ester, amines and aryl tertiary amine Medium. The carboxylic acid present in the medicinal plant serves as the main pharmaceutical product in curing ulcers, jaundice, headache, stomatitis, hemicranias, fever, pain in the liver and treatment of rheumatic joint pain. Amides, amines and amino acids are the main groups, involved in protein synthesis. The study revealed that the seed and leaf of *Syzygium cumini* contain a considerable amount of secondary metabolites and it may be considered in the future to be used in human disease management.

The GC-MS analysis of the methanolic leaf extract of Syzygium cumini confirmed the presence of 20 compounds with retention time. Interpretation of the mass spectrum of GC-MS was conducted using the database of NIST and WILEY libraries. Out of these 20 compounds, 7 compounds are majorly present in the leaf extract of Syzygium cumini respectively Dodecanoic acid, 1,2,3-propanetriyl ester (31.38%), Dodecanoic acid, 1,2,3-propanetriyl ester (31.38%), 3,5-Dimethyl-4-phenylpyridine (31.38%), 1,2,3-propanetriyl ester (18.93%), Dodecanoic acid, 1,2,3-propanetriyl ester (13.49%), Quinoline-3-carbonitrile, 2-amino-4-methyl- (13.49%), Phen-1,2-diol, 4-fluoro-5-aminoacetyl-, dimethyl ether (13.49%).

The GC-MS analysis of methanolic seed extract of *syzygium cumini* confirmed the presence of 55 compounds with retention time. Interpretation of the mass spectrum of GC-MS was conducted using the database of NIST and WILEY libraries. Out of these 55 compounds 13 compounds were majority present in the seed extract of *syzygium cumini* respectively Dodecanoic acid, 1,2,3-propanetriyl ester (21.21%), Octadecane, 3-ethyl-5-(2-ethylbutyl)- (21.21%), Dodecanoic acid, 1,2,3-propanetriyl ester (12.16%), Acetamide, 2-(4-iodopyrazol-1-yl)-N-(2-trifluoromethylphenyl)- (12.16%), Dodecanoic acid, 1,2,3-propanetriyl ester (12.16%), 1H-1,3-Benzimidazole-1-acetonitrile, 2-(difluoromethyl)- (9.31%), N-Methyl-1-adamantaneacetamide (9.31%), Acridine-9-carbaldehyde (9.31%), Dodecanoic acid, 1,2,3-propanetriyl ester (7.34%), 4-Sulfamoyl-thiophene-2-carboxylic acid (7.34%), Phthalic acid, di(oct-3-yl) ester (6.52%), 2-(Decyloxycarbonyl)benzoic acid (6.52%), Phthalic acid, 2-hexyl ester (6.52%).

Quinolone, 2,4-dimethyl in the leaf extract of *Syzygium cumini* is a main antiviral compound (WWW.Pharmaexpert.ru/pass online predict.php). Quinolines are important compounds because of their bioactive properties and medicinal uses such as antimalarial (Larsen *et al.*, 1996), anti-inflammatory (Chen *et al.*, 2001), antiasthmatic

(Roma *et al.*, 2000), antibacterial (Dube *et al.*, 1998) and tyrosine kinase inhibiting agents (Billker *et al.*, 1998). Cyclotrisiloxane and hexamethyl found in the seed extract of *Syzygium cumini* are the main antioxidant compounds that help remove harmful toxins and free radicals in the body. (Anju Krishna *et al.*, 2015). The presence of the identified bioactive components present in leaf and seed extracts of *Syzygium cumini* could be responsible for the antioxidant and antimicrobial effects of the plants. Identification of these compounds in the plants that serve as the basis for determining the possible health benefits of the plant leads to further biological and pharmacological studies.

The antioxidant or free radical scavenging activity of plant extracts of these selected medicinal plants are investigated by using methods like DPPH scavenging activity. The leaf and seed extract of *Syzygium cumini* show maximum antioxidant activity and these extracts are further subjected for antimicrobial studies.

The different solvent extracts of leaf and seed of *Syzygium cumini* and streptomycin.were used for antibacterial studies against human pathogenic bacteria, *Bacillus substilis, Escherichia coli, Staphylococcus.*

The maximum activity was found to be a 14mm zone of inhibition obtained by acetone seed extract of *Syzygium cumini* against *Staphylococcus sp.* The acetone extract of the seed of *Syzygium cumini* exhibited high antibacterial activity against *staphylococcus sp,* The diameter of the inhibition zone was 14mm. The acetone extract of *Syzygium cumini* seed exhibited more or less the same zone of inhibition compared to standard antibiotics streptomycin. The maximum bacterial effect was found in *Staphylococcus sp,* for acetone seed extracts of *Syzygium cumini.*

The maximum activity was found to be an 11mm zone of inhibition obtained by acetone leaf extract of *Syzygium cumini* against *Bacillus substilis.* The acetone extract of *Syzygium cumini* exhibited high antibacterial activity against *Bacillus substilis,* The diameter

of the inhibition zone was 11mm. The acetone extract of *Syzygium cumini* exhibited more or less the same zone of inhibition compared to standard antibiotics streptomycin..Maximum bacterial effects were found in *Bacillus substilis* for acetone extracts of *Syzygium cumini*

The antibacterial activity of seed and leaf extract of *Syzygium cumini* was nearly similar to streptomycin.. Maximum bacterial effects were found in *Staphylococcus sp* in *Syzygium cumini* seed and *Bacillus substilis* in *Syzygium cumini* leaf extract. The effects were significant in the seed and leaf of *Syzygium cumini*. The antibacterial activities of the seed and leaf of *Syzygium cumini* may be due to the presence of various phytochemicals which are known to be synthesized by plants in response to microbial infection (Cowan,1999). The mechanism of action of saponins as antimicrobial agents may be due to mebranolytic properties, rather than simply altering the surface tension of the extracellular medium(Killeen,1998). In our study seeds and leaves of *Syzygium cumini* showed extracellular saponins. The presence of tannins was also reported in seeds and leaves of *Syzygium cumini*

The antibacterial activity of tannins may be due to their intercalation with enzymes, cell envelope transport proteins and also complex with cell wall polysaccharides (Ya *et al.*,1998). Hence this plant stands as a potential candidate as a source of ingredients in drug formulation for the treatment of bacterial infection.

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COMPARITIVE STUDY ON THE PHYTOCHEMICAL SCREENING, MINERAL COMPOSITION, ANTIOXIDANT AND ANTIBACTERIAL ACTIVITY OF SOME SELECTED EDIBLE SEEDS

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By

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2022–2023

CERTIFICATE

This is to certify that this dissertation entitled **"COMPARITIVE STUDY ON THE PHYTOCHEMICAL SCREENING, MINERAL COMPOSITION, ANTIOXIDANT AND ANTIBACTERIAL ACTIVITY OF SOME SELECTED EDIBLE SEEDS"** submitted by S.MARIYA ROSELINE ANUSHIYA Reg. No. 21APBO10 to **ST. MARY'S COLLEGE (Autonomous), THOOTHUKUDI** in partial fulfilment for the award of the degree of **"Master of Science in Botany" is done by her under my supervision.** It is further certified that this dissertation or any part of this has not been submitted elsewhere for any other degree.

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DECLARATION

I do hereby declare that this work has been originally carried out by me under the guidance and supervision of **Dr. P. Hermalin, M.Sc., M. Phil., Ph.D**, Assistant Professor of Botany, St. Mary's College (Autonomous), Thoothukudi and this work has not been submitted elsewhere for the award of any other degree.

S. Mariya Roselin Anushiya.

Place: Thoothukudi **Signature of the candidate**

Date: 12.4.23 (S.MARIYA ROSELIN ANUSHIYA)

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CONTENT

INTRODUCTION

Plants provide a wide range of resources that contribute to the fundamental needs of both human beings and animals such as food, clothing, and shelter. Most of the economically important plants possess medicinal values. Plants have been utilized as therapeutic agents since ancient times. The healing properties of many herbal medicines have been recognized in many ancient cultures e.g. Chinese, Ayurvedic, and Egyptian (Sarker & Nahar, 2007).It is well acknowledged that India is endowed with an incredible diversity of medicinal plants.In recent decades, about 3.4 billion people aredependent on plant-based traditional medicines with about 88 percent of the world'sinhabitants relying mainly on traditional medicine for their primary health care.Most of the plant parts such as leaves, roots, rhizomes, stems, barks, flowers, fruits, grains, or seeds, reserve high medicinal value. Among these, seeds are considered to be one of the most nutritious human dietary foods, due to their high contents of proteins, carbohydrates, unsaturated fatty acids, vitamins, and essential minerals.Seeds consumption lowers the risk of cardiovascular heart disease (CHD), which may be partly explained by the cholesterol-lowering effect.

India is one of the largest producers and exporters of fruits in the world. India is well-known for its cultivation of rich fruit varieties with over many different species distributed throughout the country. Different varieties of major and minor fruit species and tropical fruits are cultivated and grown in the country. However, many seeds include chia, hemp, sesame, pumpkin, sunflower, mustard, nigella, papaya, mangosteen, honeydew, pomegranate, fennel, fenugreek, cumin, sweet orange, cucumber, jackfruit, mango, melons, avocado is usually consumed by humans as a dietary food. Seeds are

dried products with low water content (Łoźna*et al.,* 2020). Owing to their evolutionary adaptation to the embryonic nutrition of the plants they originate from, seeds are rich in different nutrients, such as proteins, carbohydrates, and lipids (Gama, 2018). These seeds are also processed into different by-products which have food value and food flavors. Seeds have always been used forhuman health and to cure many diseases (Ramawat *et al.,* 2008). Sinceit contains essential bioactive components such as alkaloids, carotenoids, flavonoids, glycosides, saponins, terpenoids, tannins, steroids, and polyphenolic compounds and that exhibit excellent anti-inflammatory, antioxidant properties, anticancer, anti-diabetic, anti-hyperlipidemic, anti-obesity, neurological disorders, cardiovascular, skin diseases, and chronic diseases. Many researchers have found its role in the fight against several diseases such as cancer, atherosclerosis, cerebral cardiovascular events, diabetes, hypertension, and Alzheimer's disease (Liu *et al.,* 2003; Devasagayam *et al.,* 2004).In addition, seeds are also good sources of different bioactive compounds, such as carotenoids (vitamin A), tocopherols (vitamin E), xanthophylls, and polyphenols (Lorenzo*et al.,* 2019). Indeed, phenolic compounds such as phenolic acids, flavonoids, stilbenes, and lignans are strong antioxidant compounds (Pastoriza*et al*., 2016). Polyphenols compounds are the subject of increasing scientific interest due to their potential applicability in the treatment of some chronic diseases, such as cardiovascular diseases, diabetes, osteoporosis, or neurodegenerative disorders (Pandey and Rizvi, 2009).

Polyphenol compounds act as efficient free radicals and reactive oxygen species (ROS). This reactive oxygen species acts in damaging cell membranes, attacking proteins and DNA in tissues. Carcinogenesis may also be initiated through oxidatively induced

DNA damage. Repeated damage caused by ROS throughout the span of human life increases with time and is a major cause of age-related cancers and other oxidatively induced diseases (Reynertson, 2007). Antioxidants are substances that when present in foods or the body at low concentrations compared with that of an oxidizable substrate significantly delay or prevent the oxidation of that substrate (Saha *et al.,* 2004). It will help to minimize oxidative damage as the most important approach to the primary prevention of age-related diseases since antioxidants terminate direct ROS and radical-mediated oxidative reactions (Tepe and Sokmen, 2007).

Therefore, dietary antioxidants are needed to protect against the harmful action of ROS. Well-established antioxidants derived from the diet are vitamins A, C, and E, polyphenols, and carotenoids (Pietta, 2000). Present antioxidant research of free radicals also has confirmed that food with rich antioxidants plays an essential role in the prevention of diseases caused by oxidative stress. Therefore, plant-derived antioxidants are now receivingspecial attention in recent decades. With the intention of, the present work has planned to work on the following objectives.

Scope and Objectives

- Collection of various types of edible seeds and prepare their extracts.
- To qualitatively analyze and compare the presence of their bioactive compounds.
- To quantitatively analyze their vitamins C and E using spectroscopic methods.
- To assess the antioxidant potential of selected edible seeds using aqueous extract against DPPH radical scavenging activity.
- To identify and compare their mineral composition

REVIEW OF LITERATURE

In the last decade, a growing interest in seeds as significant ingredients of the daily diet has been observed, since seeds are placed next to legumes as a source of plant proteins (Łoźna*et al.,* 2020). In addition, seeds contribute to meeting the increasing food demand, and in many cases, are also used as traditional medicines (Caballero *et al.,*2006, Preedy*et al*., 2011). Even more, their seed cakes are used for animal feed and as green manures in organic agriculture (Lokanadhan *et al.,* 2012).Seeds are dried products with low water content ((Łoźna*et al.,* 2020). Owing to their evolutionary adaptation to the embryonic nutrition of the plants they originate from, seeds are rich in different nutrients, such as proteins, carbohydrates, and lipids (Gama, 2018). In addition, seeds are also good sources of different bioactive compounds, such as carotenoids (vitamin A), tocopherols (vitamin E), xanthophylls, and polyphenols (Lorenzo *et al.,* 2019 and Panfili *et al.,* 2020). Indeed, phenolic compounds such as phenolic acids, flavonoids, stilbenes, and lignans are strong antioxidant compounds (Pastoriza*et al*., 2016).

Secondary seeds metabolites perform vital roles in human health and could possibly be nutritionally crucial. Phytochemical screening processes associated with nutritional seeds revealed the existence of many substances which include alkaloids, tannins, flavonoids, phenol, terpenoids, steroids, glycosides, saponins, etc. Numerous seeds extract components as well as phytochemicals present antioxidant / free radical scavenging properties. Secondary metabolites associated with medicinal plants provide a protection mechanism against predation through quite a number of microbes, insects as well as herbivores (Anjum *et al.,* 2012)

Polyphenols compounds are the subject of increasing scientific interest due to their potential applicability in the treatment of some chronic diseases, such as cardiovascular diseases, diabetes, osteoporosis, or neurodegenerative disorders (Pandey andRizvi, 2009). Moreover, phenolic compounds can be found in the coats (hull, husk, or skin, for instance) covering the cotyledon(s) of seeds(Arjeh*et al.*, 2020) and can be even separated from the seed matrix by extraction with an appropriate solvent (Mojzer*et al.*, 2016).Phenolic present in seeds provid eprotection again stharm fulfree radical sand have been known to reduce the risk of certain types of diseases such as cancer, coronary heart diseases (CHDs), cardiovascular diseases (CVDs), stroke, atherosclerosis, osteoporosis, inflammation and other neurodegenerative diseases associated with oxidatives tress. Plant phenolic shave multiple biological functions such as antioxidant, anti-inflammatory, anti-cancer, and anti-microbial activities (John and Grohmann,2001).

On the other hand, seeds are important dietary sources of minerals since they accumulate these compounds during plant growth to be used for further development needs (Freitas, 2012). The content of nutrients varies depending on plant variety, agricultural practices, soil and climatic conditions, as well as technological and culinary practices(Łoźna *et al.,* 2020). Mineral elements such as Ca, Co, Cr, Cu, Fe, K, Mg, Mn, Mo, Na, P, S, Se, Zn, etc. have a relevant role in human health (Ferranti, 2019).For example, seeds are low in Na and rich in K, and clinical trials and meta-analyses suggest that a high intake of potassium is linked with blood pressure reduction(Danuko *et al.,* 2017). In the case of seeds, the mineral contents of usual seeds are known (Gouveia*et al.,* 2014) but, for instance, the potential of seeds from herbal plants has received less attention from the scientific community.

Cucumber (*Cucumis sativus* L.) is a member of the important vegetables which belong to the family Cucurbitaceae like gourds, melon, pumpkins, and squash. It is widely used as medicine in traditional Indian medical practices and very much liked as a vegetable. Cucumber fruit consists of more than ninety percent of water, offers superior hydration, and is very low in calories as a food. Its flavor and texture have made it essential as a fresh addition to salads and in processed forms such as pickles and relishes. It exhibits various medicinal properties like antimicrobial activity, glycemic lowering ability, antioxidant ability, etc., and is traditionally used in various treatments. It is believed that its regular intake or application on the skin helps in reducing the aging effect, boosting metabolism, and improving immunity(Sharma *etal.,* 2020).

Sunflower plant (*Helianthus annuus)*, a leading oil seed crop that is cultivated primarily for its seeds, ranks second for edible oil production globally after soybean oil (Robertson and Burns, 1975). It is native to the Middle American region later being commercially availableat the global level (Anjum *et al.,* 2012). Sunflower is cultivated globally for its oil and protein content predominantly. Proteins present in sunflower seeds have favorable amino acid distribution. In addition to this, tocopherols, minerals, and vitamins are provided by sunflower seeds in substantial amounts(Skoric, 2009). Half a cup (64gm) of dry roasted sunflower seeds provide 370 kcal energy, 7gm of dietary fibers, and 12 gm of proteins Among vitamins, 17mg of vitamin E, 4.5gm of niacin, 0.5mg of pyridoxine,4.5mg of pantothenic acid and 151mcg of folic acid are present in 64gm of dry roasted sunflower seeds. Sunflowers seeds are also rich in minerals like calcium, copper, iron, magnesium, manganese, selenium, phosphorous, potassium, sodium, and zinc. 128 gm of sunflower seeds contains 89.6 mg calcium, 4.9

mg iron, 165 mg magnesium, 1478 mg phosphorus, 1088 mg potassium, 3.8 mg sodium, 6.8mg zinc, 2.3 mg copper, 2.7 manganese mg and 102 mcg selenium as given by USDA (2008).(Anjum *et al.,* 2012).

Sunflower seeds provide a rather significant source of zinc, a mineral that helps boost the immune system. Folate in sunflower seeds facilitates the formation of RNA, DNA, and hemoglobin. Tryptophan and choline present in sunflower seeds are useful in reducing stress, anxiety and depression, and memory enhancement (Sharma *etal.,* 2020).Sunflower seeds because of being low in sodium and saturated fats and high in magnesium, potassium, and fiber are included by the U.S. National Heart, Lung and Blood Institute in the DASH eating plan (Anjum *et al.,* 2012). Due to the presence of a wide variety of nutritional components, Sunflower seeds possess wide therapeutic dimensions with multifaceted actions. This article is to give an overview ofa wide array of uses of sunflower seeds as anti-inflammatory, antibacterial, antifungal, anticancer, cardioprotective, and dermo protective in human beings and to enlighten the therapeutic potential of the cheap and easily available seed crop-Sunflower.

Flaxseeds, scientifically known as *Linumusitatissimum* L. (AminandThakur,2014) belong to the Linaceae family (Ganorkar&Jain,2013). Inthefield of functional foods ,flax seed is emergingasone of the key sources of phytochemicals (Shahzad *et al.,* 2006). These phytochemicals (phenolic acids, cinnamicacids, flavonoids, andlignins) are antioxidants and affect cel lgrowth and viability (AminandThakur,2014). Flax seed is an essential source of high-quality protein and soluble fiber and has considerablepotentialasa source of phenolic compounds (Oomah,2001;Shahzad*et al.,*2006; AminandThakur,2014). Both soluble as well as insoluble fiber is present in flax seed with insoluble fiber

contributing to about one-third of the total fiber content.Czemplik*et al., (*2011) sugges tthat the extract sderived from new genetically modified(GM)flax types might bean effective source of antibacterial compounds and apromisingalternative to antibiotictherapy. Czemplik*etal.,*(2011) evaluated the activity of GM seed cake extract against the bacteria of clinicalr elevance-Escherichiacoli, Bacillussubtilis, and Staphylococcus aureus which are known to be the cause of antibiotic-resistantin fections. They also determined the minimal inhibit ory concentration (MIC) and indicated the bacteriostaticor bactericidalaction. The flavonoids (kaempferolandquercetin) present in flax seed sinhibit DNA synthesis in *Proteusvulgaris* or RNA synthesis in *Staphylococcusaureus* (Mori*etal.,*1987). This mixture of flavonoids, phenolicacids, and lignansisproposedtobea more effective anti-bacterial agentthana pure, single compound. The extracts of GM seed cakes may be more beneficial than isolated constituents since a bioactive in dividual component can change its properties in the presence of other compounds in the extracts (Borchers*etal.,*2004).

L. usitatissimum is often called “functional food”, “bioactive Food” and “endocrine active food” and its nutrient value together with its protective properties originate from its distinct ingredients (İşleroğlu *et.al.,* 2005).Flaxseed has also been classified as a functional food because it provides numerous health benefits in addition to serving as a source of nutrients(Pansare*et.al.,*2020). Most of the health benefits of Flax seeds are because of their rich nutritional content. It contains omega-3 essential fatty acids, Lignans, and fibers. The plant has shown diverse biological and pharmacological activities like Antioxidant, immunomodulatory, anti-inflammatory, antimicrobial,

antiprotozoal, insecticidal, analgesic, anti-hyperlipidemia, anti-hyperglycemic, anti-tumor, wound healing, and Feticidal activities(Pansare*et.al.,*2020).

The seeds of pumpkin are a nutritional treasure as they are a robust source of good-quality fat, protein, fibers, antioxidants, and other phytochemical compounds. One ounce of pumpkin seeds contains almost twice asmuchironasthreeounces of skinless chickenbrea stand they provide more fiber perounce than nuts. They are also good sources ofprotein (Hornick and Yarnell,2013).The pumpkin (*Cucurbita* spp.) is one of the most nutritious vegetables consumed all over the world, and it has currently been identified as a functional food (Al-Jahani & Cheikhousman, 2017). These wonder seeds are generally considered agroindustrial waste, but they contain interesting nutraceutical properties. The see flavor and are consumed as roasted and salted snacks in some parts of mexico, the USA, Europe, Canada, and some Asian countries (Mallek-Ayadi *et al.,* 2018) (Ayadi *et al.,* 2018).

Pumpkin seeds also work as a good antidepressant and have a 47% of anti-depressant food score, which might be due to the presence of 5-hydroxytryptophan (tryptophan intermediate) that helps in the formation of the neurotransmitter serotonin (Dotto & Chacha, 2020). The major phytosterols are campesterol, β-sitosterol, and stigmasterol, which act as sequestrants of bile acid and acyl-coenzyme and are inhibitors of cholesterol acyl-transferase (Jesch & Carr, 2017). The seeds are a valuable source of squalene, which is an organic compound originally produced by plants, animals, and humans. Squalene plays a chemopreventative role in protecting people from cancer by reducing lipid peroxidation, and it has major importance in the Mediterranean diet. Squalene protects normal but not tumor cells from chemotherapeutic toxicity. Reports

have shown that pumpkin seeds have also been reported to possess anti-cancer, antidiabetic, antitumor, antimicrobial, antimutagenic, antioxidant activities, anti-inflammatory, and antiulcer properties (Govindani *et al.,* 2012).

Sesamum indicum L. is one of the world's important oil crops. Its primary marketable products are whole seeds, seed oil, and meal. While sesame seeds have been grown in tropical regions throughout the world in India and were mentioned in early Hindu legends. In these legends, tales are told in which sesame seeds represent a symbol of immortality. From India, sesame seeds were introduced throughout the Middle East, Africa, and Asia. Sesame seeds were one of the first crops processed for oil as well as one of the earliest condiments (de Carvalho *et al.,* 2001). These seeds were brought to the United States from Africa during the late 17^{th} century. Currently, the largest commercial producers of sesame seeds include India, China and Mexico.

Sesame seeds add a nutty taste and a delicate, almost invisible crunch to many Asian dishes. They are also the main ingredients in 'tahini' (sesame seed paste) and the wonderful Middle Eastern sweet called 'halvah'. Sesame seeds may be the oldest condiment known to man dating back to as early as 1600 BC. They are highly valued for their oil which is exceptionally resistant to rancidity. "Open sesame", the famous phrase from the Arabian Nights, reflects the distinguishing feature of the sesame seed pod, which bursts open when it reaches maturity. The pods are tiny, flat ovals, measuring about 3 mm long. Seed color can vary, though they are usually beige or creamy white when husked. Sesame oil, other than its use as a cooking medium, has certain industrial applications as it is used to make hair oil, hydrogenated oil, and certain medicines (Salunkhe *et al.,* 1991; Suja *et al.,* 2004; Quasem *et al.,* 2009)

Sesame is a primary source of phytonutrients such as omega-6 fatty acids, flavonoids, antioxidants, vitamins, and dietary fiber with potential anti-cancerous and health-promoting properties. It is noteworthy to express that sesame which is rich in polyunsaturated fats (PUFA), sesamin and vitamin E extraordinarily decreases hypertension when contrasted with the circulatory strain bringing down medications. It likewise diminishes histological renal harm and degeneration of the blood vessel, an element not seen in a typical eating routine. Sesamin is significant for disease prevention and treatment of heart hypertrophy and renal hyper-strain. Insufficient intake of nutritiously balanced food hinders growth. Malnutrition and poor growth during early stages influence an expansive part of the world's total population. (Prasad, *et al.*, 2012) Studied that sesame nutraceuticals and pharmaceutical products can decrease the risk of neurological, dermatological, cancer, and heart disease. About 100 g of sesame seeds contribute a high amount of minerals, where calcium, iron, and zinc account for 1283, 15.05, and 7.7 mg, respectively. Therefore, various supplementary food products can be prepared by the addition of sesame to utilize their potential and enhance product quality by improving its taste and increasing its energy, protein, calcium, and mineral content. Incorporating sesame seeds into daily food items can make them both micro and macro-nutrient-dense, which can be used for feeding people of all ages. Commercially available nutrient supplements are limited to certain circumstances but such foods can be consumed in daily routine.

Sesame seeds are not only a good source of carbohydrates, protein, and fats (oil) but are also rich in micronutrients and bioactive components. It is known as the 'Queen of Oilseeds' as it is highly resistant to oxidation and rancidity. Sesame seeds are a

reservoir of nutrients.The proteins present in other oilseeds do not contain certain amino acids like tryptophan and methionine. However, this deficiency can be fulfilled by adding sesame, soybean meal, and flour, which are rich sources of tryptophan and methionineis helps to produce a balance in nutrition and diet. Moreover, many of these bioactivecompounds like phytosterols, tocopherols, and lignans such as sesamin and sesamolin are now being used as food additives in the production of various functional foods, that is, nutraceuticals. Likewise, these are also being utilized in pharmaceutics, cosmetics, and fungicides owing to their antiseptic properties which provide numerous health benefits to humans (Mohammed*et al.,*2022).

Materials and Methods

Plant Materials

The success of any scientific investigation mainly depends on the nature of the material used and the methods employed for the purpose. This chapter deals with methods of sample collection, preservation, and experimental methods involved in this study. The phytochemical, mineral, and antibacterial estimation of the selected edible seed of *Cucumis sativus, Linum usitatissimum, Cucurbita maxima, Sesamum indicum,* and *Helianthus annus* were collected from vallanadu Thoothukudi district, Tamil Nadu. The collected samples were dried under shade and dried and coarsely powdered using a blender. The final uniform powder was used for further analysis.

Cucumis sativus ***Linum usitatissimum***

Cucurbita maxima ***Sesamum indicum***

Helianthus annus

Phytochemical analysis

Test for Alkaloid (Wagner's test)

About 1ml of the extract was taken and a few drops of Wagner's reagent has addedto the formation of a reddish-brown precipitate indicating the presence of alkaloids.

Test for Flavonoid (Shinoda test):

About 1ml of the extract was added to a pinch of magnesium turnings and 1-2 drops of concentrated Hydrochloric acid were added. The formation of pink color indicated the presence of flavonoids.

Test for Glycoside:

About 0.5 ml of extract was dissolved in 1 ml of water and then an aqueous NaOH solution was added. The formation of yellow color indicates the presence of glycosides.

Test for phenol (Lead acetate test):

About 1 ml of extract was taken and 0.5 ml of 1% lead acetate solution was added and the formation of precipitate indicated the presence of phenolic compounds.

Test for Quinine:

A small amount of extract was treated with concentrated HCl and observed for the formation of the yellow color precipitate.

Test for Saponin (Foam test):

About 0.5 mg of extract was diluted with 20 ml of distilled water and shaken well in a graduated cylinder for 15 minutes. The formation of foam to a length of 1cm indicated the presence of saponins.

Test for steroids:

About 1 ml of extract was dissolved in 2 ml of chloroform and an equal volume of concentrated Sulphuric acid was added along the sides of the test tubes. The appearance of a brown ring indicated the presence of steroids.

Test for Tannin (Ferric chloride test):

About 1 ml of extract was taken and 0.5 ml of 5 % ferric chloride was added. The development of dark bluish-black color indicated the presence of tannins.

Nutritive value analysis:

Test for Carbohydrate (Benedict's test):

About 5ml of Benedict's solution was added to 1ml of extract and boiled in a water bath. The appearance of red or yellow or green precipitate indicated the presence of reducing sugars.

Test for Fat:

About 1 g of the sample was diluted with 10 mlof distilled water and shaken well in a graduated cylinder for 15 minutes. The formation of foam to a length of 1cm indicated the presence of lipids.

Test for protein:

Test for Terpenoid:

5ml of the extract was mixed with 2ml of chloroform and concentrated H_2SO_4 to form a layer. A reddish-brown coloration of the interface showed the presence of Terpenoids.

Test for Coumarin:

About 3 ml of 10% NaOH was added to 2 ml of plant extracts. The formation of a yellow color was an indication of the presence of coumarins.

Vitamin C: - (Baker *et al.*, 1968)

Reagents:

5% of $NaNO_2$

10% $AlCl_3H_2O$

1 M NaOH

Procedure:

100 mg of plant material was homogenized with 10 ml of two different solvents (Distilled water, Methanol) and filtered through a muslin cloth. 0.5 ml aliquots of each extract were mixed with 3 ml of distilled water and 0.2 ml 5% $NaNO_2$ solution. After 6 minutes 0.3 ml of 10% $AlCl_3.H_2O$ solution was added. After 5 minutes 1 ml of 1 M NaOH solution was added and then the total volume was made up to 2.5 ml with distilled H_2O. Following through mixing of the solution, the absorbance against the blank was determined at 510 nm. "l-ascorbic acid was used as standard and the result were expressed as mg/1g/FW".

Vitamin E:

Reagents:

Absolute alcohol

Xylene

2,2 dipyridyl

Ferric chloride solution

Standard solution

Sulphuric acid

Extraction of Tocopherol:

The plant sample (2.5g) was homogenized in 50 ml of 0.1 N sulphuric acid and allowed to stand overnight. The content of the flask was shaken vigorously and filtered through Whatman No.1 filter paper. Aliquots of the filter were used for the estimation.

Procedure:

Into 3 stoppered centrifuge tubes, 3 ml of the plant extract, 3 ml of the standard and 3 ml of water were pipette out separately. To all the tubes, 3 ml of ethanol and 3 ml of xylene were added, mixed well, and centrifuged. Xylene (2ml) layer was transferred into another stoppered tube. To each tube, 2 ml of dipyridyl reagent was added and mixed well. The mixture (3ml) was pipette out into a cuvette and the extinction was read at 460 nm. Ferric chloride solution (0.66ml) was added to all the tubes and mixed well. The red color developed was read exactly after 15 minutes at 520 nm in a spectrophotometer. Tocopherol was used as standard the concentration of tocopherol in the sample was calculated by using the formula;

Tocopherol (µg) = (Sample A 520 – A 460 × 0.29 ×0.15) / Standard A 520

Determination of total Antioxidants:

Total phenol content:

The total phenolic content was estimated by the FolinCiocalteu method as described by Singleton *et al.,* (1965) with slight modifications. The extract (1 mg. mL-1) was mixed with 5 mL of distilled water, 1 mL of sodium carbonate (20%), and 1 mL of FolinCiocalteu reagent. The mixture was allowed to stand in a water bath for 30 min at 40°C. The content of total phenolic compounds was expressed as mg of gallic acid equivalents per g dry matter (mg GAE. G-1DM). The absorbance was measured at 765 nm using a UV-Vis spectrophotometer T60 U. All the experiments were run in triplicate. The mean values and standard deviations were calculated using Microsoft Excel software (Microsoft Corporation, Redmond, WA).

Total Flavonoid content:

The flavonoid content was determined by the aluminum trichloride method using catechin as a reference compound (Zhishen *et al.,* 1999). A volume of 125µL of the extract is added to 75 µL of a 5% $NaNO_2$ solution. The mixture was allowed to stand for 6 min, then 150 µL of aluminum trichloride (10%) was added and incubated for 5 min, followed by the addition of 750 µL of NaOH (1M). The final volume of the solution was adjusted to 2500 µL with distilled water. After 15 min of incubation, the mixture turned pink and the absorbance was measured at 510 nm. The total flavonoid content was expressed as g E catechin.100g-1DM.

Total condensed tannin contents:

The tannin contents or Proanthocyanidin were determined by the method of (Broadhurst et al., 1978) with slight modification, using catechin as a reference compound. A volume of 400µL of the extract is added to 3 mL of a solution of vanillin (4% in methanol) and 1.5 mL of concentrated hydrochloric acid. After 15 min of incubation, the absorbance was read at 500 nm. The condensed tannin was expressed as g E.Catechin.100g-1DM.

Determination of Antioxidants Activity:

Free radical-scavenging ability by the use of a stable DPPH radical:

The free radical scavenging assay was measured by the 2,2-diphenyl -1- picrylhydrazyl (DPPH) method proposed Brand-Williams *et al.,* 1995. (1988) with slight modification. 1 ml aliquots of test samples were added to 3 ml of 0.004% DPPH solution prepared in methanol. The mixture was added to Trolox for 1 minute and kept at room temperature for 30 minutes in the dark. The absorbance was read at 517 nm in a UV – spectrophotometer. A low absorption of the reaction mixture indicated a high free radical scavenging activity.

DPPH scavenging activity % = ((Abs Control- Abs test/Abs control) × 100

Where A control is the absorbance of the DPPH solution without the test solution. A test is the absorbance of the DPPH with the test solution. Methanol was used as a blank. Ascorbic acid was used as a positive control.

Antibacterial activity - Disc diffusion Assay:

The antibacterial activity of each plant extract was analyzed using human pathogens, Gram-positive bacteria, *Bacillus,*and Gram-negative bacteria *E.coli* obtained from the

Department of Microbiology St, Mary's College (Autonomous), Thoothukudi. Each bacterial pathogen was subcultured in an agar medium and maintained. What man No: 1 sterile filter paper discs (5nm) were impregnated with 1 mg/ml concentration and dried aseptically at room temperature. The spread plates were prepared withthe proper concentration of agar medium. Each sample-located disc was placed in the seeded agar plate. After 24-28 hours of ±37° C incubation, the diameter of the inhibition zone was measured, for positive control, a streptomycin disc (100 µg/ml) was used; Whereas for the negative control, respective solvents were loaded on sterile discs.

Results and Discussion

Phytochemical constituents in plant samples are considered to be biologically active compounds with a variety of functions including antioxidant, antimicrobial, antifungal, hypoglycemic, anti-diabetic, anti-inflammatory, anti-carcinogenic, anti-malarial, anticholinergic properties (Hossain and Nagooru, 2011).

Phytochemical analysis

Qualitative phytochemical screening was done for the evaluation of major phytochemical constituents such as phenol, flavonoid, glycosides, alkaloids, coumarin, quinine, steroids, tannin, saponin, and terpenoids using the standard procedure of analysis. The existence of phytochemical constituents of phenol, alkaloids, coumarin, and steroids was reported in seed aqueous, ethanol, and chloroform extracts (Table 1). However, the flavonoid content was reported in aqueous and chloroform extracts. other than this glycoside and quinone was reported as aqueous extracts, which play an important role in oxidative stress and have a diverse role in medicine, including anti–cancer agents and anti-aging and arteriosclerosis (Damodar *et al*., 2011).

Meanwhile, the tannin content was reported in only the sunflower seed of aqueous and ethanolic extracts (Table 1). Tannins occurred in varying concentrations in the oilseeds, which may be considered a major barrier to the use of the seed (Enujiugha and AyodeleOni 2003). Tannin isa polyphenolic substance with various molecular weights and a variable complexity(Makkar, 2003), it may form a soluble or insoluble complex with proteins (Mole and Waterman, 1987), minerals and vitamins Salunkhe *et al.* (1990) and reduce their bioavailability. Tannins in the diet not only precipitate in oral proteins, producing an astringent sensation but also interact with dietary proteins and digestive

Table No: 1 Phytochemical ScreeningOf Selected Edible Seeds

S.NO	Test	Water					Ethanol					Chloroform				
		Cucumber	Flax	Pumpkin	Sesame	Sunflower	Cucumber	Flax	Pumpkin	Sesame	Sunflower	Cucumber	Flax	Pumpkin	Sesame	Sunflower
1	Phenol	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
2	Flavonoids	-	+	-	+	+	-	-	-	-	-	+	-	+	+	-
3	Glycoside	+	+	-	+	+	+	+	+	+	+	-	+	-	-	-
4	Alkaloid	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
5	Cumarin	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+
6	Quinine	-	+	-	+	+	-	-	+	+	+	+	+	-	+	-
7	Steroids	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
8	Tannin	-	-	-	-	+	-	-	-	-	+	-	-	-	-	-
9	Saponin	+	+	+	+	+	-	-	-	-	-	-	-	-	-	-
10	Terpenoids	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

enzymes, resulting in a variety of antinutritive and toxic effects. In addition, this tannin reduces the bioavailability of vitamins (D'Mello, 1995). So, the present phytochemical screening proved that the sunflower seed has the potential to observe vitamins.

Quantitative Estimation

Total phenolic content (TPC)

Phenolic compounds are the most widelysecondary metabolites and are believed to be responsible for antioxidant activity (Tepe*et al.,* 2006). The total phenolcontents were higher in the seeds of*Hannus* (0.895 mg/g) and *L. usitatissimum* (0.622 mg/g) than in the other seeds. Phenolic compounds have a variety of beneficial activities. They have potential antioxidants and free radical scavengers (Meenakshi *et al.* 2012). Plant phenolics have multiple biological functions such as antioxidant, anti-inflammatory, anti-cancer, and antimicrobial activities (John&Grohmann,2001).

Total Flavonoids Content (TFC)

Flavonoids are secondary metabolites and have responsible for antioxidant activity in the medicinal field. These metabolites are mostly used in plants to produce yellow and other pigments which play an important role in the colors of plants. In addition, Flavonoids are readily ingested by humans and they seem to display important anti-inflammatory, anti-allergic, and anti-cancer activities (Crozier *etal.,* 2006). The total flavonoid content was recorded in the *L.usitatissimum* (0.134mg/g) and *C.maxima* (0.127 mg/g). flavonoids are potent antioxidants and epidemic studies indicate that high flavonoidintake is correlated with decreased risk of lifestyle diseases like diabetes and cardiovascular diseases (Kaur *et al*., 2016). Flavonoids are potent water-soluble

Table No: 3 Antioxidant ActivityOf Some Selected Edible Seeds

S.NO	Seeds samples	Flavonoids	Phenol	Tannin
1	**Cucumber**	0.038	0.261	0.050
2	**Flax**	0.134	0.622	0.152
3	**Pumpkin**	0.127	0.254	0.005
4	**Sesame**	0.098	0.212	0.037
5	**Sunflower**	0.119	0.598	0.041

antioxidant and free radicles which prevent oxidative cell damage and have strong anti-cancer activity (Havsteen, 2008)

TotalTannins

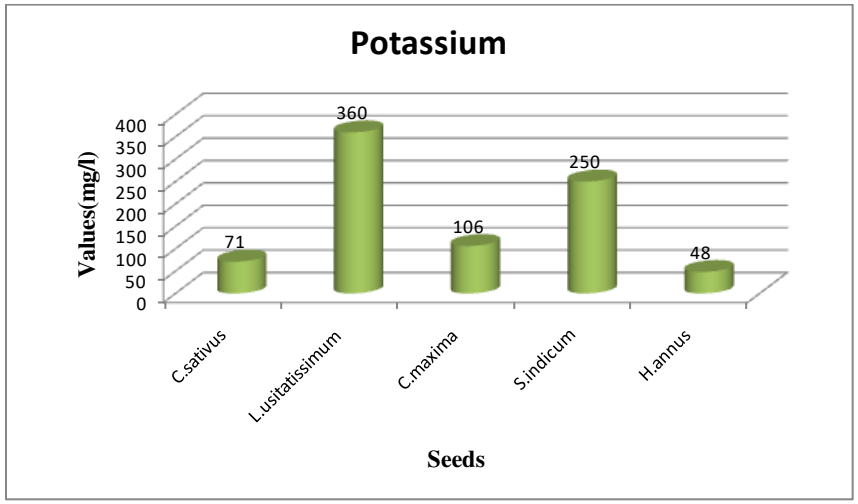
From the selected seed variety, The *L. usitatissimum* (0.152 mg/g) with showed the highest tannin content, and the lowest level was reported in *C. maxima* (0.005mg/l). The presence of tannins and flavonoids in the plants exhibited various biological activities like antibacterial, antifungal, antioxidant, and anthelmintic (Pulipati *et al.,* 2017). Lie *etal.*,2003, and Akinpelu *et al*., 2019 reported that Tannins are useful in treating inflammation, and ulcer and have remarkable activity in cancer prevention and anticancer activities.

Minerals analysis:

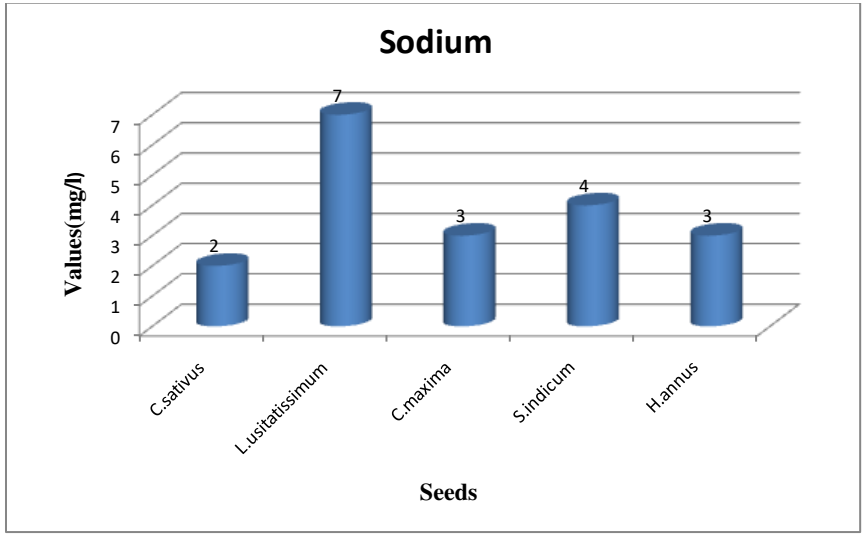
Minerals are required for normal growth, activities of muscles and skeletal development, cellular activity and oxygen transport (copper and iron), a chemical reaction in the body and intestinal absorption (magnesium), fluid balance, and nerve transmission (sodium and potassium). Deficiencies of these nutrients and minerals are known to affect the performance and health of both humans and livestock (Merck, 2005). The selected plant seeds used in this study contained an appreciable amount of minerals (Table 2), In this study, the seed's mineral composition like Na and K was analyzed using a flame photometer. According to the mineral element analysis, the *annus* seeds contained low levels of potassium (48mg/g), and the *cucumis* seed contain low levels of sodium (2 mg/g)was noted. The result of this study showed that all these five seeds contained reasonable amounts of phytochemical constituents and mineral composition,

Table No: 2 Mineral Composition of Some Selected Edible Seeds

Potassium Test



Sodium Test



which suggests the application of the seeds as supplementary sources of essential nutrients to humans and livestock.

Antioxidant

Antioxidants protect cells against the damaging effects of reactive oxygen species otherwise called, free radicals such as singlet oxygen, superoxide, peroxyl radicals, hydroxyl radicals, and peroxynitrite which results in oxidative stress leading to cellular damage (Mattson & Cheng, 2006). Natural antioxidants play a key role in the health maintenance and prevention of chronic and degenerative diseases, such as atherosclerosis, cardiac and cerebral ischemia, carcinogenesis, neurodegenerative disorders, diabetic pregnancy, rheumatic disorder, DNA damage, and aging (Uddin *et al.,* 2008; Jayasri *et al.,* 2009). Thus, free radicals are involved in a number of diseases including tumor inflammation, hemorrhagic shock, atherosclerosis, diabetes, infertility, gastrointestinal ulcerogenic, asthma, rheumatoid arthritis, cardiovascular disorders, cystic fibrosis, neurodegenerative diseases (e.g. parkinsonism, Alzheimer‟s diseases), AIDS and even early senescence (Chen *et al*., 2006; Uddin *et al*., 2008). The human body produces an insufficient amount of antioxidants which are essential for preventing oxidative stress. Free radicals generated in the body can be removed by the body's own natural antioxidant defenses such as glutathione or catalases (Sen, 1995). Therefore, this deficiency had to be compensated by making use of natural exogenous antioxidants, such as vitamin C, vitamin E, flavones, β-carotene and natural products in plants (Diplock *et al*., 1998).

Table No: 4 Total ContentOf Vitamin C & Vitamin E In Selected Edible Seeds

S.NO	Seeds Samples	Vitamin C	Vitamin E
1	C.sativus	0.032	0.515
2	L.usitatissimum	0.219	0.339
3	C.maxima	0.197	0.521
4	S.indicum	0.298	0.365
5	H.annus	0.197	0.205

Total Vitamin C

S.indicum(0.298mg/g) and *L.usitatissimum*(0.219mg/g) contain a significant amount of vitamin C. it is a vital component in a human healthy diet with the highest concentrations in animal organs. Vitamin C is a non–enzymatic and water-soluble antioxidant (Ueta *et al.,* 2003). Their main functions are enzyme activation, oxidative stress reduction, and immune function. It protects against respiratory tract infections and the risk of cardiovascular diseases and cancer treatment.

Total Vitamin E

Vitamin E is a fat-soluble nutrient found in many foods (Jacob, 1995). In the body, it acts as an antioxidant, helping to protect cells from the damage caused by free radicals. Free radicals are compounds formed when our bodies convert the food we eat into energy (Havsteen, 1983). According to this present study, the maximum level of vitamin E was noticed in *C.sativus* (0.515mg/g) and *C.maxima* (0.521mg/g) (Table 5).Though Sunflower seed is an excellent source of vitamin E /tocopherol which neutralizes free radicals, scavenges them, and prevents oxidative damage to cellular and molecular components exhibiting anti-inflammatory, cardioprotective, and antitumor action. Also, the important impact of vitamin E on the cardiovascular system makes sunflower seed oil beneficial in reducing atherosclerosis and hence complications like coronary artery disease and stroke (Singh *et al.*, 2005; Dutta and Dutta, 2003).

Table No: 5 DPPH Activity

S.NO	Samples	DPPH
1	Cucumber	1.561
2	Flax	1.551
3	Pumbkin	1.023
4	Sesame	1.421
5	Sunflower	0.651

Antioxidant activity:

DPPH scavenging activity:

The DPPH is a stable free radical and is widely used to assess the radical scavenging activity of a specific compound (Wei *et al.,* 2012). DPPH solution shows a strong absorption band at 517 nm appearing as a deep violet color. The absorption vanishes and the resulting decoloration is stoichiometric with respect to the degree of reduction. The seed samples were able to reduce stable DPPH radical to yellow color diphenyl picrylhydrazine. The degree of reduction in absorption is the reflection of the radical scavenging power of the compound.

The antioxidant activity of*Cucumis sativus, Linum usitatissimum, Cucurbita maxima, Sesamum indicum,*and*Helianthus annus*was evaluated by using a DPPH scavenging assay (Fig. 4). *C.sativus* seed shows higher scavenging activity (1.561mg/g) followed by *C.maxima* seed (1.551mg/g) as shown in (Fig 4). This result indicated the seeds of *C.sativus* and C.maxima shows higher scavenging activities than the other seeds, it could be due to the presence of phenolics groups which is responsible for redox properties, which allow acting reducing agent, hydrogen donor, and singlet oxygen quenchers (Arasali and Kadimi, 2009).

Anti – Bacterial Activity

The antimicrobial activity of variousselected seeds was investigated against the selected clinical pathogens such as *Bacillus subtilis, and Escherichia coli*, by using the disc diffusion method. All the examined plant extracts showed varying degrees of antibacterial activity against the clinical pathogens tested. The diameter of the inhibition

Table No: 6 Antibacterical Activity of Selected Edible Seeds

S.NO	Samples	*Bacillus*				*Ecoli*			
		H_2O	Chloroform	Ethanol	Streptomycine	H_2O	Chloroform	Ethanol	Streptomycine
1	Cucumber	1	0	2	4	2	0	2	4
2	Flax	2	0	0	2	4	3	4	5
3	Pumbkin	4	0	0	4	5	0	1	3
4	Sesame	3	0	4	4	2	3	4	2
5	Sunflower	3	0	0	2	5	3	3	5

zones against the species ranged from 0.5 to 0.7 mm. The study observed that the seeds of *C. maxima*and*H.annus* (Table 6) extracts have the maximum potentiality to inhibit the antibacterial activity than the other seed variety. This research work revealed that all the selected seed varieties are good and active against all the pathogens tested.

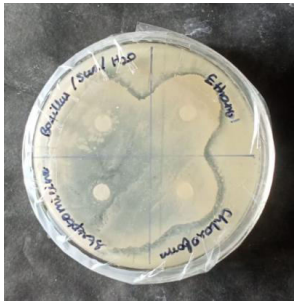
Palte 1: In Vitro Antibacterial Activity of Selected Edible Seeds Against Human Pathogens (*Bacillus)*

Cucumis sativus

Linum usitatissimum

Cucurbita maxima

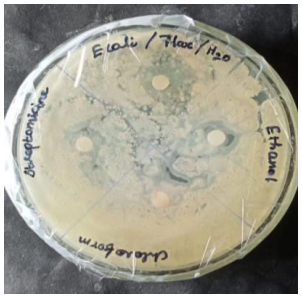
Sesamum indicum



Helianthus annus

Palte 2: In Vitro Antibacterial Activity of Selected Edible Seeds Against Human Pathogens (*E.coli)*

Cucumis sativus



Linum usitatissimum

Cucurbita maxima

Sesamum indicum

Helianthus annus

SUMMARY AND CONCLUSION

Health is the topmost priority for every individual. In the current scenario of the rising number of health problems; one needs a remedy with multifaceted actions to keep a check on a variety of factors responsible for the causation of various ailments. Seeds are an excellent nutrition package with their gold mine of healthy minerals and their niacin and folic acid contents. Seeds are considered to be the best sources of iron and zinc.Nutrients rich foods are vital for proper growth both in adults and children. Since most of the selected medicinal plant seeds are usually eaten in combination with other dietary components, some of which may be better sources of the minerals under consideration, these seeds could be of value in supplementing the minerals available from these other sources.

Based on this, the research work is mainly focused on qualitative phytochemical screening was done for the evaluation of major phytochemical constituents such as phenol, flavonoid, glycosides, alkaloids, coumarin, quinine, steroids, tannin, saponin, terpenoids using the standard procedure of analysis. The existence of phytochemical constituents of phenol, alkaloids, coumarin, and steroids was reported in seed aqueous, ethanol, and chloroform extracts (Table 1). However, the flavonoid content was reported in aqueous and chloroform extracts. other than this glycoside and quinone was reported as aqueous extracts, which play an important role in oxidative stress and have a diverse role in medicine, including anti–cancer agents and anti-aging and arteriosclerosis (Damodar *et al.*, 2011).

The selected plant seeds contained appreciable amounts of nutrients such as carbohydrates, and protein Remarkably, high contents of total phenolic compoundswere

detected in the seeds from *H.annus*, *L.usitatissimum,* and *C.maxima* also featured the highest flavonoid contents Mineral elements such as Na and K which are nutritional requirements of both humans and livestock which suggest that these seeds could be useful as a feed supplement to improve health and growth performance in humans and livestock. Taking all this information into account, seeds from different species could be a valuable source of mineral elements and phenolic compounds.

Plant phenolics have multiple biological functions such as antioxidant, anti-inflammatory, anti-cancer, and antimicrobial activities (John & Grohmann,2001). The total phenol contents were higher in the seeds of *H. annus* (0.895 mg/g) and *L. usitatissimum* (0.622 mg/g). then the other seeds. Phenolic compounds have a variety of beneficial activities. They have potential antioxidants and free radical scavengers (Meenakshi *et al.* 2012). In addition, Flavonoids are readily ingested by humans and they seem to display important anti-inflammatory, anti-allergic, and anti-cancer activities (Crozier *et al.,* 2006). The total flavonoid content was recorded in the *L. usitatissimum* (0.134 mg/g) and *C. maxima* (0.127 mg/g). flavonoids are potentialantioxidants and epidemic studies indicate that high flavonoidintake is correlated with decreased risk of lifestyle diseases like diabetes and cardiovascular diseases (Kaur *et al*., 2016). Flavonoids are potent water-solubleantioxidants and free radicles which prevent oxidative cell damage and have strong anti-cancer activity (Havsteen, 2008)

According to the mineral element analysis, the seeds contained low levels of potassium (48mg/g), and *cucumis* seed contain a low level of sodium (2 mg/g) and manganese (17 mg/g), while the higher level of calcium (170 mg/g) was noted in *Linum*. The result of this study showed that all these five seeds contained reasonable amounts of

phytochemical constituents and mineral composition, which suggests the application of the seeds as supplementary sources of essential nutrients to humans and livestock.

Thus, free radicals are involved in a number of diseases including tumour inflammation, hemorrhagic shock, atherosclerosis, diabetes, infertility, gastrointestinal ulcerogenesis, asthma, rheumatoid arthritis, cardiovascular disorders, cystic fibrosis, neurodegenerative diseases (e.g. parkinsonism, Alzheimer's diseases), AIDS and even early senescence (Chen *et al*., 2006; Uddin *et al*., 2008). Therefore, this deficiency had to be compensated by making use of natural exogenous antioxidants, such as vitamin C, vitamin E, flavones, β-carotene, and natural products in plants (Diplock *et al*., 1998). The present study showed *S. indicum* (0.298mg/g) and *L. usitatissimum* (0.219mg/g) contain a significant amount of vitamin C. Vitamin E is a fat-soluble nutrient found in many foods In the body, it acts as an antioxidant, helping to protect cells from the damages caused by free radicals. This present study showed the maximum level of vitamin E was noticed in *C. maxima* (0.521mg/g) and *C. sativus* (0.515mg/g) (Table 5).

This study was carried out to evaluate the in -vitro antibacterial activity of selected seed varieties by agar disc diffusion method against gram-negative bacteria -*E.coli*. gram positive bacteria *B. subtilis*. The data showed the maximum inhibitory zone was noticed in *C. maxima* (5mm) and *H. annus* (6mm) and the best activity was obtained with aqueous extract. The results from the current study indicate that *L. usitatissimum*and *H. annus* seed extracts contained varied types of pharmacologically active compounds like phenol, saponin, flavonoid, and antimicrobial activities.

The pharmacological effect of the phytochemical constituents such as alkaloids, glycoside, reducing sugar, and flavonoids as well as the antimicrobial activity of the plant

seeds can explain the rationale for the use of these plant seeds in the treatment of infections in traditional medicine. It is expected that using natural products as therapeutic agents will probably not elicit resistance in microorganisms. Therefore, the outcome of this research work suggests that the selected seeds could probably be a trustworthy and cheaper substitute for conventional drugs since the plant is easily obtainable and the extract can easily be made through a simple process.

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ASSESSMENT OF PTERIDOPHYTE DIVERSITY ENROUTE ENAYAM TO MARTHANDAM, KANNIYAKUMARI DISTRICT

A short term project work submitted to

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CERTIFICATE

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CONTENTS

INTRODUCTION

Pteridophytes are considered one among most primitive group and early land dwellers of vascular plants that emerged on this planet in the mid-Palaeozoic era during the Silurian period (Dudani *et al.*, 2011, 2014). Pteridophytes are a plant group that falls between non-tracheophytes and spermatophytes and account for over 48 families, 587 genera, and 12000 species worldwide (Zaman *et al.*, 2019). Most species are found in tropical and moist temperate regions followed by subtropical regions while, they also occur in different eco-geographical regions of the world where the conditions are not conducive for growth (Khullar, 2000; Dixit, 2000). Pteridophytes comprise a significant component of the forest ecosystem. The species richness is affected by the rainfall, moisture, and habitat availability (Murad *et al.*, 2000). The responsible factor for greater colonization of pteridophytes in terrestrial ecosystems is due to their adaptation to terrestrial condition by evolving specialized tissues for the translocation of water and food. Vascular tissue development not only contributed in water and food translocation but also played crucial role in mechanical support (Dudani *et al.*, 2011).

Among the diverse form of plant life, on one end are the unicellular algae, confined to moist habitats and on the other end are the higher plants capable of growing on relatively dry land. Plant life is assumed to have originated in water and in the process of evolution from a free living independent cell, the fundamental unit of structure and function evolved a multicellular organism with a division of labour among its tissue. Probably, the next advance was its remarkable manifestation, the alternation of generation. The cyclic alternation between sexual and asexual generation made possible the invasion of land and migration to land was accompanied by a corresponding complexity of internal organization was partly an adaptation to a

change of habitat from water to land and in amply illustrated by bryophytes-the amphibians of the plant kingdom. Bryophytes with a simple internal organization have a limited capacity for survival on dry land. The land plants have complex internal organization and pteridophytes occupy an intermediate position between bryophytes and higher land plants (Rashid, 1976).

Pteridophytes constitute a significant and important group in the plant kingdom. As the first true land plants, they offer a very favourable material for the study of various adaptations that have made the colonization of land possible for the plants. Pteridophytes have a long geological history on our planet. They are known from as far back as 380 million years. Fossils of pteridophytes have been obtained from rock strata belonging to Silurian and Devonian periods of the paleozoic era (Rajan, 1995).

Most of the flora in the present world is comprised of seed-bearing plants known as angiosperms, but around 280-230 million years ago, seedless vascular plants, pteridophytes dominated our planet surface (Mehra, 1961; Bir, 1987a, 1994). This is one of the most primitive plant groups and is known as the 'vascular cryptogams'. They are also known as 'ferns and fern allies'. It is estimated that pteridophytes originated during the Silurian and Devonian periods, and became the dominant plant group in the carboniferous period. They played a massive role in the establishment of plants on land during these periods. Due to their high reproductive ability and simple genetic makeup, pteridophytes quickly invaded the seashores, riverbanks, and places with very little disturbances (Kenrick and Crane, 1997). In the past, there were large fern trees that showed secondary growth, but at present, the numbers of tree ferns are very few. Currently, pteridophytes constitute the second largest floral group.

Fern and fern-allies can be seen in humid and shady places. The common types of forests that act as habitat for pteridophytes are tropical, subtropical and moist- deciduous forests. Geographically ferns can be seen from sea level to the highest mountains (Dixit, 2000). They are highly sensitive to microclimatic conditions, thus even small disturbances in the climatic and other environmental factors can cause their extinction from the natural habitats, and hence the existence of pteridophytes is largely dependent on the existence of these forests. Pteridophytes can also be seen as epiphytes on trees, lithophytes on the crevices of rocks, and even as hydrophytes completely immersed in water.

India is a subcontinent that hosts a wide variety of habitats. The climatic condition in India varies from the hot and dry Thar Desert in the North- West to the wettest place on the earth, Mawsynram in the North- East. The country is home to the dry deciduous forest in its southern part and also accommodates the Great Himalayas in the North. In this way, eachpart of this subcontinent shows a variety of habitat for pteridophytes and a large portion of these species are endemic. The main hotspots of pteridophyte diversity in India are:

- The Eastern Himalayas
- The Western Himalayas
- The Western Ghats
- The Eastern Ghats
- Central India
- Andaman and Nicobar Islands

Pteridophytes are a plant group that falls between non-tracheophytes and spermatophytes and account for over 48 families, 587 genera and 12000 species

worldwide. The richness of the species is affected by the rainfall, moisture and habitat availability. Most species are found in tropical and moist temperate regions followed by subtropical regions. They are wildly utilized as vegetables, traditional remedies, and for landscaping and gardening. Proteins, vitamins, crude fiber and minerals are all found in edible pteridophytes and steroids, terpenoids, phenolic acids, and flavonoids are only a few of the compounds found in them (Khoja *et al.,* 2022).

Pteridophytes are the seedless vascular cryptogams which occupy a crucial central position in the evolutionary history of plant kingdom between the lower non-seed bearing and higher seed bearing plants. In India, pteridophytes constitute an important component of flora next to angiosperms (Chandra *et al.,* 2008). Jain and Sastry (1980) reported 17 rare and endangered species of pteridophytes from India. The pteridophytes are moisture and shade loving plants that dependent upon the microclimatic conditions of the region for their successful survival (Rekha and Krishnan, 2017).

About 9% world Pteridophytes occurs in India or only in 2.5% landmass of the world. Ferns and Fern-allies in Indian flora and represented by 33 families 130 genera and 1267 species among them 70 species are endemic to India. In India Pteridophytes are distributed in all the phytogeographical zones of India ranging from sea level to alpine Himalayas where they grow as hydrophytes, mesophytes, lithophyte, epiphyte, hemiepiphyte, climbers etc. They are found in all ground habitats such as ravine, forest floor, on slopes, grassland, on rocks and crevices, on open walls and stone boulders and at certain places they form gregarious thickets. As epiphytes different species of Pteridophytes also distributed on different part of tree as on base of tree, bole, branches, forking. Pteridophytes, the seedless vascular plants, had a very flourishing past in dominating the vegetation on the earth about 280-230

million years ago. Although they are now largely replaced by the seed bearing vascular plants in the extant flora today, yet they constitute a fairly prominent part of the present day vegetation of the world. India with a highly variable climate has a rich diversity of its flora and Pteridophytic flora greatly contributes to its diversity. Pteridophytes also form an interesting and conscious part of our national flora with their distinctive ecological distributional pattern. On a very conservative estimate 500 species of ferns and 100 species of fern-allies are on record from India (Rawat and Satyanarayana, 2015).

There are about 12,000 species of pteridophytes occur in the world flora, of which 1,000 species into 70 families and 192 genera occur in the different parts of the present Indian political boundary. Keeping in view of large area of the country the present number of diversity is quite less. Region-wise studies reveals, that maximum number of 700 species (*i.e.* 58% of Pterido-phytes) occur in Eastern Himalayas and adjoining states. Thus, Eastern Himalayas may be termed as one of the Hot Spots diversity centre for pteridophytes. In other regions *viz.,* 400 species in Southern India,300 species in North-West India and 100 species in Central India and 125 species in Andaman and Nicobar Islands (Rawat and Satyanarayana, 2015).

According to a census, the Pteridophytic flora of India comprises of 67 families, 191 genera and more than 1,000 species (Dixit, 1984) including 47 endemic Indian ferns, less than 10% of those reported previously and 414 species of Pteridophytes (219 at risk, of which 160 - Critically endangered, 82 - Near-threatened and 113 - Rare), constituting 41-43 % of the total number of 950-1000 Pteridophytes of India. The vascular flora of our country in general has about 15,000 species and as a constituent of Indian flora of vascular plants, the ferns and fern-allies form only five percent part as far as the number of species is concerned. But, due to their abundance

in individuals as well as their conspicuousness in epiphytic vegetation and in the terrestrial vegetation along forest margins roadsides and forest floors, the contribution of ferns and fern-allies to the vegetational pattern in India rank only next to the flowering plants (Dudani *et al.,* 2011).

In view of variable climatic and altitudinal variations the Indian sub-continent represents Himalayas, Gangetic plains and Thar Desert as biodiversity centres. Maximum number of diversity of Pteridophytes observed in Himalayas, Eastern and Western Ghats. Except Pachmarhi and Parasnath hills; gangetic plains and Aravalli hills or towards North-west Hindu-Kudh much pteridophytes do not occur. The lesser rainfall from Eastern Himalayas to Western hills is responsible for a decrease in Pteridophytic vegetation (Dudani *et al.,* 2011).

Pteridophytes (ferns and fern-allies) are the most primitive vascular plants that appeared on the Earth, in the mid-Paleozoic era during the Silurian period which began 438 million years ago. They are the earliest of the plants ever evolved on the earth heralding the presence of a well-developed vascular system, xylem for water and phloem for food transport respectively and hence, are referred as 'vascular cryptogams' (Dudani *et al.,* 2014).

The arrival of pteridophytes, with specialized water and food conducting tissues, heralded an era of greater colonization in terrestrial ecosystems, to an extent that many of them could attain great heights like the flowering trees. After successfully establishing themselves as land plants, a very rapid rate of evolution was stimulated and witnessed among them with which they dominated most of the forests on earth's surface by the approach of carboniferous period. With the passage of time, especially with the evolution and dominance of the flowering plants, beginning in the late Cretaceous, the decline of the pteridophytes began Nevertheless, this interesting

group of plants, bridging the non-vascular cryptogams with the seed plants, higher in the evolutionary hierarchy, continues to occupy numerous niches on the land and in marshes and swamps and even in water bodies (Dudani *et al.,* 2011).

India has a rich and varied pteridophytic flora due to its Gondwanaland origin, its drift from south of the Equator towards Eurasia far north, carrying the progenitors of today's pteridophytes from Australia, Africa and Madagascar as well as probable endemics of its own. The rise of the Himalayas along the India-Eurasia merger line created diversified topography and varied climatic conditions ranging from warm and humid sea shores to arid deserts to elevations experiencing arctic cold, creating numerous micro-climates congenial for growth of ferns and fern allies, almost unparalleled anywhere on the Earth (Dudani *et al.,* 2014).

Moreover India's strategic geographical position would have facilitated migration of species, including several pteridophytes from Eurasia and South-east Asia and vice-versa, a notable factor that would have reduced endemism among the fern community. Today, among the vascular plants, pteridophytes form a major part next only to the angiosperms in India. Of the 12, 000 pteridophyte species enumerated in the world, around 1000 species from 70 families and 192 genera occur in India. The major centers for pteridophytes diversity are Eastern and Western Himalayas, Western Ghats, Eastern Ghats, Central India and Andaman and Nicobar Islands (Dudani *et al.,* 2014).

The Western Ghats constitute one of the 34 global biodiversity hotspots along with Sri Lanka, on account of exceptional levels of plant endemism and by serious levels of habitat loss. The rugged range of hills stretching for over 1600 km along the west coast 73° and 78° E), covering a geographical area of about 160,000 km2, is interrupted only by a 30 km break in Kerala, the Palghat Gap (Radhakrishna, 2001).

The Western Ghats have an average height of 900 m, with several cliffs rising over 1000 m. The presence of perennial streams and rivers, evergreen forests, grasslands and high altitude sholas and many other habitats of this mountain chain harbor almost 320 species of ferns and fern-allies (Dudani, 2014).

Development of vascular tissue not only contributed in water and food translocation but also played crucial role in mechanical support. Therefore, many of them such as Cyathea and Wilsonia (tree ferns) could attain great heights like trees. This interesting group of plants form an important component of forest ecosystem and act as connecting bridge between the non-vascular cryptogams and the seed plants and occupy various niches on the land, in marshes, swamps and in water bodies (Dudani *et al.,* 2011). They flourish in moist tropical and temperate forests while, they also occur in different eco-geographical regions of the world, where the conditions are not conducive for growth (Dixit, 2000). The majority of them thrive well in shady and moist places but a few survive in rock crevices and dry places while some of them such as Salvinia and Azolla grow in aquatic habitats (Bower, 1923).

According to Smith *et al.* (2006), it is estimated that there are about 9000-15000 species of pteridophytes that occur throughout the world while Moran (2008) predicted it to be approximately 13,600 species. Due to diversified topography, variable climatic conditions and geographical position, about 1200 species of pteridophytes are reported from India (Dixit, 1984, 2000; Sukumaran *et al.,* 2009; Dudani *et al.,* 2011; Patil *et al.*, 2013, Kavitha *et al.*, 2015; Kachhiyapatel *et al.,* 2015; Patel *et al.,* 2015; Rajput *et al.,* 2016), in which nearly 17% species are endemic (Sanjappa *et al.*, 2010).

First report on pteridophyte (*Ceratopteris thalictroides*) of Gujarat comes from the Flora of North Gujarat written by Saxton and Sedgwick (1918). Subsequent

studies on pteridophyte of Gujarat were taken up by contemporary researchers such as Phatak *et al.* (1953), Chavan and Mehta (1956), Gaekwad and Deshmukh (1956), Mahabale (1948, 1963), Padate (1969) and Inamdar and Shah (1967). Gujarat Ecological Commission (1996) documented 16 species of pteridophytes from different parts of the state, including forest areas. Thereafter, this group of plants was completely neglected and studies on them lagged behind. However, recently few sporadic reports about the occurrence of pteridophyte have been carried out by Patel *et al.* (2010), Dabgar (2012), and Modi and Dudani (2013).

Pteridophytes (comprises ferns and fern allies) represent the earliest vascular land plants originated some million years ago and still some of them are luxurious irrespective of all the geological and climatological disturbances which occurred from time to time, without many changes in their morphology. They form a conspicuous element of vegetation all over the earth's surface. Pteridophytes are important from the evolutionary point of view, because they show the evolution of vascular system in plants and also clearly show the process of evolution of seed habit in plants. They are considered as connecting link between higher vascular plants and lower non vascular plants. They remain primitive tracheophytes lacking flowers and seeds and propagate through haploid spores and exhibit independent alternation of generations (Kirishnan and Rekha, 2021).

Ecologically pteridophytes are adapted to almost all possible situations from tropics to temperate regions except in the Polar regions and deserts. They grow luxuriantly in moist tropical and temperate forests and their occurrence in different eco-geographically threatened regions from sea level to the highest mountains are of much interest. The world flora consists of approximately 12,000 species of pteridophytes of which around 1000 species distributed in 70 families and 192 genera

are likely to occur in India. Most of the pteridophytes diversity in India is observed in the Himalayas, Eastern and Western Ghats. The Western Ghats, is rich with more than 300 pteridophytes.

Western Ghats is the 1600 km long chain of hills of Peninsular India, ranging from the Tapti river basin of the southern Gujarat to the Kanyakumari of Tamil Nadu. It is one of the most significant geological structures, which controls the climate and culture of the Peninsular Indian states of Gujarat, Maharashtra, Karnataka, Kerala and Tamil Nadu. It offers innumerable microhabitats for the luxuriant growth of flora, including the pteridophytes, the second largest floral group.

Urbanization is a leading cause of habitat loss and biological homogenization (Mckinney, 2006). Remnant ecosystem embedded in urban areas, especially forests, provide important services such as moderating local climate, storing water, and filtering air, as well as increasing citizens' well-being (Alberti, 2010; Grimm *et al.*, 2008). However, the balance of components ensuring ecological integrity that is, biophysical structure, species composition and functional processes- is difficult to maintain in urban forest patches (Ordó˜nez and Duinker, 2012). This is because most of these components are disturbed at multiple spatial scales by human activities (Alberti, 2010). The plant communities of edge habitats are often composed of a higher proportion of competitive, pioneer, and ruderal plants than forest cores (Godefroid and Koedam, 2007; Guirado *et al.,* 2006; Lapaix and Freedman, 2010). The presence of edges may also favor the influx of invasive and exotic plants into forests (Cadenasso and Pickett, 2001). Observed changes in the composition of floristic assemblage of urban ecosystem are usually exacerbated by extensive areas of impervious surfaces that foster the formation of urban heat island (UHIs) (Bechtel and Schmidt, 2011; Godefroid and Koedam, 2007), which may in turn cause thermal and

hydric stress to organisms not adapted to these conditions (Grimm *et al.*, 2008). This anthropogenic climate disturbance, the human stranglehold over nature and the novel microhabitats created in a heterogeneous matrix differentiate urban areas from the other landscapes creating a unique distribution pattern of the plants that must be under-stood for the preservation of remaining ecosystems (Williams *et al.*, 2009). Although forests in urban areas are never pristine, it is possible to establish a scale of their relative integrity and to identify the forests whose conservation should be prioritized. Landscape metrics can be used to assess structural integrity, which is known to decrease in forests that are smaller, less connected, and surrounded by a strongly anthropized matrix (Carignan and Villard, 2002; Dale and Beyeler, 2001). Biological indicators that have either a negative or a positive response to eco-logical integrity can also be used (Carignan and Villard, 2002; McKinney, 2006; Ordó˜nez and Duinker, 2012).

Species richness estimates calculated from judiciously chosen guilds or functional groups, can effectively represent patterns and processes related to many components of ecosystems (Dale and Beyeler, 2001), but efficacy varies with sampling unit size (Dufrêne and Legendre, 1997) and a method to control this confounding effect is usually necessary (Bräuniger *et al.*, 2010). Species richness is a fundamental measure of community and regional diversity and underlies many ecological models that area analyzed by a number of species recorded in the samples (Tessler *et al.*, 2016). Elevation gradients can serve as natural experiments for studies on community and ecosystem responses to long-term changes in climate in a changing world (Sundqvist *et al.*, 2013). Pattern of species richness along elevation gradients is a classic subject in ecology and biogeography (Ah-Peng *et al.*, 2012).

Studies on relationship between species richness and elevation gradients

resulted in development of a more complete understanding about the nature and more effective plans for conserving biological diversity in the context of global change Grytnes and Beaman, 2006). Ecology of terrestrial and marine ecosystems has been studied over a century, and human utilization of both realms was documented going back thousands of years (Knapp *et al.,* 2017). The regional patterns of species richness are a consequence of many interacting factors, such as plant productivity, competition, geographical area, historical or evolutional development, regional species dynamics, regional species pool, environmental variables, and human activity (Zobel, 1997).

Species richness on elevation gradient studies identified two main correlation patterns such as monotonic and humped. "Monotonic" was referred to as decrease in species richness with increasing elevation, and "humped" was referred to as highest distribution and species richness near the middle of the elevation gradient (Rahbek, 1995; Grytnes and Beaman 2006). Carpenter (2005) stated that 49% of altitudinal gradient studies across the globe on different vegetation showed a humped species richness trend with 500 m or lower to 1500 m or higher elevation and that 24% of the studies showed little change in species richness at lower elevations and decline at higher elevations. Plant species richness declines monotonically above an elevation gradient of 1500 m (Vazquez and Givnish, 1998). Global climate is probably the principal determinant of the vegetation pattern, which has considerable influence on distribution, structure, and ecology of the forest ecosystem, and it is assumed that changes in climate would alter constitution of an ecosystem (Singh *et al.,* 2015).

Climate variables seem to be most important for explaining species richness patterns with elevation gradients for all kinds of living organisms (Bhattarai *et al.,* 2004). Climate factors, viz., temperature, potential evapotranspiration, length of the

growing season, humidity, air pressure, nutrient availability, ultraviolet radiation, and rainfall, are varied for elevation, all of which can have an effect on distribution of species and their richness along the gradient in any forest ecosystem (Funnell and Parish, 2001). The community structure and ecosystem processes always differ along elevation gradients in almost all vegetation. Ecosystem and ecological responses to elevation are commonly driven by changes in temperature, and many community and ecosystem level variables often respond similarly to elevation across contrasting gradients (Sundqvist *et al.,* 2013). A majority of elevation gradient studies are focused on woody plants (Kitayama, 1992; Liberman *et al.,* 1996), although the largest share of plant species in several forests belongs to nonwoody plants (Gentry and Dodson, 1987). Epiphytic plants that grow on the surface of trees contribute to more than half of the plant species richness in tropical forests, which provides unique microclimates and habitats for other species; thus, their decline could negatively affect many animals and plants that rely on them (Wang *et al.,* 2017).

Pteridophytes have been a popular subject of studies on species elevation relationships, with the highest diversity in tropical and subtropical mountains (Linder, 2001; Bhattarai *et al.,* 2004). It is generally accepted that tropical regions are reported to have higher species richness than temperature areas (Pianka, 1966), but documentation of diversity patterns within tropics is limited in particular to vascular cryptogams (Gentry, 1982). Pteridophytes shows a peculiar type of life cycle, that combines wind-dispersed spores and mostly with free-living gametophytes. This unique characteristic feature improves a surplus level of complexity when making biogeographical comparisons of pteridophytes with other vascular plants (Watkins *et al.,* 2006).

Pteridophytes are mostly related to its ecological structure, and very few

studies have addressed the change of diversity along elevation gradients. A majority of previous reports on distribution of pteridophytes along elevation gradients in different geographical regions show a "humped" distribution pattern (Watkins *et al.*, 2006). In India, contributions about the taxonomy, ecology, ethnobotany, and distribution pattern of pteridophytes are available in the literature from time to time, but enough attention has not been given toward the diversity of this unique group along elevation gradients.

Ecological indicators (EIs) are useful tools to link empirical results, models, and theories with environmental applications. They are broadly employed in the classification of environments and in the evaluation of natural and/or anthropic disturbance or stress (Siddig *et al.,* 2016). One definition considers EIs as a species or group of species that readily reflects the abiotic or biotic state of an environment (Dale and Beyeler, 2001; Heink and Kowarik, 2010). This definition includes only species and/or group of them, but other taxonomic levels or still ecological attributes could be adopted.

The total number of pteridophyte species present in India is c. 1100 and of these 337 taxa are considered to be threatened or endangered (nearly one third of the total). It should be realised that IUCN listing (IUCN, 2010) is organised by countries and the global rarity and endangerment of species is therefore often somewhat masked in an area where the floras are intimately related. This particularly applies to the two major groups of Sino-Himalayan and S. E. Asian/Malesian elements present in India which extend across the eastern borders into China, Myanmar etc. It also applies to the Lankan/ Indian peninsular element in the south, which contains the highest number of Indian endemics. A list of Asian globally threatened species of narrow distribution is given by Ebihara *et al.* (2012) for which the 76 Indian, Nepalese and

Bhutanese species listed have been extracted from the present paper. The present list is reduced compared to that of 414 threatened pteridophytes given by Chandra *et al.* (2008) as it concerns only the top six IUCN categories, EX (Extinct), EW (Extinct in the wild), CR (Critically endangered), EN (Endangered), VU (Vulnerable) and NT (Near threatened), whereas Chandra et al. (2008) list was a more preliminary one which did not set out to follow the IUCN categories until more information became available. The IUCN categories given here apply to political India only.

In addition more information about the status of species in Arunachal Pradesh has become available (Fraser-Jenkins, 2010b) and has revealed that a number of species that are very rare else-where in India are much more common in the far North-East in Arunachal Pradesh and some other North-Easternmost States of India. Adjustment has also been made to the status of a number of species, either taxonomically, or for IUCN category, now that more information from Indian herbaria, particularly CAL, BSA and LWG, has become available to the author. A few of the species that have now been excluded for taxonomic or other reasons have been listed here for explanatory reasons, but in square brackets and without categories. Many other species previously estimated to be endangered and endemic have been elucidated taxonomically by Fraser-Jenkins (1997, 2008a, 2008b) and Chandra *et al.* (2008) and excluded.

The classification of Fraser-Jenkins (2010a) has been used in the list, with some modifications according to more recent work. In general this is similar to that of Kramer and Green (1990) and Smith *et al.* (2006). But it is less similar to two molecular cladonomy lists recently produced by Christenhusz *et al.* (2011) and Rothfels *et al.* (2012), which are seen here as being insufficiently taxonomically based and to recognize too many groups that have no possible morphotaxonomic

significance. They also split many other groups that have been more successfully sunk into recognisable categories of more major value. Their schemes are therefore not accepted here as being applicable to taxonomic classification and are seen as being of less use to Botanists.

The Western Ghats, one among 34 global biodiversity hotspot centers, is rich in floral and faunal diversity with great endemism throughout the plant and animal kingdom (Daniels, 2003). The criteria for a region to be recognized as a "Biodiversity Hotspot" by Conservation International (2005) is the presence of a minimum of 1500 endemic species of vascular plants and the loss of at least 70 percent of its original habitat. Due to the distribution of endemism, Nayar and Geevarghese (1993) compared the ecological niches in the Western Ghats to be similar to that of an Island. The mountain chain has perennial streams, rivers, evergreen forests, and high altitude sholas, thus making a perfect habitat for ferns and fern-allies.

The Western Ghats covers a distance of 1600 km from Kanyakumari in Tamil Nadu to Tapti valley in Gujarat state with an area of 160000 km2. It runs parallel to the western coast of India traversing the states of Gujarat, Maharashtra, Goa, Karnataka, Kerala, and Tamil Nadu. The continuous stretch of the Western Ghats is interrupted by a 30 km break-in Kerala. The interrupted part is known as 'Palghat Gap' (Radhakrishna, 2001). Mountain ranges like Nilgiri Hills, Anamallays Hills and Palni Hills are present in the Ghats, and they have various forest types like scrub forests at an elevation of 200-500 m, moist deciduous forests (500-900 m), tropical moist evergreen forests (1200-1500 m) and Shola forest above 1500 m. The Western Ghats gets an annual rainfall of 1000 to 5000 mm.

Based on the studies done by Manickan and Irudayaraj (1992), the Western Ghats harbors 349 pteridophyte species out of 1200 Indian pteridophyte species.

According to Manickam and Irudayaraj (1992), a large portion of pteridophyte species in the Western Ghats is endemic. The most diverse genera present here is *Asplenium, Selaginella, Pteris, Athyrium, Diplazium*. Dudani *et al.* (2011) found that the major families seen in the Western Ghats were Aspleniaceae, Polypodiaceae, Thelypteridaceae, Selaginellaceae. According to Dixit (2000), based on the available literature, Karnataka holds the maximum diversity of pteridophytes in the family Aspleniaceae which is comprised of 27 species. Other major pteridophyte families were Polypodiaceae, Athyriaceae, Thelypteridaceae, Selaginellaceae, Pteridaceae.

Studies on pteridophytes are gaining momentum and this is shedding light on the medicinal and economic importance of these plants. Understanding the flora of a region always helps in understanding the change in the ecosystem and in-vitro and ex-situ conservation can be widely exploited to bring back the threatened species from the verge of extinction and preserve this plant group for our coming generations.

REVIEW OF LITERATURE

The study of pteridophytes of South India began with Van Rheede (1703) who included 20 illustration of 16 species of ferns and fern allies from Kerala and their description in *Hortus Malabaricus*. Linnaeus (1753) referred to this work while naming the Indian species in his *Species Plantarum*. Manilal (2003) transliterated Van Rheede's *Hortus Malabaricus* to English. References to ferns of south India can also be found in Hooker's *Icones Plantarum* (1837-1854) and *species filicum* (1846-1864).

The most significant contribution on the south Indian pteridology is by Col.R.H. Beddome, conservation of forests, Madras presidency. Based on his field work and study of earlier works (Hooker and Greville, 1829-1831; Hooker, 1846-1864; Hooker and Baker, 1868; Moore, 1857-1862). Beddome published his monumental work, Ferns of southern India (1864), which recorded 240 species of Pteridophytes from peninsular India. Following this, he published *Ferns of British India* (1866) and Supplement to the Ferns of Southern India and Ferns of British India (1876). In 1883, the Handbook to the ferns of British India, Ceylon and the Malay peninsula was published and supplement was added in 1892. Even to-day, Beddome's work remains as one of the most important and useful reference for the identification of Indian fern and flora. Since then, the concepts of various taxonomic groups of ferns have considerably been changed and altered. Nayar and Kaur (1974) and Chandra and Kaur (1994) have published the nomenclatural equivalents to the ferns described by Beddome.

The earliest major work on the pteridophytes of the Nilgiris was by Kunze (1851) who reported eighty two fern species and 12 fern allies and described 22 new species. Sharma *et al.* (1977) have listed 147 species of ferns and fern allies from the Nilgiris. Fraser-Jenkins (2010b) studied the pteridophyte diversity of Nepal.

Pteridology in India

The different accounts presented by Prof. R.C. Ching, dealing with the Himalayan ferns and C.B. Clarke's study of the North Indian ferns (Clarke, 1880) are perhaps the most noteworthy since Beddome's studies. Afterwards, Baynes (1887) beautifully illustrated 33 species of ferns from India in 17 plates in his *Album of Indian Ferns*.

During the past fifty years, there has been remarkable contribution in almost all fields of Pteridology by Indian botanists (Bir, 1987b). The studies on the ferns of peninsular India have been reviewed by Nair and Bhargavan (1981) which gives an insight in to the historical heritage of our culture related to the studies on ferns and their utilization in the indigenous system of medicine. Madhusoodanan *et al.* (2001) reviewed the studies and contributions made by different pteridologists on the various aspects of ferns and fern-allies of South India from 1947 to 1997.

Dixit and Vohra (1984) had given a brief account of all the genera reported from India in their 'A dictionary of Pteridophytes'. Dixit (1984) published A Census of Indian Pteridophytes giving author citations and distribution of ferns and fern allies occurring in India. Later Chandra (2000a) updated the nomenclature of 219 taxa. Chandra (2000b) also enumerated the ferns occurring in India with all their synonyms and distribution. Fraser-Jenkins (2008a) published a taxonomic revision of three hundred ferns occurring in India and provided a revised census list.

Various checklists of the fern flora of limited geographical regions have been published from time to time. The only attempt at a serious floristic study of any region is ferns and fern allies of Meghalaya state by Baishya and Rao (1982) which includes 256 species of ferns and fern allies along with key to their identification, and brief taxonomic account of each taxon. But it lacks detailed morphological descriptions of

taxa and taxonomic notes are not precise; hence the work is only of limited utility. An enumeration of species of plants of Palni hills (South India) is published by Manikam and Ninan (1976), listing 133 species. The ecology of 150 taxa of ferns of the Palni Hills have been studied by Manikam and Ninan (1976). Manikam (1986) also published Fern Flora of Palni Hills which includes detailed descriptions on ecological data of 137 species (including 8 new species) belonging to 64 genera and 13 varieties. Subsequently, Manikam and Irudayaraj (1992) published Pteridophyte Flora of the Western Ghats–South India which includes descriptions and illustrations of 239 species occurring in Western Ghats south of Palghat gap. Nayar and Geevarghese (1993) published the Fern Flora of Malabar giving elaborate descriptions of 170 species of ferns from this area.

Blatter and Almedia (1922) published Ferns of Bombay and provided a brief description of the taxa along with their description. Holtum (1976) has dealt the family Thelypteridaceae including 10 species for the flora of Hassan district, Karnataka. Nair *et al.* (1988, 1992a, 1992b and 1994) provided brief description and ecological account of the fern allies and ferns of Kerala. Pteridophytes of Karnataka state have been enumerated by Rajagopal (1999) published Polymorphic ferns of the Western Ghats which explains the polymorphism in 10 south Indian ferns. Pullaiah *et al.* (2003) published Pteridophyte in Andhra Pradesh, India enumerating a total of 89 species of ferns and fern allies belonging to 51 genera spread over 32 families.

Monographic and revisionary works

Holttum (1965) gave a detailed account on the Tree – Ferns of the genus *cyathea* in Asia and discussed about the taxonomy, nomenclature and distribution of nine species. Panigrahi (1975) revised the genus *Pityrogramma* in Asia and discussed

about the taxonomy, nomenclature and distribution of four species of *Pityogramma* naturalised in Asia. Holttum (1983) revised the family Thelypteridaceae in Europe.

Alston (1945) was the first person who enumerate 58 species of *Selaginella* from British India and provided a key for identification. Out of which 44 species are confined to the present political boundaries of India. Alston (1952) made a revision of the West Indian species of *Selaginiella.* Gupta (1962) revised the genus *Marsilea* in India. Later Panigrahi and Dixit (1969) studied the family Osmundaceae in India. Afterward Dixit published revisionary works on *Lycopodiaceae* of India (1988) and "Selaginellaceae of India*".* Singh *et al.,* (1989) revised the genus *Asplenium* in India and Fraser-Jenkins (1991) studied the genus *Polystichum* in India

In South India, revisionary works on families like Aspleniaceae (Azeez *et al.,* 2008), Hymenophyllaceae (Hameed *et al.,* 2003) Polypodiaceae (Nampy and Madhusoodanan, 1998), Thelypteridaceae (Leena and Madhusoodanan 1994; Manikam and Irudayaraj 1990), Pteridaceae (Manickam and Irudayaraj, 1991), Selaginellaceae (Nisha *et al.,* 2010) and genera like Microsorium (Madhusoodan and Nampy, 1993) *Adiantum* and *Cheilanthus* (Kurup, 2008), *Pteris* (Sreenvias, 2011) were carried out. Jyothi and Madhusoodanan (1993) studied the Cheilanthoid ferns of South India and Kurup *et al.* (2008) revised the primitive ferns of South India which includes 23 species coming under 12 genera.

Status of pteridophytes

The world conservation monitoring centre at Cambridge, England, listed 1650 threatened species of pteridophytes world-wide (Jermy, 1990), under the following categories: presumed Extinct-20, Endangered-67, vulnerable-9, rare-354, candidate species for conservation -1318. Based on their new criteria for species to be include in the red list, IUCN (1998) listed 770 threatened species of Pteridophytes world –wide.

Much study has been carried out in India on threatened species of flowering plants and their conservation, but ferns and fern–allies, which constitute edible and valuable part of the national flora, have attracted less notice. First attempt in this respect was the work done by Jain and Sastry (1980) who listed 17 rare and endangered pteridophytes from India along with Angiosperms. Afterward, Dixit (1983) reported 25 rare and interesting pteridophytes; whereas Datta (1983) listed only five pteridophytes as rare. Bir (1987b) identified 104 rare and endangered pteridophytes from different parts of India. Nayar and Sastry (1987, 1988, 1990) include 31 threatened pteridophytes in the Red Data Book of India Plants. Madhusoodanan (1991) listed rare and endangered ferns of Western Ghats of Kerala and later on, Manickam (1995) reported 44 rare and endangered species of ferns from the Southern Western Ghats. Bennianmin *et al.* (2008) developed a key for identification of rare and endangered ferns and fern allies in the Western Ghats.

Chandra *et al.* (2008) lists 29 fern-allies and 358 ferns as threatened taxa of pteridophytes from India. According to them 219 species of pteridophytes to be considered as 'At risk' in India and among them, 160 come into the IUCN category of 'Critically endangered'; 82 species to be considered as 'Near-threatened' and 113 species to be considered under the category of 'Rare'. Recently, Ebihara *et al.* (2012) provided a list of narrowly distributed Asian pteridophyte taxa towards an assessment of globally threatened species. Out of the total 886 taxa enumerated by them, 577 taxa occur in Southeast Asia, 215 taxa occur in East Asia and 101 taxa occur in South Asia.

Ferns are the most diverse group and the oldest lineage of vascular plants and the second-most speciose after angiosperms with approximately 12000 species. But, currently many pteridophyts extinction are fragmentation, degradation and habitat

destruction, commercial collection, pathogens, predators and invasive species, climate change and pollution. These ferns are not only taxonomic oddities but those are plats with dynamic relationship to their environment. The Western Ghats of Peninsular India is of grate phyto-geographical importance which constitutes one of the 34 global biodiversity hotspot centres, on account of exceptional levels of plant endemism because of its diversified topograpghy and varied climatic conditions. Recently Fracer-Jenkins reviewed pteridophytic numbers to be 1000 species of fern and fern allies in India, Western Himalyas and Western Ghats supported 399 and 349 pteridophytes species of fern and fern allies in India, repectively. In central Western ghats, Karnataka region houses richest pteridophytic diversity (Dudani *et al.,* 2014).

Kumar (1998) documented 159 species from Munnar forest divison. Among them 109 species were terrestrial and 50 epiphytic. A checklist of rare and endangered species found in different forests of Munnar were also provided. Dudani *et al.* (2014) stated that the major families of pteritophytes found in the Western Ghats are Aspleniaceae, Polypodiaceae, Thelypteridaceae, Selaginellaceae, Pteridaceae. Whereas, in the generic level, maximum diversity was observed in the genus *Asplenium, Selaginella, Pteris, Athyrium, Diplazium, etc.* The Western Ghats also harbours endemic species like *Polystichum manickamianum, Cyathea nilgiriensis, Bolbitis semicordata, Selaginella radicata*. Many endangered pteridophytes like *Psilotum nudum, Tectaria zeylanica, Lindsaea malabarica, Cheilanthes rufa.* may soon face the brunt of extinction.

According to Moran (2008), there are 13,600 species of ferns globally, and of this, approximately 1200 species with 70 families and 192 genera are seen in India. In India, there are 1200 pteridophyte species with 70 families and 192 genera. The pteridophyte hotspots in India are the Himalayas, Western Ghats, Eastern Ghats,

Central India, and Andaman and the Nicobar Islands. The Western Ghats occupies only 6% of the India landmass and still holds a pteridophyte diversity of 383 species.

Pteridophytes are the seedless vascular cryptogams which occupy a crucial central position in the evolutionary history of plant kingdom between the lower non-seed bearing and higher seed bearing plants. In India, pteridophytes constitute an important component of flora next to angiosperms (Chandra *et al.*, 2008). Jain and Sastry (1980) reported 17 rare and endangered species of pteridophytes from India.

According to a census, the Pteridophytic flora of India comprises of 67 families, 191 genera and more than 1,000 species (Dixit, 1984) including 47 endemic Indian ferns, less than 10% of those reported previously and 414 species of Pteridophytes (219 At risk, of which 160 critically endangered, 82 Near-threatened and 113 Rare), constituting 41-43 % of the total number of 950- 1000 Pteridophytes of India. Chandra Shubhash (2000) recorded 34 families, 144 genera and more than 1100 species of ferns with about 235 endemic species from Indian region. The vascular flora of our country in general has about 15,000 species and as a constituent of Indian flora of vascular plants, the ferns and fern-allies form only five percent part as far as the number of species is concerned. But, due to their abundance in individuals as well as their conspicuousness in epiphytic vegetation and in the terrestrial vegetation along forest margins roadsides and forest floors, the contribution of ferns and fern-allies to the vegetational pattern in India rank only next to the flowering plants.

In the nineteenth century R. H. Beddome, C.B. Clarke and C.W. Hope produced commendable works on the taxonomy of ferns of Indian subcontinent. A major boost to fern studies in India came with the establishment of the Indian Fern Society in 1983. This organization has helped to bring together the pteridologists of

the country, and is promoting interest in the study of ferns through meetings and through its publications. With the start of Indian Fern Journal in 1984, a channel has been created for communication with international groups of pteridologists. Bir wrote an account of "Pteridology in India" giving details of work done in various fields. Foreign scientists did lot of work on Indian Pteridophytes. Of which R.H. Beddome, C.B. Clarke and C.W. Hope are the pioneers, worked upto the end of 19th century. Beddome's Handbook of the ferns of British India", Ceylon and Malay Peninsula" is the only authentic useful work even to-day. Regional flora on Nagaland (Jamir and Rao, 1988) Meghalaya, (Baishya and Rao, 1981), Tirupura (Singh and Panigrahi, 2005). (Arunachal Pradesh) and North-West Himalayas have already been published. Checklist on pteridophytes of North-East India; Darjeeling and Sikkim Himalayas, Western Himalayas, Mount Abu in Rajasthan and other places viz., Pachmarhi, Tamiya and Patalkot in Madhya Pradesh, Kambab; Shevroy and Pachaku-Tattachi hills and Bombay, Mahabaleshwar, Mather and Kanara in Western Ghats have been published. Dixit (1984) published “A census of Indian pteridophytes” and “Dictionary of Pteridophytes of India” respectively.

Each species of fern has its own preferences of micro habitat depending on the temperature, humidity, soil type, moisture, pH, light intensity, etc., and in many cases are very specific indicators of the conditions they need (Shaikh and Dongare, 2009). It is well observed and noted that most species of ferns succeed under high humidity and shade conditions (Page, 1979), unless they are species that prefer more xeric conditions and are more heliophilous. *Cyathea* sps. and *Angiopteris* sp. are among those perennial ferns which prefer swampy/ moist habitats with low light intensities for their growth. Similarly, another endemic fern *Osmunda huegeliana* prefers the moist and humid banks of free flowing perennial streams and rivers (Dudani *et al.*,

2012). Another endemic fern species *Bolbitis subcrenatoides* is commonly found growing as terrestrial inside the fully covered forest floors and sometimes along road cuttings or edges in the forest. *Pteridium revolutum*syn. *Pteridium aquilinum*is an adventive alien species (Fraser-Jenkins, 2008a) of the region, an escape from gardens into the wild surroundings of Western Ghats, which has a preference for growing gregariously on fully exposed grassy slopes. *Lygodium flexosum* is the only scandent or climbing fern recorded in the current study prefers to grow among the bushes along the partially or fully exposed roadsides. The commonly growing terrestrial species such as *Blechnum orientale* and *Dicranopteris linearis* were observed to be growing abundantly and forming thickets in many places. The common lithophytic fern species in the study region were *Pityrogramma calomelanos, Leucostegia truncata* and *Adiantum philippense* while the common epiphytic fern species observed were *Drynaria quercifolia* and *Lepisorus nudus*.

Most of the pteridophytes recorded in this study have various medicinal and other miscellaneous applications. Among all, *Pteridium revolutum* is perhaps the most widely used terrestrial fern species for various purposes. The tender fronds of this fern are used as vegetables and also in soup preparations while the rhizomes are boiled or roasted and eaten. The rhizome of this fern is astringent, anthelmintic and is useful in diarrhea and inflammation of the gastric and intestinal mucous membranes. The rhizome is boiled in oil and is made into an ointment for wounds. The dried fronds of the fern are also employed as packaging material and have also been tried as a source for paper pulp (Manickam and Irudayaraj, 1992). Another widely occurring fern in the study region – *Tectariaco adunata*, has antibacterial properties and is used in the cases of asthma, bronchitis, honey bee stings and the cooked tender portion of this fern is employed for curing stomach trouble (Dixit and Vohra, 1984; Manandhar,

1996; Das, 1997). The climbing fern *Lygodium flexosum* is used as an expectorant and in the treatment of rheumatism, sprains, scabies, ulcers, eczema and coughs (Singh *et al.*, 1989; Manandhar, 1996).

Other important medicinal applications of the pteridophytes include: use of the rhizome of *Drynaria aquercifolia* as antibacterial, antiinflammatory, tonic, in the treatment of typhoid fever, dyspepsia, cough, diarrhea, ulcers and other inflammations (Dixit and Vohra, 1984; Warrier *et al.*, 1996); use of the fronds of *Osmunda huegeliana* as tonic, styptic and also for the treatment of rickets, rheumatism and for intestinal gripping (Dixit and Vohra, 1984); use of the fronds of *Pityrogramma calomelanos* for the treatment of asthma, cold and chest congestion (Dixit and Vohra, 1984); use of the rhizome of *Leucostegia truncata* as antibacterial and in the treatment of constipation (Benjamin and Manickam, 2007). Besides these exemplary medicinal properties, the pteridophytes have long been greatly valued as ornamentals. They are used to enhance the beauty of the landscape and find their place in gardens, nurseries and during functions for beautification purposes.

Ferns and lycophytes – forming together the paraphyletic pteridophytes – are important components in tropical rainforests globally (Mehltreter, 2010). They are often less recorded in biodiversity surveys compared to timber trees or birds, although several studies demonstrated the high value of ferns and lycophytes as indicators to characterize tropical rainforest biosystems (Mehltreter, 2010; Pouteau *et al*., 2016; Silva *et al*., 2018). Despite the documentation of these plants, they also tend to be less utilized compared to some flowering plant lineage, but their socio-economic importance has been highlighted in several studies focusing on their utilization of ferns in the Philippines (Zamora and Co, 1986; Amoroso, 1990; Buot, 1999; Banaticla and Buot, 2004; Barcelona, 2005; Coritico *et al.*, 2020). Unfortunately, there is little

to no information about these plants occurring at SINP, which inhibits efforts to explore the effectiveness of this protected area for fern and lycophyte conservation.

The pteridophytes tend to increase in number in the north-south direction of Western Ghats, obviously due to the more number of rainy months and higher altitudes with cooler climates. Maharashtra has 64 species of Pteridophytes, most of them confined to northern Western Ghats, (Manickam and Irudayaraj, 2003), Karnataka has about 174 species of pteridophytes, mostly growing in central Western Ghats (Rajagopal and Bhat, 1998) and Kerala and Tamil Nadu together, especially in a block south of Palghat gap alone account for 239 species (Manickam and Irudayaraj, 1992). There have been some notable studies on the pteridophytes of central Western Ghats in Karnataka with the earliest record of 75 ferns species from North Canara district (Matchperson, 1986). After a gap of more than three decades, Blatter and Almeida (1922) included 90 species of ferns from North Canara district in their work 'Ferns of Bombay'. Subsequent studies on pteridophytes of Karnataka included collection of four species of *Selaginella* from the state by Alston (1945); listing of 25 species of pteridophytes by Kammathy *et al.,* (1967); preparation of an artificial key for the 70 species of pteridophytes recorded from Mysore city and its neighbouring areas; inclusion of 10 species of fern family Thelypteridaceae in 'Flora of Hassan District' by Holttum (1976) and inclusion of 12 species of fern by Yoganarasimhan *et al.,* (1981) in 'Flora of Chikmagalur District.

After a significant time gap, there has been a surge in various research aspects of pteridophytes of central Western Ghats. Some of the recent studies on ferns and fern-allies include the record of 23 species of pteridophytes in Madhuguni state forest of central Western Ghats by Deepa *et al.* (2011); enumeration of 22 species of pteridophytes from Agumbe forest of central Western Ghats (Nataraja *et al.,* 2011);

survey and record of 21 species of pteridophytes in the Yana sacred forests of central Western Ghats by Dudani *et al.* (2013) and record of 38 taxa of pteridophytes from Kemmangundi forest of Karnataka by Deepa *et al.* (2013). As it is evident from the available literatures, there is still a serious depth of information pertaining to the pteridophyte diversity of many important biodiversity rich areas of Karnataka. Hence, this study was taken up with the aim of exploring the pteridophytic diversity as a part of an ecological study carried out by our multidisciplinary team during 2010-2011 in the Sakleshpur taluk of Hassan district.

Each species of fern has its own preferences of micro habitat depending on the temperature, humidity, soil type, moisture, pH, light intensity, etc., and in many cases are very specific indicators of the conditions they need (Shaikh and Dongare, 2009). *Pteridium revolutum* syn. *Pteridium aquilinum* is an adventive alien species (Fraser-Jenkins, 2008a) of the region, an escape from gardens into the wild surroundings of Western Ghats, which has a preference for growing gregariously on fully exposed grassy slopes. *Lygodium flexousum* is the only scandent or climbing fern recorded in the current study prefers to grow among the bushes along the partially or fully exposed roadsides. The commonly growing terrestrial species such as *Blechnum orientale* and *Dicranopteris linearis* were observed to be growing abundantly and forming thickets in many places. The common lithophytic fern species in the study region were *Pityrogramma calomelanos, Leucostegia truncata* and *Adiantum philippense* while the common epiphytic fern species observed were *Drynaria quercifolia* and *Lepisorus nudus*. Most of the pteridophytes recorded in this study have various medicinal and other miscellaneous applications. Among all, *Pteridium revolutum* is perhaps the most widely used terrestrial fern species for various purposes. The tender fronds of this fern are used as vegetables and also in

soup preparations while the rhizomes are boiled or roasted and eaten. The rhizome of this fern is astringent, anthelmintic and is useful in diarrhea and inflammation of the gastric and intestinal mucous membranes. The rhizome is boiled in oil and is made into an ointment for wounds. The dried fronds of the fern are also employed as packaging material and have also been tried as a source for paper pulp (Manickam and Irudayaraj, 1992).

Another widely occurring fern in the study region is *Tectariaco adunata*, has antibacterial properties and is used in the cases of asthma, bronchitis, honey bee stings and the cooked tender portion of this fern is employed for curing stomach trouble (Dixit and Vohra, 1984; Manandhar, 1996; Das, 1997). The climbing fern *Lygodium flexosum* is used as an expectorant and in the treatment of rheumatism, sprains, scabies, ulcers, eczema and coughs (Singh *et al.*, 1989; Manandhar, 1996). Other important medicinal applications of the pteridophytes include: use of the rhizome of *Drynaria quercifolia* as antibacterial, anti-inflammatory, tonic, in the treatment of typhoid fever, dyspepsia, cough, diarrhea, ulcers and other inflammations (Dixit and Vohra, 1984; Warrier *et al.*, 1996); use of the fronds of *Osmunda huegeliana* as tonic, styptic and also for the treatment of rickets, rheumatism and for intestinal gripping (Dixit and Vohra, 1984); use of the fronds of *Pityrogramma calomelanos* for the treatment of asthma, cold and chest congestion (Dixit and Vohra, 1984); use of the rhizome of *Leucostegia truncata* as antibacterial and in the treatment of constipation (Benjamin and Manickam, 2007). Besides these exemplary medicinal properties, the pteridophytes have long been greatly valued as ornamentals. They are used to enhance the beauty of the landscape and find their place in gardens, nurseries and during functions for beautification purposes.

However, as the humid places and water bodies are prioritized by the humans for settlements, farming, power generation, setting up of industries etc. the most sensitive pteridophytes depending upon such habitats tend to vanish from the region. Kumar (1998) studied the fern flora of Kerala with special reference to the Sylvan Valley of Munnar. He recorded 159 species of ferns and fern allies belonging to 70 genera and 29 families. It was also found that once abundant species of Munnar forest like *Osmunda hugelina, Angiopteris evecta, Cyathea spp*., *Diplazium spp. Polystichum spp*. have become rare due to deforestation and man-made and natural forest fire.

Kavitha *et al.,* studied the diversity of pteridophytes of Ponmudi hills in 2015. The study area is carpeted by thick tropical forest with a diversity of 28 pteridophyte species. The dominant forest types were tropical evergreen and moist deciduous. Joseph and Thomas (2015) collected 15 chasmophytic pteridophyte species from the Urumbikkara hills of Idukki district of Kerala and, the species belonged to 11 families and 11 genera. Joseph *et al.* (2017) studied the pteridophyte flora of Dr. Salim Ali Bird Sanctuary, Thattekad, Ernakulam; and reported the presence of 30 species of ferns and fern- allies belonging to 23 genera. The sanctuary covers an area of 25.16 km and has tropical evergreen forests, semi evergreen forests, tropical moist deciduous forests, and Mahogany plantations. The pteridophyte flora of Akamala forest station, Thrissur district, kerala was documented by Rekha and Athira (2017). The study area was home to 24 species of pteridophytes. Of these, 2 species belonged to the 'endangered' category, 4 species belonged to the 'rare' category and one species belonged to the 'at risk' category.

Tamil Nadu is the southernmost state of India. It has a geographic area of 1,30,060 km and a forest cover of 26,281 km and that is 20.2% of the total area of the

state (Ruma, 2018).The state has four major geographical divisions such as the Eastern and coastal plains, central uplands, western Karnataka plateau, and the central Eastern Ghats. There are nine types of forests in the state and among these tropical dry deciduous forest occupies 46.98% of the total forest area. The average rainfall that Tamil Nadu receives is 3000- 5000 mm per year. Since Tamil Nadu has both the Western Ghats and the Eastern Ghats passing through it, it holds a rich pteridophyte flora and many studies have been conducted to explore this diversity. Sukumaran *et al.* (2009) recorded.

The diversity of pteridophytes in miniature sacred forests of the Kanniyakumari district in Southern Western Ghats. A total of 24 species were reported from here and out of these 3 were endemic, 3 were endangered and 8 were rare. In a study conducted by Abraham and Ramachandran (2013), six species were added to the pteridophytic flora of Tamil Nadu.

The pteridophyte composition of the Banjalaya forest region was documented by Ashwini and Parashurama (2014). Nineteen pteridophyte species were collected and these belonged to 11 families. *Athyrium hohenackeranum* (Kunze) T. Moore was found to be the most abundant species. The wet evergreen forests of Sakleshpur, which is considered as the 'Hottest hotspot of biodiversity' was surveyed by Dudani *et al.* (2014) and a total of 45 species of pteridophytes were reported from this region. Parashurama *et al.* (2016) assessed the pteridophyte diversity in Mudigere taluk, Central Western Ghats, Karnataka, and a total of 26 species of pteridophytes belonging to 17 families were obtained from the study area. Their habitat was also observed and it was recorded that 22 species were terrestrial.

Maharashtra is a state with a geographic area of 3,07,713 km (Ruma, 2018). Maharashtra has a forest cover of 21% and has mountain ranges with tropical rain

forests. 17% of the state has deciduous forests. There are 3 game reserves, 5 national parks, and 24 bird sanctuaries (Shelar and Madhuri, 2016). 64 fern species have been reported from Maharashtra and most of these are confined to the northern Western Ghats (Manickam and Irudayaraj, 2003). A new addition was done to the flora of Maharashtra by Patil *et al.* (2016). Goa has a geographical area of 3702 km and of this, 2229 km is forest area. 95% of its forest area has 'protected area' status due to the presence of four wildlife sanctuaries. Datar and Laksminarasimham (2010) conducted a study to compile data on the pteridophyte diversity of Goa and concluded that Goa has a pteridophyte flora comprising of 47 pteridophyte species and these belonged to 32 genera under 20 families.

Gujarat has a land area of 1,96,244 km and a forest cover of 14,757 sq km (Ruma, 2018). In 1996, the Gujarat Ecological Commission documented 16 pteridophyte species from different parts of Gujarat. Later few studies were done by Patel *et al.* (2010), Dabgar (2012) and Modi and Dudani (2013). Rajput *et al.* (2016) assessed the pteridophyte diversity of Gujarat through a field study of three years, in which 23 species were collected. It was also noted that *Equisetum debile* was extinct from the wild and *Isoetes coromandeliana* was on the verge of extinction. Eight species were also recorded for the first time in the state was extinct from the wild and *Isoetes coromandeliana* was on the verge of extinction. Eight species were also recorded for the first time in the state.

Kumar (1998) studied the fern flora of Kerala with special reference to the Sylvan Valley of Munnar and recorded 159 species of ferns and fern allies belonging to 70 genera and 29 families. It was also reported that once abundant species of Munnar forest like *Osmunda hugelina, Angiopteris evecta, Cyathea spp., Diplazium spp. Polystichum spp.* etc., have become rare due to deforestation, man-made and

natural forest fires. Kavitha *et al*. (2017) studied the fern allies from Sitheri hills, Eastern Ghats of Tamil Nadu.

Tamil Nadu is the southernmost state of India. It has a geographic area of 1,30,060 km^2 and a forest cover of 26,281 km^2 and that is 20.2% of the total area of the state (Talukdar, 2018).The state has four major geographical divisions such as the Eastern and coastal plains, central uplands, western Karnataka plateau, and the central Eastern Ghats. There are nine types of forests in the state and among these tropical dry deciduous forest occupies 46.98% of the total forest area. The average rainfall that Tamil Nadu receives is 3000- 5000 mm per year.

Since Tamil Nadu has both the Western Ghats and the Eastern Ghats passing through it, it holds a rich pteridophyte flora and many studies have been conducted to explore this diversity. Sukumaran *et al.* (2009) recorded the diversity of pteridophytes in miniature sacred forests of the Kanniyakumari district in Southern Western Ghats. A total of 24 species were reported from here and out of these 3 were endemic, 3 were endangered and 8 were rare.

In a study conducted by Abraham and Ramachandran (2013), six species were added to the pteridophytic flora of Nilgiris viz. *Asplenium bipinnatum* Roxb. (Aspleniaceae), *Cheilanthes viridis* Sw. (Pteridaceae), *Huperzia phlegmaria* Rothm. (Lycopodiaceae), *Selaginella ciliaris* (Retz.) Spring. (Selaginellaceae), *Selaginella intermedia* (Blume) Spring. (Selaginellaceae) and *Trichomanes bipunctatum* Poir. (Hymenophyllaceae).

Sathish and Vijayakanth (2016) too added six fern species to the recorded fern flora of Kolli hills in Tamil Nadu. The six species added were *Adiantum latifolium, Oleandra musifolia, Diplazium cognatum, Bolbitis appendiculata,*

Leptochilus thwaitesianus, and *Phymatosorus membranifolium.* And in 2017, Vijayakanth *et al.,* added two new ferns to the fern flora of Tamil Nadu. These ferns are *Athyrium parasnathense* of family Athyriaceae and *Leptochilus metallicus* belonging to Polypodiaceae. Kumari and Jeeva (2018) studied the pteridophytes along the Thamiraparani River in Tamil Nadu and reported the presence of 65 pteridophytes species alongthe Thamiraparani river and in this 33% were terrestrial, 12% were aquatic, 11% were lithophytes and 13.8% wereepiphytes.

Alagesaboopathi *et al.* (2018) documented 14 species of pteridophytes from the Kanjamalai Hills of Salem District. A study on thepteridophyte diversity of the Kilavarai freshwater river in Kodaikanal was conducted (Packiaraj and Suresh, 2019) and, 36 species belonging to 25 genera distributed among 19 families were reported. The dominant species belonged to Adiantaceae, Polypodiaceae, Pteridaceae, and Cheilanthaceae families.

Karnataka has a major portion of the Central Western Ghats and also hosts a large portion of its endemism. Most of the pteridophytes species are seen in the central Western Ghats (Rajagopal and Bhat, 1998).The studies on the pteridophyte diversity began with the work by Blatter and D'almeida (1922) and 75 species were recorded in the 'Ferns of Bombay'. Followed by this many studies were conducted, Alston (1945) recorded four species of *Selaginella*; Kammathy *et al.*, (1967) listed 25 species; Holttum (1976) included 10 fern species from the family Thelypteridaceae in 'Flora of Hassan District' and Yoganarasimham *et al.,* (1981) added 12 species in 'Flora of Chikmangalur District'. Match person recorded 90 fern species from the North Canara district (Matchperson, 1986).

Again after a significant time gap, more research was being conducted to

provide detailed data on the pteridophyte diversity of Karnataka. Deepa *et al.* (2013b) studied the distribution of pteridophytes in the Kigga forest of Central Western Ghats in Karnataka. The species diversity was calculated using Shannon's diversity index and Simpson's diversity. *Aleuritopteris anceps* (Blanf.) Panigrahi was the most abundant species in the studied area. Deepa *et al.* (2017) later enumerated 23 pteridophytes in Madhuguni forest of Central Western Ghats in Karnataka. The majority of the pteridophyte species were terrestrial with an exception of two epiphytes, one aquatic and one climbing fern.

The pteridophyte composition of the Banjalaya forest region was documented by Ashwini and Parashurama (2014). 19 pteridophyte species were collected and these belonged to 11 families. *Athyrium hohenackeranum* (Kunze) T. Moore was found to be the most abundant species. The wet evergreen forests of Sakleshpur, which is considered as the 'Hottest hotspot of biodiversity' was surveyed by Dudani *et al.*, (2014) and a total of 45 species of pteridophytes were reported from this region. Parashurama *et al.* (2016) assessed the pteridophyte diversity in Mudigere taluk, Central Western Ghats, Karnataka, and a total of 26 species of pteridophytes belonging to 17 families were obtained from the study area. Their habitat was also observed and it was recorded that 22 species were terrestrial.

Conservation statuses of Western Ghats pteridophytes

The study and recording of endemic pteridophytes of India were first done by Chandra (1982). In his study, 96 species were recorded and in a later study with Kaur and Chandra (1994), 41 more species were reported. According to Dixit (1984), 214 pteridophyte species are endemic to India. In 2008b, Fraser-Jenkins removed many endemic species, as these were pseudo- endemics and was mistaken

by pteridologists due to their synonyms and lack of species understanding. Currently, a total of 49 pteridophytes are recognized as endemic to India. The majority of the endemic pteridophytes (33 species) are reported from Deccan Peninsula and the Western Ghats. Some of the endemic species in the Western Ghats are *Polystichum manickamii, Cyathea nilgiriensis, Bolbitis semicordata, Selaginella radicata* etc. Endangered species in this region include *Psilotum nudum, Tectaria zeylanica, Lindsaea malabarica, Cheilanthes rufa*.

According to Fraser-Jenkins (2012), out of the total 1100 indigenous pteridophytes of India; 337 species are considered to be threatened or endangered. The conservation statuses of the pteridophytes were determined based on the IUCN (2010) listing. According to this study,12 species were Extinct (EX), 4 were Extinct in the wild (EW), 95 were critically endangered (CR), 117 were Endangered (EN), 67 were Vulnerable (V) and 43 were nearthreatened (NT).

OBJECTIVES

The present study was carried out with the following objectives

i) To document the presence of variety of pteridophytes in the form of different taxas.

ii) To rationalize a relationship between diversity and the similarity of ferns in the study area.

iii) To analyze the further distribution of species in the study area categorized under IUCN.

MATERIALS AND METHODS

Study area:

The study region was mainly confined to the collection along the roadside water bodies enroute Enayam to Marthandam, Kanniyakumari district, Tamil Nadu, India. The geographical co-ordinates of the locations surveyed were noted down using pre-calibrated GPS. The field collection was carried out in and around the location with Lat 8.236932, Long. 77.214392 to Lat 8.310582, Long 77.218474.

Survey methodology:

Intensive field exploration was done enroute from Enayam to Marthandam, Kanniyakumari district during November 2022 to March 2023, to document and collect ferns and fern allies. For some of the common, locally uncommon and valuable species of pteridophyte in the study area, photographs alone were taken.

Identification of specimens:

The taxa were identified using appropriate floras, journals, monographs and revisions and verified with the help of illustrations and Floras, particularly Ferns of southern India (Beddome, 1864); Ferns of British India (Beddome, 1866) Pteridophyte Flora of Western Ghats (Manickam and Irudayaraj, 1992); Pteridophytes of the Western Ghats A Pictorial Guide (Benniamin and Sundari, 2020). The identities of some doubtful specimens were verified with the help of eminent pteriodologists. The specimens were then dried, processed and labelled by standard herbarium method given by Jain and Rao (1977). Field observations such as habitat, ecology and distribution were also noted. The processed specimens of all taxa collected were incorporated in the PG and Research Department of Botany, St. Mary's College

(Autonomous), Thoothukudi for future reference. Special emphasis was given to the occurrence of pteridophytes categorized under IUCN pteridophytes as evaluated by Fraser-Jenkins (2012).

RESULTS

The current study reveals the distribution of 24 species (Table – 1, Plate - I) of pteridophytes along the study area enroute from Enayam to Marthandam, Kanniyakumari district, Tamil Nadu. Twenty four species of pteridophytes belongs to ten families namely Pteridaceae, Thelypteridaceae, Gleicheniaceae, Davalliaceae, Nephrolepidiaceae, Salviniaceae, Azollaceae, Linsaeaceae, Masileaceae and Hymenophyllaceae. The botanical description is given below.

***Azolla pinnata* subsp. *asiatica* R.M.K. Saunders & K. Fowler**

Common name: Mosquito fern

Description:

Azolla pinnata subsp. *asiatica* is small, 1.5-2.5 cm long, with a straight main axis with pinnately arranged side branches, progressively longer towards the base, thus roughly triangular in shape; the basal branches themselves becoming pinnate and eventually fragmenting as the main axis decomposes to form new plants. Roots have fine lateral rootlets, giving a feathery appearance in the water. Leaves minute, 1overlapping in two ranks, upper lobe green, brownish green or reddish, lower lobe translucent brown; minute, short, pillae, cylindrical unicellular hairs often present on the upper lobes.

***Davallia repens* (L.f.) Kuhn**

Common name: Dwarf Hares-Foot Fern

Description:

Rhizome slender, white-waxy, densely covered with appressed red-brown scales with pale scarious margins bearing fragile deciduous hairs. Fronds in Australian material generally simple and pinnatifid, narrowly triangular, coriaceous. Stipe 0.5–12 cm long, bearing similar scales to rhizome; rachis with scattered scales

Table 1 – Distribution of pteridophytes in the Kamamkuli pond

S. No	Name of the species	Family	Habitat	Distribution in site of collection	IUCN Category
1.	*Davallia repens* (L.Fil) Kuhn	Davalliaceae	Terrestrial	++	VU
2.	*Lygodium flexousum* (L.) Sw.	Gleicheniaceae	Terrestrial	+	-
3.	*Trichomanes obscurum* Blume.	Hymenophyllaceae	Terrestrial	++	-
4.	*Nephrolepis multiflora* (Roxb.) Jarret	Nephrolepidiaceae	Terrestrial	+++	-
5.	*Adiantum caudatum* L.	Pteridaceae	Terrestrial	+++	-
6.	*Adiantum latifolium* Lam.	Pteridaceae	Terrestrial	+++	-
7.	*Adiantum philippense* L.	Pteridaceae	Lithophytic	+	-
8.	*Ceratopteris thalictroides* (L.) Brongn	Pteridaceae	Aquatic	++	-
9.	*Mickelopteris cordata* (Roxb. Ex Hook. & Grev.) Fraser-Jenk	Pteridaceae	Terrestrial	++	-
10.	*Pityrogramma austroamericana* Domin	Pteridaceae	Terrestrial	++	-
11.	*Pityrogramma calomelanos* (L.) Link	Pteridaceae	Terrestrial	++	-
12.	*Pteris vittata* L.	Pteridaceae	Terrestrial	++	-
13.	*Salvinia molesta* D.S. Mitch.	Salviniaceae	Aquatic	+++	-
14.	*Thelypteris articulata* (Houlston & T.Moore) Tagawa & K.Iwats	Thelypteridaceae	Terrestrial	++	-
15.	*Thelypteris dentata* (Forssk.) E.P. John.	Thelypteridaceae	Terrestrial	++	-
16.	*Thelypteris ochthodes* (Kunze) Ching.	Thelypteridaceae	Terrestrial	++	-
17.	*Thelypteris paludosa* (Blume) K. Iwats	Thelypteridaceae	Terrestrial	++	EN

PLATE - I

Azolla pinnata subsp.asiatica

Davallia repens

Lygodium flexousum

Trichomanes obscurum

Lindsaea ensifolia

Marsilea quadrifolia

Nephrolepis multiflora

Acrostichum aureum

PLATE - II

Adiantum caudatum

Adiantum latifolium

Adiantum philippense

Ceratopteris thalictroides

Mickelopteris cordata

Pityrogramma austroamericana

Pityrogramma calomelanas

Pteris vittata

PLATE - III

Pteris ensiformis

Salvinia natans

Salvinia molesta

Thelypteris articulata

Thelypteris dentata

Thelypteris ochthodes

Thelypteris paludosa

Thelypteris interrupta

Figure - 1 Percentage distribution of families of pteridophytes

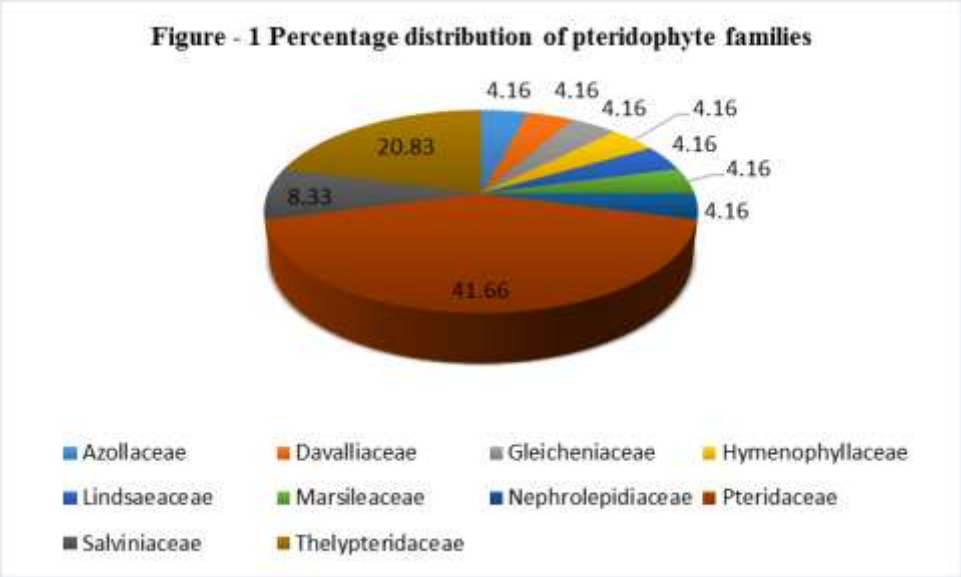
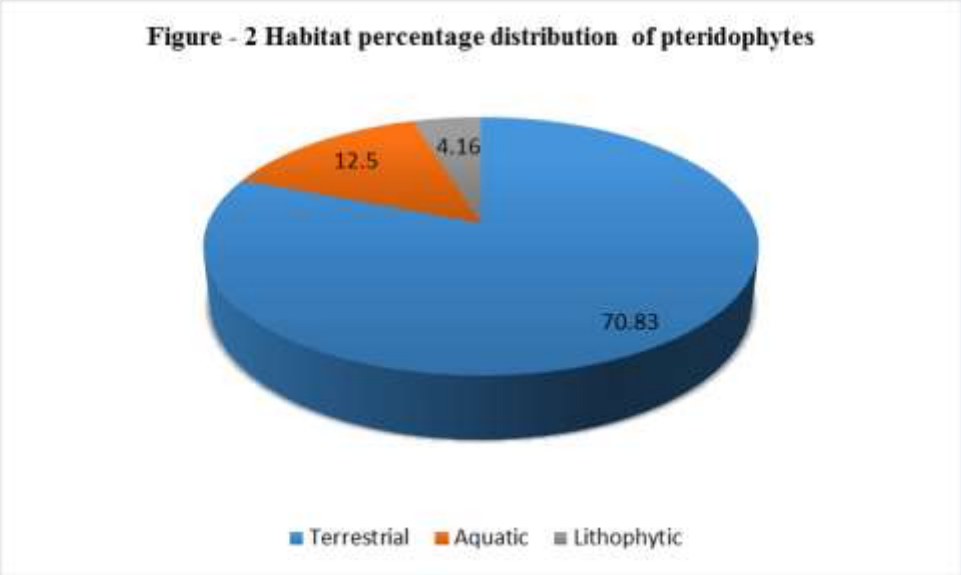


Figure - 2 Habitat percentage distribution of pteridophytes



on lower surface, glabrous on upper surface. Pinnae deeply lobed almost to rachis; widest point 8–85 mm; upper surface glabrous; lower surface rarely with a few dark simple hairs; margins highly variable in degree of lobing but basal lobe pair always distinct, usually larger and asymmetrical with much deeper lobing on the basal side; apex rounded on sterile lobes to acuminate on fertile lobes; false veins absent. Sori marginal, grouped near apices of pinna lobes, mostly in sinuses formed by a marginal tooth projecting from basal side of sorus; indusium generally elongate, attached at a narrow base, apex extending beyond margin.

***Lygodium flexousum* (L.) Sw.**

Common name: Maidenhair Creeper

Description:

Terrestrial herbs with long creeping rhizome, 5-8 mm thick, with fibrous roots at base; densely dark brown hairy, hairs 1 mm long, multicellualr, uniseriate, tubular. Fronds pale green, 4-5 m long, climbing, tripinnate; stipe stout, dark brown, densely hairy at base, strminaceous, glabrous above, wiry, 3-5 mm thick, rounded beneath, flattened above; primary pinnae alternate, stalked, forked once, with dormant bud on axis, each forked branch with 2-3 pairs of simple or forked pinnules alternately; pinnules oblong-lanceolate, simple or forked, or auriclate, apex subacute, acute or acuminate, base cuneate in simple pinnules, subtruncate or cordate in branched or forked pinnules; margin regularly or irregularly serrulate in sterile pinnules; costa raised above and below; veins distinct above and below, forked twice or thrice, free, reaching the margin, axis of main branches and costa pubescent; sporangia yellowish-brown, crowded on fingerlike lobes of fertile pinnae, lobes 3 x 1.5 mm, sporangia five pairs, alternate, indusiate. Spores 48 µm in diameter, yellowish-green (Plate).

***Trichomanes obscurum* Blume.**

Description:

Terrestrial herb with erect, or short creeping, dark or reddish brown, stiff hairy rhizome. Fronds tripinnate, dark bluish green; stipe long, hairy; lamina ovate-acuminate in outline; primary pinnae elliptic in outline; secondary pinnae lanceolate in outline; pinnules linear, dissected, acute; veins indistinct. Sori dark brown, confined to basal acroscopic segments of pinnules; indusia cup-shaped, narrowly winged; truncate at apex, receptacles exerted. Sporangial capsule subglobose. Spores globose, trilete, finely granulose.

***Lindsaea ensifolia* Sw.**

Description:

Rhizome short- to long-creeping; scales narrowly triangular, pale reddish brown. Fronds monomorphic. Stipes distant, 10–35 cm long, usually shorter than lamina, stramineous to reddish brown. Lamina 1-pinnate, narrowly oblong, narrowly ovate or linear; Pinnules spreading to strongly ascending, narrowly ovate to linear, only moderately reduced towards apex; base asymmetrical; apex acute to obtuse, sometimes acuminate. Terminal pinnule similar in shape and size to laterals, usually free, occasionally connected with 1 or 2 uppermost lateral pinnules; midrib distinct. Sori continuous; indusium wide, entire. Spores trilete, pale brown.

***Marsilea quadrifolia* L.**

Common name: Water clover or pepperwort

Description:

Marsilea quadrifolia, is an aquatic fern that grows from creeping rhizomes anchored in the muddy bottoms of shallow ponds and lakes. From creeping rhizomes, thin green stalks rise to the water surface, each stalk bearing a single shamrock-like leaf with four wedge-shaped leaflets. Leaves usually float on or just below the water

surface, however in very shallow water the stalks often are able emerge above the surface.

***Nephrolepis multiflora* (Roxb.) Jarret**

Common name: Asian sword fern

Description:

Stem scales appressed, bicolored with margins transparent. Tubers absent. Petiole moderately to densely scaly; scales appressed, dark brown with pale margins. Blade sparsely to moderately scaly, hairy abaxially, hairs pale brown. Scales scattered to dense, brown, margins pale. Central pinnae narrowly deltate, sometimes elliptic, base rounded basiscopically, slightly auriculate to truncate acroscopically, acroscopic lobe acute to oblong, margins biserrate to irregularly serrate to serrulate, apex attenuate and occasionally slightly falcate; costae adaxially densely hairy, hairs pale, erect. Indusia circular to horseshoe-shaped.

***Acrostichum aureum* L.**

Common name: Leather Fern

Description:

Acrostichum aureum is a mangrove fern, growing up to 3 m tall, and thrives well under full sun. The stem (rhizome) of this species is stout, erect, and covered with relatively large scales. Dark green, erect fronds are composed of 24 - 30 pinnae in alternate arrangement. The petiole of the frond is usually about 1 m long. The 5 - 8 pairs of pinnae near the tip are fertile with their underside covered in reddish brown sporangia (except the midrib), while the remaining pinnae are infertile. The pinnae are oblong with a blunt tip and bright red when immature. Fibrous rhizomes have a scaly surface.

***Adiantum caudatum* L.**

Common name: Walking fern

Description:

Rhizome erect, short; scales concolorous, golden brown, with entire margins. Fronds tufted, long. Stipe long, dark brown, hairs multicellular and dark brown to brown. Lamina 1-pinnate, lanceolate in outline; rachis dark brown, hairs multicellular and dark brown to brown. Apex usually prolonged into a whiplike stolon rooting at tip to form new plantlet. Pinnules alternate, or lower ones subopposite, horizontally spreading or slightly obliquely spreading; lower pinnules gradually reduced, suboblong; brownish green; both surfaces sparsely multicellular hirsute; base asymmetrical; lower margins entire, upper margins deeply divided into many narrow lobes; lobes linear, margins entire, upper part again lobed into fine linear segments. Sori 5–12 per pinna; false indusia dark brown, orbicular or oblong, hairy, upper margins flat and straight, entire, persistent (Plate - II).

***Adiantum latifolium* Lam.**

Description:

Terrestrial herb with long creeping, densely scaly rhizome, 3-4 mm thick. Scales lanceolate or linear, very narrow pointed at apex, dark reddish brown, clathrate, entire or with few outgrowths. Fronds 40-55 x 20-28 cm; stipe 25-36 cm long, black polished, scaly beneath, hairy above; lamina deltoid in outline, bipinnate; primary pinnae, oblong, lanceolate in outline, 5-6, alternate, largest in middle; pinnules obtuse to rounded at apex, serrate; sessile or subsessile, acroscopic base truncate, basiscopic base cuneate, excised, coriaceous; veins very close, repeatedly forking, anastomosing, largest pinnae in the middle, terminal pinnae rhomboid, larger.

Sori oblong or elliptic, yellowish brown. Sporangial capsule, subglobose. Spores triangular, yellowish, verrucoid.

***Adiantum philippense* L.**

Common name: Maidenhair Fern

Description:

Terrestrial herb with erect, rhizome. Scales linear or lanceolate, acuminate, dark brown to black, clathrate, entire. Fronds simply pinnate; stipe 17-23 cm long, scaly at base, pinkish-brown polished above, lamina 30 x 8-10 cm, lanceolate in outline; pinnae, semicircular, acroscopic margin shallowly lobed, basiscopic base entire, coriaceous or subcoriaceous; veins forked to anastomosing. Sori linear 0.6-2 cm, dark brown, indusia brownish. Sporangial capsule subglobose, stalk 250 µm long. Spores brownish yellow, planoconvex, thinly granulose.

***Ceratopteris thalictroides* (L.) Brongn**

Common names: And water Horn fern, Indian fern, Oriental wate rfern, Water fern, Water sprite

Description:

Aquatic or semi aquatic herbs with erect rhizome, 4 x 2-3 cm, with thick long, fibrous, fleshy roots. Scales pale brown, membranous, ovate or elliptic, acute, entire. Fronds dimorphic; stipe 15-20 cm long; sterile lamina simply pinnate; pinnae ovate or deltoid in outline, variously lobed, coriaceous, fleshy, glabrous, veins anastomosing, indistinct; fertile lamina simply pinnate, pinnae tripinnatifid, lobes linear, fleshy. Sporangial capsule sessile. Spores trilete with thickly folded ridges.

***Mickelopteris cordata* (Roxb. Ex Hook. & Grev.) Fraser-Jenk**

Common name: Heart fern

Description:

Extremely attractive fern is characterized by its heart-shaped leaves, which are also covered with numerous dark hairs. The leaves sit on long black-brown petioles. In addition, it is dimorphic and, above a certain size, forms not only its trophophylls for photosynthesis but also sporophylls for spore production, which are characterized, among other things, by significantly longer petioles. In its preferred habitats in Southeast Asia it grows both as epiphytic and terrestrial in loose substrate congregations.

***Pityrogramma austroamericana* Domin**

Description:

Terrestrial herbs with erect rhizome, 3-5 x 2-3 cm. Scales lanceolate, long acuminate, pale brown. Fronds 20-30 x 5-10 cm, bipinnate; stipe 8-10 cm long, scaly beneath, dark brown polished, grooved above; lamina ovate in outline; pinnae lanceolate, acuminate in outline, progressively reduced towards apex; pinnules lanceolate, acute, margins serrate, basal pinnules auricled, progressively reduced towards apex, glabrous above, covered by yellow powder below. Sori acrostichoid when mature. Sporangial capsule globose. Spores dark brown, tetrahedral, trilete, granulose.

***Pityrogramma calomelanos* (L.) Link**

Common name: Silver Fern, Silverback Fern

Description:

Terrestrial herb with erect, densely scaly rhizome. Scales, brownish, linear, entire. Fronds bipinnate; stipe, dark-pinkish brown, polished; lamina triangular in outline; pinnae, lanceolate, acuminate in outline, pinnules rhomboidal to lanceolate, acute, lobed to serrate, pinnae and pinnules progressively reduced to apex, rachis and costa grooved above, raised below; lower surface of pinnules white crusted. Sori

acrostichoid. Sporangial capsule, globose, stalk 250 µm long. Spores triangular in outline, yellowish with pinkish thickenings.

***Pteris ensiformis* Burm.f.**

Common name: Slender bracken

Description:

Short slender terrestrial fern. Rhizome short creeping to suberect, scaly; scales narrowly triangular, brown. Fronds dimorphic. Stipe to 200 mm long, pale green; base scaly. Lamina 1-pinnate, with pinnae increasing in length from apex to base. Sterile lamina; lower pinnae pinnate to pinnatifid, oblong to ovate, irregularly serrate. Fertile lamina to 300 mm long, narrower than sterile; pinnae forked near their base; ultimate segments linear to narrowly oblong, serrate only at their apices. Veins free, oblique, 1-2-forked. Sori continuous from near base to near apices of fertile pinnae or pinnules; paraphyses scarce.

***Pteris vittata* L.**

Common name: Chinese brake fern or ladder brake fern

Description:

Rhizome suberect, densely scaly at apex; scales ovate lanceolate, entire, acuminate, up to 3×1mm, pale brown; stipes bent at base 6 × 0.1cm ,round at the abaxial surface, grooved at the adaxial side, pale brown, scaly at the base. Lamina lanceolate, simply pinnate, narrowing towards the base, terminated by a pinna larger than the lateral ones; pinnae up to 1cm apart, lower ones opposite, upper ones sub-opposite, sessile, ascending, middle pinnae up to 6×0.7cm, terminal one up to 10× 0.5cm, base cuneate or truncate, apex acuminate, margin serrate in the non-soral part, entire in the rest, veins obscure, forked once from or above the base. Sori linear, continuous along the margin except the apex (Plate - III).

***Salvinia molesta* D.S. Mitch.**

Common name: Kariba weed, African payal

Description

Aquatic free floating ferns. Stem spongy, terete, up to 2mm thick, brown, branched with nodes and internodes, submerged leaves modified into root-like organs, up to 5cm long, covered by brown hairs. Normal leaves born at the nodes in two opposite pairs, erect floating, sessile ovate to oblong, entire, pale green, lower surface glabrous, upper surface with dense hairs born on the intervenal areas, stiff, erect with a common stalk, branched into four, separate hooked branches; veins slightly distinct below, anastomosing, areoles parallel, elongated; herbaceous in texture; normal leaves spongy due to the presence of hairs; sporocarps born in clusters on submerged leaves.

***Salvinia natans* (Linn) All.**

Common name: floating fern, floating watermoss, floating moss

Description:

Floating fronds sessile, base rounded or subcordate, margin entire, apex obtuse; lateral veins on each side of costa, each one with 5-8 low dome-shaped papillae, each with a terminal bunch; lamina deep green on upper surface, densely brown villous on lower surface; submersed fronds finely dissected into linear segments, covered with hairs, and acting as roots.

***Thelypteris articulata* (Houlston & T.Moore) Tagawa & K.Iwats**

Description:

Terrestrial fern. Rhizomes erect, sparsely scaly at the apex. Scales basifixed, ovate, acuminate, base broad, margin glandular hairy, pale brown, concolorous. Fronds tufted, simple pinnate; stipes scaly at base above glabrous, acicular hairy; lamina lanceolate, glabrous above, hairy below, dark green, with a terminal pinna similar to lateral ones; texture sub coriaceous; pinnae upto 12 pairs, sub opposite or

alternate, sessile or sub sessile, margin shallowly lobed, apex acuminate, acroscopic base broad cuneate to truncate, basiscopic base cuneate, lower 2-3 pairs of pinnae slightly deflexed; pinnae lobes cut down one fourth way to the costa, segments 15-20 pairs, acute, slightly ascending. Veins distinct, anastomosing, upper 2-3 pairs free, lower surface sparsely hairy. Sori round, small, median on veins, arranged in two rows; indusia glabrous; sporangia bearing club shaped glandular hairs on stalk; spores monolete, pale brown.

***Thelypteris dentata* (Forssk.) E.P. John.**

Common name: Downy maiden fern

Description:

Terrestrial fern. Rhizomes suberect or shortly creeping, apices densely scaly. Scales light-brown linear-lanceolate, acuminate apex, margin entire. Fronds subclustered; stipe bases covered with scales, dark brown, stramineous distally; lamina simple pinnate, bases slightly narrowed, apices acuminate, brownish green when dried, shortly hairy adaxially, densely puberulent abaxially, sometimes with glandular hairs, texture herbaceous to papery; lateral pinnae 15–20 pairs, proximal 2 to 3 pairs progressively shortened; pinnae lanceolate to oblanceolate, sessile, subopposite, bases rounded-truncate, apices acuminate; pinnae lobes more than half way cut down to the costae, segments 15–25 pairs, oblong, slightly oblique, basal acroscopic one slightly longer, rounded-obtuse at apices, margin entire. Veinlets 6–8 pairs per segment, proximal pair anastomosing, next pair running to sinus membrane, intervenal area with several acicular hairs,. Sori orbicular, arranged in two rows on both side of the costule; indusia shortly hairy, brown. Spores monolete, irregularly cristate.

***Thelypteris ochthodes* (Kunze) Ching.**

Description:

Terrestrial herbs. Rhizome short creeping or suberect, rarely erect, up to 6 cm thick; scales broadly ovate, about 4 x 5 mm, uniformly pale brown, apex acuminate, margin entire or with few small elongate outgrowths. Stipes up to 105 x 1 cm, dark brown, sparsely scaly at base, grey brown or purple-brown, glabrous above, rounded below, grooved above, three or four pairs, dark brown aerophores present along each side of the stipe. Lamina ovate or lanceolate, terminating with a pinna having larger lobes at the basal part, acuminate, pinnae up to 35 pairs, sessile, subopposite or alternate, about 10 pairs of basal pinnae abruptly reduced to tubecles, pinnae in the distal part of the lamina progressively reduced; rachis tetragonal, grooved on each side, except the lower side; largest pinna, linear lanceolate, apex acuminate, base truncate or subtruncate, margin lobed; lobes oblong, apex acute or rounded, entire; costa distinctly raised, rounded below; pinnae dark green above, pale green below; texture subcoriaceous; densely hairy, hairs long, soft, sender, pale brown acicular, margin of the lobes bears few short, stiff acicular hairs. Sori supra-median on each vein, except few pairs of distal part of the lobe, yellowish-green; indusia with few acicular hairs; spores, yellowish-green, exine densely, coarsely tuberculate.

***Thelypteris paludosa* (Blume) K. Iwats**

Common name: Marsh fern

Description:

Perennial fern has erect to ascending compound. Fertile leaves tend to be a little smaller in size than infertile leaves; they are both deciduous and die down during the winter. The compound leaves are pinnate-pinnatifid in structure and lanceolate to lanceolate-oblong in outline; their petioles are pale tan or pale purplish tan and mostly glabrous. The blade tissue of these leaves is light green and hairless on both the upper

and lower sides. In contrast, the central stalk of the compound leaf and the lateral stalks of the pinnatifid leaflets are finely pubescent on their lower sides. Each compound leaf has 10-40 pairs of leaflets; these leaflets are deeply pinnatifid and narrowly lanceolate-oblong. Often, a compound leaf and its leaflets are slightly curved and twisted. The lobes of the leaflets are oblong or oblong-lanceolate in shape, while their margins are smooth and strongly involute (curved downward). The lobes are spaced close together along the length of each leaflet. On the lower surface of each lobe, there is a central vein with several lateral veins. Each lateral vein becomes forked and divides into two veins.

***Thelypteris interrupta* (Wild.) K.Iwats.**

Common name: Hottentot fern, Willdenow's fern

Description:

Stems long-creeping, cordlike. Leaves monomorphic, evergreen. Petiole straw-colored to tan, scaleless. Blade broadest at base, gradually narrowed distally to pinnatifid apex. Pinnae segments deltate, rounded to acute; proximal pair of veins from adjacent segments united at acute or obtuse angle below sinus, with excurrent vein. Indument abaxially of hairs on costae and veins, or hairs often lacking, costae also with tan, ovate scales; veins, costules, and costae adaxially glabrous or sparsely pubescent; blade tissue without hairs on both sides, or hairy abaxially, usually with red to orange, shiny, sessile, hemispheric glands abaxially. Sori round.

The genus the *Thelypteris* was recorded with five different species like *Thelypteris articulata, Thelypteris dentata, Thelypteris ochthodes, Thelypteris paludosa* and *Thelypteris interrupta.* Whereas the genus *Adiantum* was reported with three species namely *Adiantum latifolium. Adiantum caudatum* and *Adiantum*

philippense. The genus *Pityrogramma* was recorded with two species namely *Pityrogramma austroamericana* and *Pityrogramma calomelanos.* Family pteridaceae was reported with ten different species and six different genus. Families like Azollaceae, Devalliaceae, Gleicheniaceae, Hymenophyllaceae, Lindsaeaceae, marsileaceae, and nepholepidiaceae, and were represented by a single genus

Most of the pteritophytes recorded were terrestrial in its habit. *Adiantum philippense*.one was lithophytic *Azolla pinnata*, subsp. *asiatica*, *Marsilea quadrifolia*, *Ceratopteris thalictroides*, and *Salvinia natans* were aquatic. *Lygodium flexousum* was the only climber in habit.

Azolla pinnata subsp *asiatica, Nephrolepis multiflora Adiantum caudatum Adiantum latifolium Salvinia natans, Salvinia molesta Thelypteris interrupta* were dominantly reported in the site of study species like *Davallia repens, Marsilea quadrifolia, Ceratopteris thalictroides, Mickelopteris cordata, Pityrogramma austromericana, Ptryogramma calomelanos, Pteris vittata, Thelypteris articulata , Thelypteris dentata, Thelypteris ochthodes* and *Thelypteris paludosa* were recorded in least. *Lygodium flexousum, Acrostichum aureum* and *Adiantum philippense* were distributed meagerly. In the current investigation the families Azollaceae, Davalliaceae, Gleicheniaceae, Hymenophyllaceae, Lindsaeaceae, Marsiliaceae and Nephrolepidiaceae the percentage is 4.16 Pteridaceae (41.66), Salviniaceae (8.33) and Thelypteridaceae (20.83) (Figure – 1). Terrestrial habitat of pteridophytes is (70.83), aquatic (4.16) (Figure – 2).

IUCN has categorized certain pteridophytes based on their distribution. In the present study two species of pteritophytes were categorized under IUCN was reported. Of the 24 species of pteritophytes collected only two species were

catagorized under IUCN. *Davallea repens* was catagorized as vulinerable by IUCN where us *Thelypteris paludosa* was categorized as endangered under IUCN.

DISCUSSION

The tropical wet evergreen and semi-evergreen vegetation along road side encompass a wide array of floristic diversity which includes angiosperms, pteridophytes, bryophytes and fungi. The presence of many perennial streams, waterfalls and other moist habitats support rich growth of pteridophytes. The current investigation on the distribution of pteridophytes enroute Enayam to Marthandam, Kanniyakumari district, Tamil Nadu reveals the distribution 24 different species of pteridophytes belonging to ten different families. The study gives a broad outlook about the pteridophytes of the study area where 24 pteridophytes were encountered. Majority of the pteridophytes belong to the family Pteridaceae (10), followed by Thelypteridaceae (5) and the remaining families are Azollaceae (1), Davalliaceae (1), Gleicheniaceae (1), Lindsaeaceae (1), Marsiliaceae (1), Nephrolepidiaceae (1), Salviniaceae (1), Hymenophhyllyaceae (1). Among 24 pteridophytes 17 are terrestrial, 2 are aquatic and 1 climber. At that place the pteridophytes like *Adiantum caudatum*, *Adiantum latifolium, Nephrolepis multiflora,* and *Salvinia natans* are commonly distributed.

The 24 species recorded belongs the different families like Azollaceae, Davalliaceae, Gleicccheniaceae, Hymenophyllaceae Lindsaeaceae, Marsilieaceae, Nephrolepidiaceae, Pteridaceae, Salviniaceae and Thelypteridaceae. Among the families the Pteridaceae was documented with diverse species, whereas family Thelypteridaceae was reported with five species. Family Salviniaceae was recorded with two species. All the members of the genus *Thelypteris* were terrestrial in habit though they were terrestrial this members were documented along the water banks for survival. Water bodies which contain moisture content is the driving force behind diversity. The other families like Azollaceae, Davalliaceae, Gleicheniaceae,

Hymenophyllaceae, Marsileaceae and Nephrolepidiaceae was documented with single species. The diversity in the genus Thelypteridaceae was observed which might to due to the environmental hues which favoured the survival. This followed by genus *Adiantum* which was recorded with three species.

Among the members of the genus *Thelypteris paludosa* was enlisted under endangered category by IUCN. *Thelypteris interrupta* was growing in wild occupying the entire most land. The rhizome were very aggressive in its growth whereas the other species of *Thelypteris dentata, Thelypteris ochthodes* and *Thelypteris paludosa* were occasional.

Adiantum caudata and *Adiantum latifolium* were terrestrial in its habitat whereas *Adiantum philippense* was lithophytic in habit. *Adiantum caudatum* and *Adiantum latifolium* was dominantly reported. In the study area *Adiantum latifolium* was of rapidly colonising the study area and it was occupying the entire land which might be due to scattering of fragments of rhizomes

Nephrolepis multiflora which attracted the by passers with its beautiful fronds were dominantly documented on the barren land along with grasses as weeds. *Nephrolepis multiflor*a belongs to the family Nephrolepidiaceae and it was represented by a single genus. The species was growing abundantly due to its aggressive rhizome.

Azolla pinnata subsp *asiatica* belonging to the family Azollaceae was aquatic in habit and dominantly documented in the water bodies of the study area. Luxuriant growth of *Azolla pinnata* sub sp *asiatica* was recorded. It is also used as fertilizer and poultry feed.

Family Davalliaceae was documented with a single species *Davallia repens* terrestrial in habit and occasionally distributed. It was categorized by IUCN under vulnerable category.

Genus *Lygodium* representing the family Gleicheniaceae was reported with a single species *Lygodium flexousum* which was the only climber in habit it was available in spore stage and the member was observed meagrely in the 3 study area. *Trichomanes obscurum* of the family Hymenophyllaceae was terrestrial in *Lindsaea ensifolia* belonging to the family Lindsaeaceae was terrestrial in habit and occasional in distribution.

The family Marsileaceae was represented by a single species *Marsilea quadrifolia* aquatic in habit and moderately distributed. *Acrostichum aureum* belonging to the family pteridaceae was documented with meagre in distribution and it was terrestrial in habit located along the banks of water bodies. *Ceratopteris thalictroides* belongs to the family pteridaceae was aquatic in habit some were found submerged in water and some distributed along the bunds of running waters it was occasionally documented.

Mickelopteris cordata attracted with the heart shaped leaves was terrestrial in habit even though terrestrial it was found growing embedded with soil it was occasionally reported. The genus *Pityrogramma* was documented with two species namely *Pitryogramma austroamericana* and *Pityrogramma calomelanos*. It belongs to the family Pteridaceae and terrestrial in habit occasionally reported. Though the members were terrestrial lush growh of the plants was recorded along the banks of water bodies and bunds of running waters with high moisture content. The genus *Pteris* was represented by two species namely *Pteris ensiformis* and *Pteris*

vittata belonging to family Pteridaceae, terrestrial in habit along the banks of water bodies. It was occasionally reported. The family Salviniaceae was documented with two species *Salvinia natans* and *Salvinia molesta,* aquatic in habit and commonly reported.

Global climate is probably the principal determinant of the vegetation pattern, which has considerable influence on distribution, structure, and ecology of the forest ecosystem, and it is assumed that changes in climate would alter constitution of an ecosystem (Singh *et al.,* 2015). Climate variables seem to be most important for explaining species richness patterns with elevation gradients for all kinds of living organisms (Bhattarai *et al.,* 2004). Climate factors, viz., temperature, potential evapotranspiration, length of the growing season, humidity, air pressure, nutrient availability, ultraviolet radiation, and rainfall, are varied for elevation, all of which can have an effect on distribution of species and their richness along the gradient in any forest ecosystem (Funnell and Parish, 2001). Majority of the pteridophytes collected was terrestrial in habit. It shows that the microclimatic conditions and ambient temperature in the surrounding environment was present which influenced the survival of the terrestrial pteridophytes. Though the pteridophytes where terrestrial they were collected along the banks of water bodies which clearly indicates that the microclimatic conditions are an essential factor for their survival. Four species namely *Marsilea quadrifolia, Ceratopteris thalictroides, Salvinia molesta* and *Salvinia natans* were aquatic in habitat. *Lygodium flexousum* was a climber and *Adiantum philippense* was lithophytic in habitat.

Distribution of pteridophytes in the site of collection is an indicating factor of its diversity. The diversity is also influenced by the canopy of vegetation. *Nephrolepis multiflora, Adiantum caudatum, Adiantum latifolium* and *Salvinia molesta* was

dominantly reported in the study area *Nephrolepis multiflora, Adiantum latifolium* occupied the entire barren land area around the water bodies with moisture content. Whereas *Salvinia molesta* was collected from the small water bodies along the pond. *Davallia repens, Adiantum caudatum, Adiantum latifolium, Adiantum philippense, Ceratopteris thalictroides, Mickelopteris cordata, Pityrogramma austroamericana, Pityrogramma calomelanos, Pteris vittata, Thelypteris articulata, Thelypteris dentata, Thelypteris ochthodes* and *Thelypteris paludosa* was occasionally reported in the study area. *Lygodium flexosum and Adiantum philippense* was reported with a single species alone. *Adiantum philippense* was found inside a cave like appearance with water from small stream running where the genus *Adiantum* was surviving with a single species alone.

Each species of fern has its own preferences of micro habitat depending on the temperature, humidity, soil type, moisture, pH, light intensity, etc., and in many cases are very specific indicators of the conditions they need (Shaikh and Dongare, 2009). It is well observed and noted that most species of ferns succeed under high humidity and shade conditions (Page, 1979), unless they are species that prefer more xeric conditions and are more heliophilous. *Lygodium flexousum* is the only scandent or climbing fern recorded in the current study prefers to grow among the bushes along the partially or fully exposed roadsides.

Among the collected pteridophytes two species namely *Davallia repens and Thelypteris palusoda* was enlisted under IUCN Category as vulnerable and endangered respectively. Majority of the pteridophytes obtained during the study are of economically important. According to Fraser-Jenkins (2012), out of the total 1100 indigenous pteridophyte of India 337 species are considered to be threatened or endangered. According to the current study there was only one species each under

vulnerable and endangered category respectively and the remaining were not enlisted under IUCN category.

Small urban patches contribute less evaporative cooling from trees, and are therefore more affected than large forests by the diffusion of dry and warm microclimatic conditions from the matrix to the forest interior (Grimm *et al.,* 2008). High water availability usually stimulates pteridophyte growth and controls the diversity of favourable habitats (Richard *et al.,* 2000). Contrary to such expectations, it was documented that lower pteridophyte richness in pond surrounded by large proportions of water bodies than in patches surrounded by small water bodies. Water is a key element for pteridophytes to thrive, the recurrence of anthropogenic disturbances in ecosystems located near water bodies, could have caused a cumulative population decline, and eventually the local extinction of a large number of pteridophytes. Actually, some of the highest rates of extinction have been found for native herbaceous plants living in urban riparian forests, while the introduction of exotic plants continues year after year, leaving floristically degraded sites (DeCandido *et al.,* 2004). At the microhabitat scale, abundant shrub cover was negatively correlated to pteridophyte richness, indicating that sample plots dominated by shrubs had a lower number of pteridophyte species. In general, pteridophytes are poor competitor against ligneous plants (Grime, 1985), which are more tolerant of anthropogenic disturbances in urban areas (LaPaix and Freedman, 2010). Despite the impact of biological interactions, the number of pteridophyte species in assemblages was essentially controlled by physicochemical conditions associated to topographic and edaphic properties. Moisture and soil pH are two parameters recognized as important gradients along which ferns are distributed in temperate forests (Richard *et al.,* 2000), and this is supported by the results of current study too.

The results showed that pteridophyte richness was negatively correlated with shrub cover, but positively correlated to soil moisture. Shrub density in edge habitats is often high, while air and soil moisture are low, due to lateral exposure to solar radiation (Murcia, 1995). These conditions may have induced higher interspecific competition, influenced fertility during the gametophyte stage, and caused hydric stresses on the fronds, as several pteridophytes have no physiological mechanism effective to control water loss (Page, 2002). Ionic concentration and pH imbalances in cells may cause physiological gaps (Bryson and Barker, 2002). Surprisingly, several species of pteridophytes tolerate high salt concentrations in their mature aerial organs (Bryson and Barker, 2002), but their prothallus cells may be intolerant to salts (Bogdanovi´c *et al.,* 2012). This phenomenon illustrates the limitations engendered by the two distinct stages (gametophyte and sporophyte) in the pteridophyte life cycle (Page, 2002).

Studies on pteridophytes are gaining momentum and this is shedding light on the medicinal and economic importance of these plants. Understanding the flora of a region always helps in understanding the change in the ecosystem and in-vitro and ex-situ conservation can be widely exploited to bring back the threatened species from the verge of extinction and preserve this plant group for our coming generations. Even though the present study was carried out in a short duration, 24 pteridophytes were encountered. A detailed investigation may yield further number of pteridophytes from the study area.

Coupled with the environmental factors, the incessant collection of ferns from the natural habitat by visitors and locals for ornamental purposes, medicinal purposes and during excursions have also increased the pressure for survival on these plants.

Such threats apart, nevertheless, the same region, one of the last bastions of pteridophytic wealth in the study area.

SUMMARY AND CONCLUSION

The 24 species of pteridophytes recorded during the investigation enroute from Enayam to Marthandam belongs the different families like Azollaceae, Davalliaceae, Gleicccheniaceae, Hymenophyllaceae, Lindsaeaceae, Marsilieaceae, Nephrolepidiaceae, Pteridaceae, Salviniaceae and Thelypteridaceae. Among the families the Pteridaceae was documented with diverse species, whereas family Thelypteridaceae was reported with five species. Family Salviniaceae was recorded with two species. All the members of the genus *Thelypteris* were terrestrial in habit though they were terrestrial this members were documented along the water banks for survival. Water bodies which contain moisture content is the driving force behind diversity. The other families like Azollaceae, Davalliaceae, Gleicheniaceae, Hymenophyllaceae, Marsileaceae and Nephrolepidiaceae was documented with single species. The diversity in the genus Thelypteridaceae was observed which might to due to the environmental hues which favoured the survival. This followed by genus *Adiantun* which was recorded with three species.

Among the members of the genus *Thelypteris paludosa* was enlisted under endangered category by IUCN. Family Davalliaceae was documented with a single species *Davallia repens* terrestrial in habit and occasionally distributed. It was categorized by IUCN under vulnerable category.

In conclusion detailed survey on the pteridophyte flora of a particular region becomes significant only if it can do any good to the conservation practices of these endangered plants. One of the key reasons for neglect faced by pteridophyte is a lack of knowledge or interest among botanists. So it is a way out of the near endangered

status of these plants to familiarise this group and include them with almost equal meaning to the Angiosperms.

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PHYTOCHEMICAL SCREENING, FTIR, ANTIBACTERIAL AND ANTHELMINTIC ACTIVITIES OF SELECTED SEAWEEDS FROM THE MANAPAD COASTAL REGION

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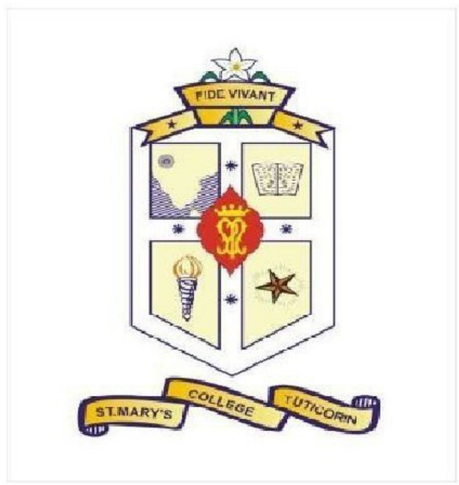

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CERTIFICATION

This is to certify that this dissertation entitled, **PHYTOCHEMICAL SCREENING, FTIR, ANTIBACTERIAL AND ANTHELMINTIC ACTIVITIES OF SELECTED SEAWEEDS FROM THE MANAPAD COASTAL REGION** submitted by **E. PONMARI, Reg.No. 21APBO12** to ST. MARY'S COLLEGE (Autonomous), THOOTHUKUDI in partial fulfilment for the award of the degree of **"Master of Science in Botany"** is done by her under my supervision. It is further certified that this dissertation or any part of this has not been submitted elsewhere for any other degree.

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I do here by declare that this dissertation entitled, **PHYTOCHEMICAL SCREENING, FTIR, ANTIBACTERIAL AND ANTHELMINTIC ACTIVITIES OF SELECTED SEAWEEDS FROM THE MANAPAD COASTAL REGION** submitted by me in partial fulfilment for the award of the degree of "**Master of Science in Botany**", in the result of my original and independent work carried out under the guidance of **Dr. Mrs. F. Dayana Lobo, M.Sc., M.Phil., Ph.D.,** Assistant Professor, Department of Botany, St. Mary's College (Autonomous) Thoothukudi and it has not been submitted elsewhere for the award of any other degree.

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CONTENT

INTRODUCTION

INTRODUCTION

Seaweeds, otherwise known as marine algae are primitive non-flowering photosynthetic macrophytes occurring in tidal regions of seas and oceans that occupy 71% of the globe and they are natural renewable resources. Green, brown and red seaweeds are generally distributed in the intertidal, tidal and subtidal regions respectively (Rao, 2018).

Macroscopic marine algae is one of the important living resources of the ocean. Agar, carrageenan and alginate are popular examples of seaweeds – these have been used as food for human beings, feed for animals, fertilizers for plants and source of various chemicals. In the recent past, seaweeds have also been gaining momentum as new experimental systems for biological research (Vijaya Kumar, 2003).

Seaweeds are considered as ecologically and biologically important component in the marine ecosystems. Seaweeds make a substantial contribution to marine primary production and provide habitat for near shore benthic communities. Seaweeds or marine algae have long made up a key part of the Asian diet and are also consumed in other parts of the world, such as in Ireland and Wales. Seaweed has often been used as a food for people who are sick and has been credited with health-giving properties. Today, seaweed supplements for human use are usually considered to be sources of iodine or minerals but may offer other therapeutic benefits. Seaweeds are key space occupiers of rocky shores and interact with other organisms and hence play a key role in overall coastal biodiversity. They are found on rocks in the intertidal zone as a giant underwater forest.

Seaweeds grow abundantly along the Indian coastline particularly in rocky shore regions; rich seaweed beds occur around Visakhapatnam in the eastern coast, Mahabalipuram, Gulf of Mannar, Tiruchendur, Tuticorin and Kerala in the southern coast; Veraval and Gulf of Kutch in the western coast; Andaman and Nicobar Islands and Lakshadweep. Seaweeds are under threat in developing countries, where they are being disturbed by a variety of human activities. Increasing concern on destruction of seaweed resources and alterations in the diversity of various life forms makes it necessary the studies on the taxonomy and species diversity for a better management of marine algae (Doss and Rukshana , 2016).

Brown seaweeds from the phylum Phaeophyta (meaning "dusky plants"), is the most prevalent type of seaweed. Brown or yellow-brown in color, brown algae are found in the waters of both temperate or arctic climates. While not roots in the true sense, brown algae typically have root like structures called " holdfast" that are used to anchor the algae to a surface. Seaweeds can thrive in both salt and freshwater, but the brown algae known as kelp grows only in saltwater, most often along rocky coastlines. There are about 30 kelp varieties (Jennifer Kennedy, 2019).

Red seaweeds from the phylum Rhodophyta. There are more than 6000 species of red algae. Red algae gain their often brilliant colors thanks to the pigment phycoerythrin. The ability to absorb blue light allows red algae to live at greater depths than either brown or green algae (Jennifer Kennedy, 2019).

Green seaweeds from the phylum Chlorophyta. More than 4000 species of green algae exists on the planet. Green algae can be found in marine or freshwater habitats, and some even thrive in moist soils. These algae come in three forms: unicellular, colonial or multicellular (Jennifer Kennedy, 2019).

The active Phytochemical produced by seaweeds include, alkaloids, glycocide, betecyanin, anthraquinonen, flavonoids, phenols, saponins, steroid, tannins, terpenes, quinines, proteins and carbohydrates . In recent years, advances have been made in the development of antimicrobial compounds in an effort to check the harmful effects of microorganisms (Rao, 1995).

Bacterial disease results when the harmful bacteria enter the organism then multiply and invade the body's defence mechanism. These pathogenic bacteria enter the body through inhalation, ingestion or damaged skin tissue. The inability of the immune system to stop the bacteria from reproducing and spreading consequently results in the symptoms of bacterial disease. The antimicrobial resistance is the foremost problem all over the world with present antibiotic therapy in treating infectious diseases. The development of drug resistance by microorganisms reduces the effectiveness of modern drugs. Thus, resistance to antibacterial agents poses threat in many areas of the world especially in the developing countries. The integration of traditional and modern medicine is gaining increase recognition globally (Abebe, 1996, WHO, 2000).

Turbinaria ornata is a very common brown algae found intertidally on Hawaiian reefs and throughout the Pacific and Indian Ocean. It belongs to the class Phaeophyceae and family Sargassaceae. It grows in a variety of habitats including rocky intertidal, tide pools, intertidal benches, reef flats and deeper water. It is normal found in small clusters attached to the crevices of basalt rocks in high wave action areas as well as in the crevices of coral heads at 20-30 meters deep. They are mostly light yellowish brown to dark brown with dark brown spots. Turbinaria ornata has a wide variety of health benefits and is being researched for pharmaceutical purposes because of its antioxidant, anti-inflammatory, antidiabetic, antiproliferative and neuroprotective effects on humans (Abbott, 2001).

Gracilaria is belongs to the class Rhodophyceae. Gracilaria is one of the Genus in Family Gracilariaceae with more than 100 species worldwide, inhabiting temperate and tropical seawaters, covering from intertidal to subtidal areas. Gracilaria is important as source of income in country such as Chile, where they have been cultured commercially with total landings of 120,000 wet metric tons. From ecological aspect, Gracilaria act as natural habitat for aquatic organisms and protect them from predators, waves and tides and certain fishes, crabs, and isopods prefer Gracilaria as food. Gracilaria is used as food and in the preparation of food products. Originally and especially in China, Gracilaria species were used as food and as binding material in the preparation of lime for painting walls (Abbott, 2001)

Halimeda tuna is belongs to the class Chlorophyceae. The green macroalgae of the genus Halimeda are important calcifying organisms in tropical and sub-tropical marine ecosystems (Maria Elizabeth bandeira- pedrosa et al., 2004). Macroscopically, the thallus is characterized by a series of green articulated coin shaped segments made rigid by the impregnation of calcium carbonate as aragonite. Halimeda genus has been investigated over the past years because of their source of natural antioxidants, neyro and hepato-protectant compounds. Halimeda tuna are a source of hydrophilic antioxitants, which could be further recommended for the prevention of oxidative stess-relaed disturbances and can be of usefulness either as dietetic supplements or as food ingredients. Some species of Halimeda are used as fertilizers to recondition acidic soils. They are also used as animal feed and reportedly have anti-bacterial and anti-fungal properties (Abbott, 2001)

SCOPE AND OBJECTIVES

SCOPE AND OBJECTIVES

Seaweeds constitute one of the commercially important marine living renewable resources. They are good source of food and medicine. Seaweeds derived natural products such alkaloids, flavonoids, terpenoids have received considerable attention in recent years due to their diverse pharmacological properties including antioxidant and antimicrobial activities. According to World Health Organisation (WHO), seaweeds should be greatest source to obtain an array of drugs. Selected seaweeds such as Turbinaria ornata, Gracilaria corticata and Halimeda tuna were selected for study. Hence the present investigation was undertaken with the following objectives:

1. Enlisting and recording the availability status of seaweeds in Manapad.
2. To qualitatively screen the presence of phytochemicals in selected seaweeds such as Halimeda tuna, Turbinaria ornata and Gracilaria corticata.
3. To elucidate the effectiveness of seaweeds in controlling human pathogenic bacteria such as Escherichia coli, Bacillus subtilis and Staphylococcus aureus.
4. To evaluate anthelmintic activity in selected seaweeds of Halimeda tuna, Turbinaria ornata and Gracilaria corticata.
5. To identify the functional groups present in the seaweeds of Halimeda tuna, Turbinaria ornata and Gracilaria corticata (FTIR).

REVIEW OF LITERATURE

LITERATURE REVIEW

Seaweeds are considered as a source of bioactive compounds as they are able to produce a great variety of secondary metabolites characterised by a broad spectrum of biological activities. Compounds with antioxidant, antiviral, antifungal and antimicrobial activities have been detected in brown, red and green algae (Yuan et al., 2005; Bansemir et al., 2006; Chew et al., 2008). Seaweeds are rich and varied source of bioactive natural products and have been studied as potential bioactive and pharmaceutical agents. They are used in traditional remedies in many parts of the world. Extracted substances from seaweeds have phytochemical, antibacterial actions and other properties include antifungal activities and growth inhibition of plants (Abdussalam, 1990; Scheuer, 1990; Rizvi and Shameel, 2003; Su et al., 1973; Burkholder and Sharma, 1969; Chapman, 1980; Arasaki, 1983; Abbott, 1988).

Phytochemical activity and Antibacterial activity:

Gopalan Rajkumar and Periyakali Saravana Bhavan, 2017 understand the phytochemical components of the edible seaweed, Turbinaria ornata, it was subjected to hexanic, acetonic and methanolic extractions. The alkaloids, Terpenoids, Flavonoids, polyphenols and quinines were present in the hexanic extract of T.ornata whereas alkaloids, Terpenoids and Flavonoids were absent in acetonic and methanolic extracts of T.ornata. Overall, presence of five secondary bioactive components {neophytadiene; 2- hexadecen-1-ol,3,7,11,15-tetramethyl-,[R-[R*, R*-(E)]; 17-pentatriacontene; 4,8,12,16-octadecatetraen-1-ol,4,9,13,17-tetramethyl; squalene} has been identified in T.ornata, of which four bioactive compounds except 4,8,12,16-octadecatetraen-1-ol,4,9,13,17-tetramethyl have been recorded in methanolic extract. Moreover, the presence of 17-pentatriacontene and squalene were unique only to methanolic extract of T.ornata.

Gopalan Rajkumar and Periyakali Saravana Bhavan, 2017 studied the phytochemical screenings of the red alga, Gracilaria corticata was subjected to hexanic, acetonic and methanolic extactions. Hexanic extract of G. corticata showed presence of alkaloids, terpenoids, flavonoids, polyphenols and quinones. Acetonic extract of G. corticata showed presence of alkaloids, tannins, polyphenols, saponins, cardiac glycosides and quinones. In case, methanolic extract of G. corticata contained tannins, polyphenols, saponins, cardiac glycosides and

quinones. Further, GC-MS analyses of G. corticata revealed that presence of 17 secondary compounds (6 from hexanic, 4 from acetonic and 7 from methanolic extracts). Among the 17 compounds, 8 compounds (1 from acetonic and 6 from methanolic and 1 from all the three extracts) are possessed bioactive properties based on literature. Thus, G. corticata has significant amount of primary and secondary phytochemicals. Among the three solvents extractions, methanolic extract possessed more numbers of secondary as well as bioactive compounds

Rajan Renuka Remya et al., 2022 studied the marine biosphere is the primary source that has produced excellent bioactive metabolites. Natural compounds isolated from various algae, especially brown algae, gained interest because of their wide variety of biological activities and biocompatibility. Among brown algae, Turbinaria ornata, a highly prevalent alga, because of the presence of bioactive substances, primarily polysaccharides and proteins, could be used for a broad range of pharmaceutical applications. Also a few natural compounds isolated from the Turbinaria species have been used for the biogenic nanoparticle synthesis that was considered to be potential for desired biological applications.

Alireza Ghannadi et al., 2016 studied Turbinaria ornata methanolic extract with more bioactive compounds has been investigated for its in vitro total antioxidant activity, DPPH scavenging assay, reducing power assay and antioxidant potential. Our preliminary phytochemical analysis showed that Turbinaria ornata methanolic extract (TOME) constituents carbohydrates, alkaloids, saponins, phenolic compounds, flavonoids, tannins, coumarins, steroids and terpenoids. The methanolic extract of Turbinaria ornata at the concentration of 100 µg showed 89.11% of total antioxidant activity. The free radicals NO, H2O2 and SOD scavenging activities were enhanced with an increase in the concentration of the methanolic extract of Turbinaria ornata. Further, the methanolic extract of Turbinaria ornata at selected 0.5, 0.75 and 1(mg/ml) concentrations showed significant reduction in H2O2 -induced hemolysis.

Gnanaprakasam et al., 2017 investigated the antibacterial activity in hexane, chloroform, ethyl acetate, acetone and methanol extracts of Gracilaria corticata J. Ag against bacterial and fungal strains viz., Bacillus subtilis, Streptococcus pyogenes, Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, Salmonella typhimurium, Vibrio cholerae, Shigella flexneri, Proteus mirabilis and Proteus vulgaris. Fungal strains Candida albicans, Candida krusei, Candida guilliermondi, Candida parapsilosis, Candida tropicalis, Candida

glabrata, four dermatophytes viz., Trichophyton rubrum, Trichophyton mentagrophytes, Microsporum gypseum and Epidermophyton flocossum. The extracts of G. corticata were extracted with different solvents viz., hexane, chloroform, ethyl acetate and methanol against bacterial and fungal strains by using disc diffusion method. The ethyl acetate extract of G. corticata showed the highest antibacterial and antifungal activity against all the bacterial and fungal strains tested than the other extracts. The mean zones of inhibition produced by the extracts in agar disc diffusion assays were from 7.1 to 16.0 mm. The Minimum Inhibitory Concentrations (MIC) was between 125 and 500µg/ml, while the Minimum Bactericidal Concentrations and Minimum Fungicidal Concentrations (MFC) were between 250 and 500µg/ml. The highest mean zone of inhibition (16.0 mm) was observed in ethyl acetate extract of G. corticata against B. subtilis. The lowest MIC (125µg/ml), MBC and MFC (250µg/ml) values was observed in ethyl acetate extract of G. corticata against B. subtilis.

Kannan et al., 2014 evaluated the phytochemicals, the antioxidant activity of marine algae Gracilaria corticata (G. corticata) and Spirulina platensis. The phytochemicals present in the selected marine algae G. corticata and S. platensis were screened and their antioxidant activities were tested. The marine algae was collected, shade dried, powdered and extracted with methanol. The presence of a variety of chemical constituents, such as saponins, phenols, glycosides, flavonoids and alkaloids were analyzed in these marine algae by TLC and HPLC method. Their antioxidant activities were studied by Fentons method and DPPH assay. Phytochemical screening showed the presence of active molecules. The selected algae is having antioxidant potential.

Rashida Qari and Abdul Rahim Khan, 2019 studied prominent pathogenic enteric include strains of Escherichia coli and Salmonella typhi which are responsible for diarrhea. The antibacterial characteristics of methanol, acetone, diethyl ether and ethanol extracts of three Gracilaria species (Gracilaria corticata J. Agardh, Gracilaria dentate J. Agardh and Gracilaria pygmaea Borgesen) collected from different shores (Buleji, Paradise Point, Manora channel and Mubarak village) of Pakistan coast. All these species were tested in vitro for their antidiarrheal activities against diarrhea causing E.coli and Salmonella typhi in five children of different ages (one, two, five, nine and ten years) stool culture that were affected by diarrhea.

Dayuti, 2017 reported the chemical analysis of red algae contained terpenoid, acetogenic and aromatic compounds, which have a wide range of biological activities, such as anti-microbial, anti-inflammatory and anti-viral. The objectives of this research was to evaluate the effect of extraction solvent and time on antibacterial activity of red algae (Gracilaria verrucosa), and to explore the bioactive compound contained with in Gracilaria verrucosa. The method in this study used descriptive research. These finding revealed that the highest inhibition activity among all extracts was obtained with the ratio of methanol:aquades (75:25) and extraction time around 72 hours against Escherichia coli and Salmonella typhimurium. The bioactive compounds of Gracilaria verrucosa tested by phytochemical analysis consisted of flavonoid, alkaloid and saponin. Those secondary metabolites may be approximated as antibacterial substances.

Jayashree et al., 2018 evaluated the biological activity of Red algae, Gracilaria corticata, collected from the southeast coast of Mandapam, Ramanathapuram, Tamil Nadu. Seaweeds are rich in bioactive compounds. Seaweeds are highly diverse group of organism from secondary metabolites of the natural source are a potential source. The marine seaweed is the interesting group of their broad spectrum of biological activities such as antibacterial, antioxidant and anticancer. The antibacterial activity of G. corticata was tested against Staphylococcus aureus and Escherichia coli by disc diffusion method. In vitro antioxidant activity was determined by 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging assay. 3-(4,5-dimethylthiazol-2-lye)-2,5-diphenyltetrazolium bromide assay was employed to study the anticancer activity against (MDA-MB 231) human breast cancer cell line. The DPPH assay screening of methanolic extract of G. corticata showed specific activity of inhibition. In antibacterial shows the growth of two virulent strains of pathogenic bacteria, E. coli and Bacillus subtilis. In anticancer activity obtained results indicated that the methanol extracts of G. corticata were cytotoxic against (MDA-MB 231) human breast cancer cell.

Abimannan arulkumar et al.,2018 studied the increasing of resistance pathogenic microorganisms to majority of antibiotics, there is an urgent need for exploring plant based drugs and bioactive compounds with least side effects. The study was aimed to determine the level of phytochemicals, antioxidant, antibacterial properties of the edible red seaweeds, Gracilaria corticata and G.edulis. The extraction with methanol yielded 7.10 ± 0.16 and 6.39 ± 0.16 %

extracts from G.corticata and G.edulis respectively. The G.corticata possess higher total phenol content (4.00 ± 0.35 mg GAE/g) compare to G.edulis (3.4 ± 0.21 mgGAE/g). G.corticata and G.edulis extracts significantly varied in total flavonoid content i.e 3.33 ± 0.12 and 2.6 ± 0.08 mg CE/g DW respectively. In this investigation, G. edulis presented the highest 2, 2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity (23.95%) when compare to G. corticata (20.32%). G.edulis showed significantly higher 2, 2′-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) radical scavenging activity (40.24%) compare to G. corticata (32.65%). In addition, G. corticata exhibited higher nitric oxide (NO*) radical scavenging activity (36.78%) than G. edulis 35.25%. Antimicrobial properties of 70% methanol and DMSO extracts were found effective against Bacillus subtilis. GC-MS analysis revealed the presence of phytochemical compounds including sulfurousacid, 2-ethyl hexyl isohexyl ester, pentatriacontane, eugenol and phthalic acid played a vital role in antioxidant and antibacterial activities.

Manoj kumar Narashimhan et al., 2013 investigated the bioactive properties of three seaweed samples, Enteromorpha antenna, Enteromorpha linza and Gracilaria corticata were collected from the shoreline of Mahabalipuram, Tamilnadu. Bioactive components were extracted by using various solvents. Antioxidant analysis methods like scavenging activity of nitric oxide, hydrogen peroxide, hydroxyl radicals, free radical scavenging (DPPH), FRAP (ferric reducing ability plasma) ability and reducing power were carried out. MTT assay was employed to study the anticancer activity against cancer cell lines Hep-G2, MCF7 and normal VERO cell lines. It was found that methanolic extracts elicited higher total phenolic content, higher percentage scavenging activity of nitric oxide, hydrogen peroxide, hydroxyl radicals, free radical scavenging (DPPH), FRAP (ferric reducing ability plasma) ability and reducing power. Different concentrations of crude methanolic extracts of seaweeds showed potential antimicrobial activity by well diffusion method. Crude methanolic extract of G. corticata had significant anticancer activity followed by E. antenna and E. linza on cancer cell lines Hep-G2, MCF7 and normal VERO cell lines by MTT assay. The methanolic extracts of seaweeds Enteromorpha antenna, Enteromorpha linza and Gracilaria corticata possess high total phenolic content and shows a good free radical scavenging activity and hence are proven to have better

antioxidant activity and they might be good candidates for further investigations in order to develop potential anticancer drugs.

Geetha Devi and Shree Devi Kumari, 2022 studied the phytochemicals such as flavonoids, tannin, saponins, terpenoids, glycosides, steroids, fat and fixed oil and several bioactive compounds were observed by using GC-MS analysis in red algae Gracilaria corticata. The antibacterial activity of various extracts of G.corticata extracts revealed that the methanol and chloroform extracts had the greatest zone of inhibition (19 mm) against the pathogenic bacteria Klebsiella pneumonia, Staphylococcus aureus, E. coli, Bacillus subtilis, Lactobacillus sp, Enterococcus sp and Pseudomonas aeruginosa, while the aqueous extract had the least zone of inhibition (9mm). Antioxidant activity of various extracts of G.corticata were evaluated by 2, 2-diphenyl-1- picrylhydrazyl (DPPH), 2, Ferric Reducing Antioxidant potential (FRAP), and Hydrogen Peroxide Assays and methanolic extract of G.corticata have much better H2O2 Scavenging activity (IC 50 value 60.904 µg/ml) than the standard medication, Ascorbic acid (IC50 value 259.849 µg/ml). In vitro anticancer activity tested against Human Breast Adenocarcinoma cell line (MCF-7), Lung Cancer cell line (A549), and Colorectal Carcinoma cell line (HCT-116) of methanolic extract of G. corticata was tested. Among the 3 cancer cell lines highest inhibitory percentage was observed in HCT-116 cell lines (IC50 value 108.10 µg/mL-1). The methanolic extract of G. corticata inhibited α-glucosidase with high IC50 value of 55.2015 µg/ml indicated the Antidiabetic actvitiy. All these finding may lend credence to the use of G. corticata in traditional medicine.

Diyah Fatimah Oktaviani et al., 2019 investigated on natural products which can against fouling bacteria is really essential. This research reported the potentials of seaweeds Turbinaria ornata and Chaetomorpha antennina against fouling bacteria which have been tested on the inhibition zone and phytochemical contents. The seaweed samples were extracted using various solvents such as hexane, ethyl acetate and methanol. The results showed that C. antennina had more potentials against the fouling bacteria than T. ornata regarding to the maximum inhibition zone. C. antennina extract had 6 mm at 10 µl/disk when T. ornata only had 2 mm at 10 µl/disk. In addition, C. anteninna extracts also had more phytochemical contents (phenol, flavonoid, steroid, saponin, alkaloid and triterpenoid) than T. ornata extracts (phenol, steroid, saponin, and alkaloid). Having larger maximum inhibition zone and more phytochemical

contents indicated that C. anteninna can be the natural source candidate for antibacterial agent especially fouling bacteria.

Mohamed Shaibi et.al., 2022 reported in Turbinaria ornata (Turner) J., tropical brown algae, was found in the South Pacific and Indian Ocean ecosystems. In accordance with recent studies, Turbinaria ornata J. has potent anti-inflammatory effects. Therefore, this study is aimed to explore the biological activities of ethanolic extract of T. ornata J. by analyzing the presence of phytochemical components, antioxidant property, antimicrobial activity, and the wound healing activity. From the results, phytochemical analysis of ethanolic extract of T.ornata J. showed the presence of alkaloids, saponins, oils, total phenolic, and total flavonoid content which were estimated to be 0.683±0.001 Abs and 0.433±0.001 Abs, respectively. Antioxidant activity of the ethanolic extract of T. ornata J. extract showed remarkable DPPH radical scavenging activity of about 58.8% at 200 μg/mL and total antioxidant activity of 0.257 Abs at 100 μg/ml concentration, as compared to that of their respective controls. The ethanolic extract of T. ornata J. exhibited the maximum zone of inhibition against the clinical pathogens like Pseudomonas aeruginosa, Escherichia coli, Staphylococcus aureus, Candida albicans, and Methicillin-resistant Staphylococcus aureus with their potent antimicrobial activity. Wound healing effects of the ethanolic extract of T. ornata J were analyzed by using zebra fish model.

Fatma Mohamad et al., 2016 investigated the potential antibacterial activities of ethanol extracts of Turbinaria ornata (T. ornata), Oleic acid (OA) and palmitic acid (PA) extracted from T. ornata as well as mixtures of OA and PA (1:1) against some bacterial species. Brown seaweed T. ornata was collected from Hurghada shores, Red Sea coast of Egypt. OA and PA were extracted from T. ornata. Ethanol extracts of T. ornata, OA, PA and mixtures of these two fatty acids (1:1) were tested for their antibacterial activities against Escherichia coli, Bacillus cereus, Bacillus subtilis, Klebsiella pneumonia and Staphylococcus aureus by the disc diffusion method. Ethanol extracts of T. ornata, OA, PA and mixtures of OA and PA (1:1) showed antibacterial activities that increased significantly (least significant difference at 0.05 level) with increasing concentrations against all tested bacteria. Different concentrations of ethanol extracts of T. ornata and extracted OA showed its highest activity against Bacillus subtilis, while PA and mixtures of PA and OA (1:1) showed its highest activity against Bacillus cereus. The maximum inhibition activities were shown for mixtures of OA and PA (1:1).

Scanning electron microscope showed that mixtures of OA and PA (1:1) caused plasmolysis and reduction in cell size of Escherichia coli. Different concentrations of T. ornata and its fatty acids showed activities against all tested bacteria. Therefore, it is a potential source of natural antimicrobial compounds.

Sornalakshmi et al., 2021 investigated the qualitiative and quantitative phytochemical analysis of red seaweed Gracilaria corticata. DPPH radical scavenging activity using different solvent extracts also performed. Preliminary phytochemical screening confirmed the existence of proteins, carbohydrates, lipids, aromatic acids, alkaloids, phenols, flavonoids, tannins, terpenoids, steroids, saponins, coumarins, quinones, anthroquinones and catechins. G. corticata contains 16.7% protein, 1.0 % lipid, 7.9% carbohydrate, 3.14 mg GAE/g DW phenol and 1.05 mg GAE/g DW flavonoid. Maximum DPPH radical scavenging ability was recorded by ethanol extract (74.5%) followed by methanol (73.82%), water (73.62%), chloroform (36.9%) and petroleum ether (18.2%). No scavenging activity recorded by benzene extract.

Krishnaveni Eahamban et al., 2012 investigated the preliminary phytochemical analysis and UV-VIS, HPTC profiling and the antibacterial activity of Gracilaria corticata J. Ag extracts against the Gram positive and Gram negative bacteria. Preliminary phytochemical screening was carried out by Harborne method. The G. corticata extracts were tested against bacteria by the agar disc diffusion method. The results of the presence study showed the presence of alkaloids, steroids, phenolic groups, saponins, tannin, flavonoids, terpenoids, glycosides and sugars. Proteins, xantoproteins, coumarins and catechin did not show any positive result for their presence in any of the six extracts of Gracilaria corticata tested. The result of the present study revealed the various behavior character of Gracilaria corticata crude drug. The UV-VIS spectrum profile of Gracilaria corticata methanolic, petroleum ether, benzene and aqueous extracts profiles were recorded. The HPLC profile of Gracilaria corticata petroleum ether benzene and aqueous extracts were tabulated. The maximum (9/12 bacterial pathogens) degree of antibacterial activity was observed in isopropanol soxhlet extracts followed by isopropanol cold extracts (7/12 bacterial pathogens).

Johnsi christobel et al., 2011 reported in aqueous extract of seven species of marine macroalgae were screened for their antimicrobial potency against ten pathogenic bacterial strains. Ulva fasciata, Gracilaria corticata, Sargassum wightii and Padina tetrastromatica

showed significantly higher activity against 70% of the tested bacterial isolates. The maximum zone of inhibition was noted for the red alga G.corticata against Proteus mirabilis (17mm) and brown alga P. tetrastromatica against the pathogens Staphylococcus aureus and Vibrio harveyi (15mm). The general trend of inhibitory activity was higher towards Gram negative bacteria.

Indira et al., 2013 examined the seaweed (Halimeda tuna) for antibacterial and antifungal activity in vitro using the well diffusion method, minimum inhibitory concentration, minimum bactericidal concentration and minimum fungicidal concentration. The activity was against 10 bacterial strains (Staphylococcus aureus, Salmonella typhimurium, Salmonella paratyphi, Klebsiella oxytoca, Escherichia coli, Proteus mirabillis, Lactobacillus vulgaris, Pseudomonas sp., Klebsiella pneumonia and Vibrio cholerae) and nine fungal strains (Aspergillus niger, Aspergillus flavus, Alternaria alternaria, Candida albicans, Epidermophyton floccossum, Trichophyton mentagrophytes, Trichophyton rubrum, Pencillium sp. and Rhizopus sp.). The methanolic extracts in the present study exhibited a broad spectrum of antimicrobial activity compared to the ethanolic and chloroform extracts.

Kannan, 2014 elucitated an intensive spectroscopic (FT-IR and EDS) analyses were made on both brown algae Sargassum wightii and red algae Gracilaria corticata. The samples were collected from Vedalai, Gulf of Mannar, near Rameswaram coastal region, Tamil Nadu. The collected samples were stored in a cool place and the SEM diagrams for cross sectional area of the samples were taken. To identify the frequency of functional groups in the samples, FT- IR technique was performed. The band sat 3371 cm-1, 2924 cm-1 and 2358 cm-1 corresponding to N-H/O-H, C-Hand C-O stretching vibrations respectively in various amines, hydroxyl and Carboxylic groups. Elements such as S, Cl, Fe, Zn were found to occur in largest amount in Sargassum wightii while Fe, Si, Cl, S, and Na are in highest proportion in the Gracilaria corticata. The main functional groups involved in the seaweeds uptake are Carboxyl, Sulfydryl and Hydroxyl which are the prime constituents of the seaweeds. The estimation of trace elements in the seaweeds is of great value so that the seaweeds can be treated with necessary nutrient materials in order to ensure proper growth, yields and make them resistant to disease and toxicity.

Radhika and Mohaideen, 2015 presented the work at two seaweeds Ulva lactuca and Gracilaria corticata, which were collected from Hare Island in the Gulf of Mannar of Tuticorin

coast. Ethanol was taken as the solvent for extraction. The crude extract was purified using column chromatography. Antibacterial activity of crude and column purified fractions were tested against Klebsiella, Aeromonas, Staphylococcus, Escherichia and Pseudomonas using well-diffusion method. Maximum zone of inhibition (9 mm) was found in the crude extract of G. corticata against Pseudomonas sp. Minimum zone of inhibition (4 mm) was found in U. lactuca fraction against Escherichia coli. Highest antibacterial activity was obtained in red seaweed, whereas, green seaweed showed less antibacterial activity.

Rashida Qari and Shaima haider investigated the intensive spectroscopic FTIR analysis. It was determined in all three species of G. corticata, G. dentata and G. longissima. The bands at 414.7/cm to 3917.2/cm represents stretching and bending vibrations of alcohol O-H, amine N-H, alkane C-H, alkyne C=C, nitriles C=N, carboxyl C=O, nitro aromatic N=O, alkane C-C, nitro methane C-N, aliphatic amines C-N, sulfoxides S=O, alkene C-H alkyl halide C-Cl, C-I groups. The ash content of all studied species (G. corticata, G. dentata and G. longissima) was in the range of 20-30%, while the carbohydrate content was in the range of 22-24%.

STUDY AREA

STUDY AREA

The study area is situated on the distal end of Gulf of Mannar Biosphere Reserve. The rocky shore of Manapad inhabits an astonishing biodiversity, representing nearly almost all the invertebrate phyla and urochordates. Hard rocky bottom of this area greatly supports the algal diversity and provide suitable shelter and feeding ground for grazers.

PLATE 1:

MATERIALS AND METHODS

MATERIAL AND METHODS

The selected seaweeds such as Halimeda tuna, Turbinaria ornata and Gracilaria corticata were collected from the Manapad coastal, Gulf of Mannar, Thoothukudi were described below

HALIMEDA TUNA

Kingdom: Plantae

Phylum : Chlorophyta

Class : Chlorophyceae

Order : Bryopsidales

Family : Halimedaceae

Genus : Halimeda

Species : tuna

DESCRIPTION:

Halimeda tuna is a calcareous green seaweed, attached to the seabed by a holdfast. Each individual thallus (frond) consists of a single cell forming a tube with multiple cell nuclei. The cytoplasm is mobile and the nuclei, chloroplasts and other cell contents are free to move around inside the cell wall.

SIZE:

Rod shape with a size ranging from 2 to 54 nm

HABITAT:

Found in the tropical and subtropical regions.

DISTIBUTION:

Found in Indo-Pacific region, the Mediterranean Sea, Indian oceans and the western Atlantic Ocean.

USES:

Some species of Halimeda are used as fertilizers to recondition acidic soils. They are also used s animal feed and reportedly have anti bacterial and antifungal properties.

TURBINARIA ORNATA

Kingdom: Plantae

Phylum : Phaeophyta

Class : Phaeophyceae

Order : Fucales

Family : Sargassaceae

Genus : Turbinaria

Species : ornata

DESCRIPTION:

It is a very common brown algae characterized by an upright thallus with radially branched axes bearing blades. It has a tough texture. The blades come in various forms described as stipitate, turbinate, crowned and obpyramidal. Its appearance resembles that of a long pinecone.

SIZE:

2-20 cm tall

HABITAT:

Found in tropical and subtropical regions.

DISTRIBUTION:

Found intertidally on Hawaiian reefs and throughout the Pacific and Indian Ocean.

USES:

Used as a potential source for reducing postprandial hyperglycemia in humans making it an alternative therapeutic approach in treating diabetes.

GRACILARIA CORTICATA

Kingdom: Plantae

Phylum : Rhodophyta

Class : Rhodophyceae

Order : Gracilariales

Family : Gracilariaceae

Genus : Gracilaria

Species : corticata

DESCRIPTION:

The thallus consists of bundles of that and much divided blades with 2-3 mm broad segments branching numerous marginal projection line the edge of the segment in a pinnate fashion, the colour of the plant vary from deep purple to grass green.

SIZE:

Grows up to a height of 10-12 cm long

HABITAT:

It is seen in intertidal region of the coast.

DISTRIBUTION:

Indian oceans, Dwarka, OKha (Gujarat), Bombay, Malvan, Ratnagiri, Maharashtra, Goa, Karwar, Honawar, Bhatkal (Karnataka).

USES:

It can be used for agar production, food, animal feed.

COLLECTION OF SEAWEEDS:

Seaweeds, a group of non- flowering plants endowed with broad spectrum of neutraceutical and pharmaceutical components with significant species diversity are found inhabited in Manapad of Gulf of mannar. Manapad is situated 51.3 Km away from Tuticorin. A field survey was undertaken in the selected sampling stations of Manapad coast of Tamil Nadu, India (8.3775° N; 78.0522° E) over a period of three months from January to March 2023. During low tide by random sampling method. After collection, seaweeds were washed with seawater to remove all extraneous matters such as epiphytes, shells, associated fauna and adhering sand particles. Later, seaweeds were thoroughly cleaned with tap water to remove salt on the surface

and preserved by wet (5% formalin in seawater) and dry preservation (herbaria) methods. Seaweed species were identified by referring to authentic floras and books (Srinivasan 1969; Misra, 1996; Fritsch, 1977). Diversity and availability status of seaweeds were recorded as most abundant (++++), abundant (+++), less abundant (++) and sparse (+). Seaweeds encountered in Manapad were also photographed.

Preparation of seaweed powder:

Dominated seaweeds such as Halimeda tuna, Turbinaria ornta and Gracilaria corticata were air-dried under shade at 30 to 35°C and 65-70% relative humidity and were powdered in an electrical miller, sieved and stored. These powdered samples were extracted using solvents like ethanol, methanol, acetone, hexane and aqueous by soxhlet apparatus. All extracts were used to find out the phytochemical constituents such as alkaloids, quinines, carbohydrates, saponins, terpenoids, steroids, tannins, glycosides, proteins, coumarin, phenols, flavonoids, vitamin-C and free radical scavenging activity.

QUALITATIVE PHYTOCHEMICAL SCREENING:

Phytochemical analysis was carried out for all extracts using standard methods (**Horbone 1984, Kojate et al., 1995, Harborne 1998**).

Test for Alkaloid (Wagner's test):

About 1 ml of extract was taken and few drops of Wagner's reagent was added and the formation of a reddish brown precipitate indicate the presence of alkaloids.

Test for Flavonoid (Shinoda Test):

About 1 ml of extract was added to pinch of magnesium turnings and 1-2 drops of concentrated hydrochloric acid was added. Formation of pink color indicated the presence of Flavonoids.

Test for Phenol (Lead acetate test):

About 1 ml of extract was taken and 0.5 ml of 1% lead acetate solution was added and the formation of precipitate indicated the presence of tannins and phenolic compounds.

Test for Quinines:

A small amount of extract was treated with concentrated HCL and observed for the formation of yellow colour precipitate.

Test for Tannin (Ferric chloride test):

About 1 ml of extract was taken and 0.5 ml of 5% ferric chloride was added. The development of dark bluish black color indicated the presence of tannins.

Test for Steroid:

About 1 ml of extract was dissolved in 2 ml of chloroform and equal volume of concentrated sulphuric acid was added along the sides of the test tube. Appearance of brown ring indicated the presence of steroids.

Test for Carbohydrate (Benedict's test):

About 5 ml of Benedict's solution was added to 1 ml of extract and boiled in water bath. The appearance of red or yellow or green precipitate indicated the presence of reducing sugars.

Test for Saponin (Foam test):

About 0.5 mg of extract was diluted with 20 ml distilled water and shaken well in a graduated cylinder for 15 min. The formation of foam to a length of 1 cm indicated the presence of saponins.

Test for Glycoside:

About o.5 ml of extract was dissolved in 1 ml of water and then aqueous NaOH solution was added. Formation of yellow color indicated the presence of glycosides.

Test for Protein:

Ninhydrin test:

About 0.5 ml of extract was taken and 2 drops of freshly prepared 0.2% ninhydrin reagent was added and heated. The appearance of pink or purple color indicated the presence of proteins, peptides or amino acids.

Test for Terpenoid:

Five ml of the extract was mixed with 2 ml of chloroform and concentrated sulphuric acid to form a layer. A reddish brown coloration of the interface showed the presence of terpenoids.

Test for Coumarin:

About 3 ml of 10% NaOH were added to 2 ml of plant extracts. The formation of a yellow color was an indication for the presence of coumarins.

ANTIBACTERIAL ACTIVITY:

The powdered seaweed samples (20 gms) were extracted with 300 ml of hexane, benzene, chloroform, methanol, petroleum ether and water using soxhlet apparatus. Solvent was evaporated under vacuum and the concentrates were used for antibacterial assay.

Antibacterial activity – Disc diffusion Assay (Bauer et al., 1966).

Antibacterial activity of each seaweed extract was analysed using human pathogens., Gram positive bacteria, Bacillus subtilis and Staphylococcus aureus. Gram negative bacteria Escherichia coli obtained from the Department of Microbiology; St. Mary's College (Autonomous), Thoothukudi. Each bacterial pathogen was subcultured in agar medium and maintained. Whatman No.l sterile filter paper discs (6mm) were impregnated with different seaweed extract concentrations 0.5 mg/ ml, 1 mg/ ml, 1.5 mg/ ml, 2 mg/ ml, 2.5 mg/ ml and 3 mg/ ml and dried aseptically at room temperature. The spread plates were prepared by proper concentration of inocula. Each sample loaded disc was placed in the seeded agar plate. After 24-48 hours of ±37°C incubation, the diameter of the inhibition zone was measured. For positive control, streptomycin disc (100 µg/ ml) was used, whereas for negative control,

respective solvents were loaded on sterile discs. All the assays were carried out in triplicates. The seaweed extract concentration which has effected minimum inhibition (MIC = 2.5 mg/ ml) was used for further studies.

Anthelmintic Activity (Kumar et al., 2010):

Preparation of extract:

Dried seaweed powder (Turbinaria ornata, Gracilaria corticata and Halimeda tuna) (0.1 gram) was extracted with 10 ml of ethanol, methanol, acetone, hexane and aqueous for 24 hrs. The extracts were filtered through filter paper (What man No.1). The filtrate was collected and concentrated till a syrupy mass was obtained and dried at room temperature. The dried extracts were dissolved in normal saline and used for anthelmintic activity.

Experimental animal:

Due to its anatomical and physiological similarity to human intestinal parasite, anthelmintic operation was conducted on adult earthworms.

Experimental design:

In the present investigation the earthworms were divided into the following 6 groups. Each group consists of 3 earthworms.

Group I: Earthworms were placed in normal saline and served as control.

Group II: Earthworms were placed in ethanol extract of seaweeds (Turbinaria ornate, Gracilaria corticata and Halimeda tuna) at the dose of 10mg/ml.

Group III: Earthworms were placed in methanol extract of seaweeds (Turbinaria ornate, Gracilaria corticata and Halimeda tuna) at the dose of 10mg/ml.

Group IV: Earthworms were placed in acetone extract of seaweeds (Turbinaria ornate, Gracilaria corticata and Halimeda tuna) at the dose of 10mg/ml.

Group V: Earthworms were placed in hexane extract of seaweeds (Turbinaria ornate, Gracilaria corticata and Halimeda tuna) at the dose of 10mg/ml.

Group VI: Earthworms were placed in aqueous extract of seaweeds (Turbinaria ornate, Gracilaria corticata and Halimeda tuna) at the dose of 10mg/ml.

Group VII: Earthworms were placed in standard drug albendazole at the dose of 10mg/ml served as standard.

FT-IR (Fourier transforms infra-red spectroscopy) spectroscopic analysis:

One milligram of Halimeda tuna, Turbinaria ornata and Gracilaria corticata powder was mixed with 100 mg of dry potassium bromide (FT-IR grade) and then compressed into a pellet using hydraulic press (500-1000 psi). The pellet was immediately put into the sample holder and FT-IR (Systronics 166) spectra were recorded in the range of 400-4000 cm-1

RESULTS AND DISCUSSIONS

RESULTS AND DISCUSSION

Sea vegetables, which are commonly referred to as marine algae or seaweeds, have been a stable food since ancient times in countries located by the sea, viz., U.K, Ireland, Norway, the Pacific Islands, African countries and American countries. In modern days, they have become primarily associated with Asian cuisine, Japan which has the world's largest seaweed consumption per capita with 10-15%, is also accorded with significantly lower rate of cancer, thyroid diseases, heart diseases and dementia (Fitz Gerald et al., 2011). Today China, Japan and the republic of Korea are the largest consumer of seaweeds as food. Seaweeds are considered to be the food supplement for 21st century and as source for proteins, lipids, polysaccharides, minerals, vitamins and enzymes. In India, seaweeds are generally being used as raw material for the production of agar, alginates and seaweed liquid fertilizer, in spite of their great potential as therapeutic health booster, beauty enhancer and the source of nutrition. Hence it becomes essential to popularise seaweeds as health food which will help to feed undernourished people in India. For this purpose it requires a thorough study on the occurrence, availability and diversity of seaweeds both locally and regionally, their neutraceutical and therapeutical value which provide a baseline data for further exploration. So in the present study an attempt has been made picturise the diversity and availability status of seaweeds, found along the coast of Manapad, Gulf of Mannar, Thoothukudi.

Diversity of seaweeds:

Seaweeds were surveyed during four months viz., December, January, February and March at unique and distinct stations during low- tide in the year 2022-2023 by random sampling method. The study indicated that, Manapad was endowed with numerous taxa belonging to Chlorophycea (9 species), Phaeophyceae (12 species) and Rhodophyceae (9 species). Nine taxa were found as most abundant in this area. In Manapad region the post monsoon season (February to May) was noted for proliferous growth of different members of Chlorophyceae, Phaeophyceae and Rhodophyceae. The supply of nutrients from Manapad after the short rainy season, and the associated climatic conditions would probably favoured the growth and establishment of these seaweed species during post- monsoon season. Distributional diversity and abundance of species were high in the coast of Manapad. So this area is suitable for seaweed cultivation and harvest.

Table 1: Availability status of Chlorophycean seaweeds in Manapad, Gulf of Mannar

S.No	Seaweeds encountered	Availability status during months			
		December	January	February	March
1	Chaetomorpha antennina	+	+++	+++	+++
2	Caulerpa sertulariodes	+	+	+	++
3	Caulerpa scalpelliformis	+	++	++	++
4	Caulerpa taxifolia	+	+	+	+
5	Enteromorpha compressa	+	++	++	++
6	Halimeda macroloba	+	+	++	+
7	Halimeda tuna	++	++	+++	+++
8	Ulva fasciata	+	+	++	++
9	Ulva lactuca	+	++	+++	+++

Most abundant "+++", Abundant "++", Less abundant "+", Absent "-" Periodical collections were made in every month during low tide December, January, February and March.

Table 2: Availability status of Phaeophycean seaweeds in Manapad, Gulf of Mannar

S.No	Seaweeds encountered	Availability status during months			
		December	January	February	March
1	Sargassum wightii	+++	+++	+++	+++
2	Cystophyllum muriaticum	++	++	++	++
3	Sargassum polycystum	+	++	++	+++
4	Sargassum swartzii	++	+	++	++
5	Sargassum fluitans	++	+	+	+
6	Turbinaria ornata	++	++	+++	+++
7	Turbinaria conoides	++	++	+	+
8	Sargassum cristaefolium	+	+	+	+
9	Padina tetrastromatica	++	++	++	+++
10	Stoechospermum marginatum	+	++	++	+++
11	Padina gymnospora	++	+++	+++	+++
12	Spathoglossum asperum	+	+	+	++

Most abundant "+++", Abundant "++", Less abundant "+", Absent "-" Periodical collections were made in every month during low tide December, January , February and March.

Table 3: Availability status of Rhodophycean seaweeds in Manapad, Gulf of Mannar

S.No	Seaweeds encountered	Availability status during months			
		December	January	February	March
1	Gracilaria parvispora	+	+	+	++
2	Gracilaria corticata	+	++	+++	+++
3	Acanthophora spicifera	+	+	++	++
4	Hypnea valentiae	+	+	++	++
5	Botrycladia occidentalis	+	+	+	+
6	Hypnea musiformis	+	+	+	+
7	Rhodymenia palmate	+	+	+	+
8	Carynomorpha prismatica	+	+	+	+
9	Gracilaria verrucosa	+	+	+	+

Most abundant "+++", Abundant "++", Less abundant "+", Absent "-" Periodical collections were made in every month during low tide December, January, February and March.

CAULERPA SCALPELLIFORMIS

Kingdom: Plantae

Phylum : Chlorophyta

Class : Chlorophyceae

Order : Siphonales

Family : Caulerpaceae

Genus : Caulerpa

Species : scalpelliformis

DESCRIPTION:

The thallus is bright yellowish green to olive green is colour, with extensive stolonifous prostrate axes bearing upright fronds to stolons terate, naked attached to the substratum by fine rhizoidal branches. Upright branches terate near the base, bearing closely set, determinate lateral ramuli is an alternate distichous pattern.

SIZE:

Grows up to 20cm height 3 cm broad

HABITAT:

Found in the intertidal and subtidal region.

DISTRIBUTION:

Found abundantly along the coasts of Tamil Nadu, Maharashtra, Kerala and Karnataka.

USES:

This species is edible and the slightly sour and pungent nature "of the young and fresh thalli give it a spicy taste

ULVA LACTUCA

Kingdom: Plantae

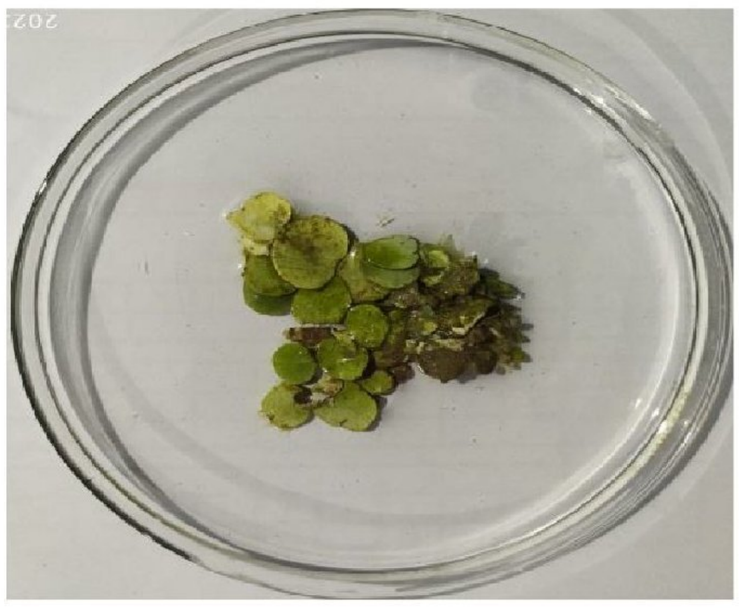
Phylum : Chlorophyta

Class : Chlorophyceae

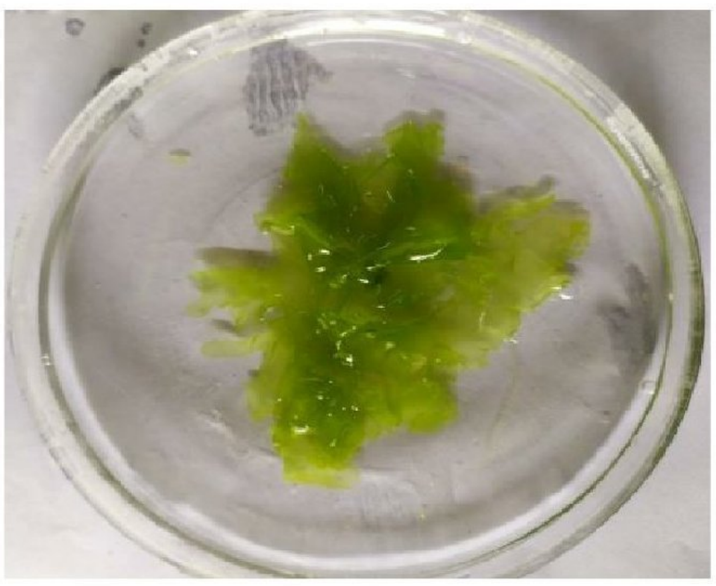
Order : Ulvales

Plate 2: Chlorophyceaen members encountered in Manapad of Gulf of Mannar, Thoothukudi District

Caulera scalpelliformis

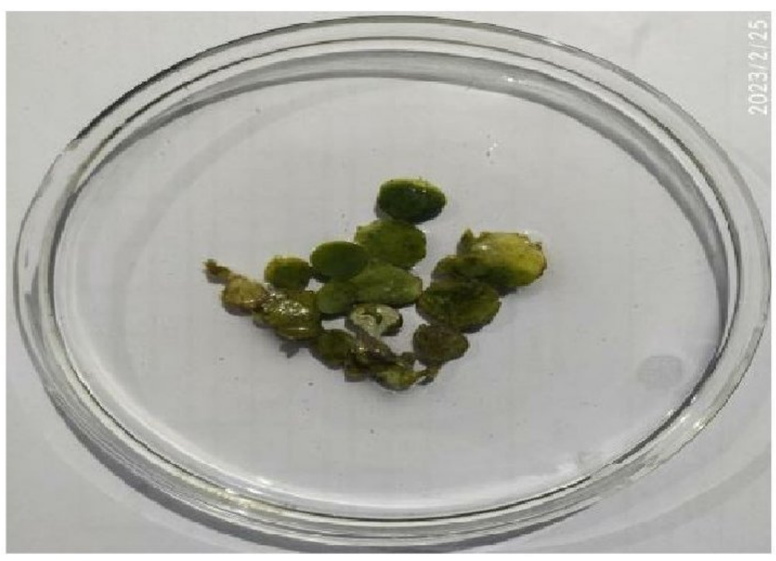


Halimeda tuna



Ulva lactuca

Chaetomorpha antennia



Halimeda macroloba

Ulva fasciata

Caulerpa sertularioides

Family : Ulvaceae

Genus : Ulva

Species : lactuca

DESCRIPTION:

The plant is bright to light green in colour. They are attached to the substratum by holdfast composed of rhizoidal out growth from the lower cells, the stalk inconspicuous or apparently absent, blade lanceolate to rounded, often somewhat lobed and undulate or folded and relatively broad. Margin of thallus ruffled and wary and folded thallus lobes varying is thickness. The rhozoids emerge to the substratum and become closely intermingled to one another forming a pseudoparenchymatous holdfast.

SIZE:

6 cm long

HABITAT:

Found along the intertidal region and in shallow waters of estuaries and mangroves.

DISTRIBUTION:

Along the coasts of Tamil Nadu, Gujarat, Maharashtra, Goa, Karnataka, Kerala and Lashadweep.

USES:

It is used as soap, mixed sea vegetables salad, cooked with other vegetables and meat.

CHAETOMORPHA ANTENNINA

Kingdom: Plantae

Phylum : Chlorophyta

Class : Chlorophyceae

Order : Cladophorales

Family : Cladophoraceae

Genus : Chaetomorpha

Species : antennina

DESCRIPTION:

Chaetomorpha antennina has a pantropical distribution and is easily identifiable by its characteristics erect, brush like tufts, composed of straight, rigid filaments borne on a long clavate basal cell with annular constrictions.

SIZE:

10 mm long

HABITAT:

Subtropical and tropical regions

DISTRIBUTION:

Along the coasts of Atlantic, Indian and Pacific Oceans

USES:

It is discovered to have antibacterial, antifungal, anti-inflammatory, antimalarial, antioxidant, antidote and radical scavenging properties.

HALIMEDA MACROLOBA

Kingdom: Plantae

Phylum : Chlorophyta

Class : Chlorophyceae

Order : Bryopsidales

Family : Halimedaceae

Genus : Halimeda

Species : macroloba

DESCRIPTION:

Halimeda is a genus of green macroalgae. The algal body (thallus) is composed of calcified green segments. Calcium carbonate is deposited in its tissues, making in inedible to most herbivores.

SIZE:

1.5 to 2cm wide and up to 3 cm tall

HABITATS:

Grown in fine, compact sand on the shallow reef flats in areas washed regularly by mild currents.

DISTRIBUTION:

Highly abundant in Indian ocean, Thai-Malay Peninsula and the Florida Keys

USES:

Bioindicators of the quality of water

CAULERPA SERTULARIOIDES

Kingdom: Plantae

Phylum : Chlorophyta

Class : Ulvophyceae

Order ; Bryopsidales

Family : Caulerpaceae

Genus : Caulerpa

Species : sertularioides

DESCRIPTION:

It is a small delicate green alga, also known as green feather algae is a species of seaweeds in the Caulerpaceae family found in warm water environments.

SIZE:

Each branchlet has a length of 3 to 11 mm (0.118 to 0.433 in)

HABITATS:

Found in tropical regions

DISTRIBUTION:

It is native to tropical waters, including the Caribbean, Indo-Pacific Indian ocean and Red sea.

USES:

It is used to antifungal, lowers blood pressure; source of sitostrerol, caulerpin and caulerpicin, palmitic acid.

ULVA FASCIATA

Kingdom: Plantae

Phylum : Chlorophyta

Class : Ulvophyceae

Order : Ulvales

Family : Ulvaceae

Genus : Ulva

Species : fasciata

DESCRIPTION:

Bright grass green to dark green, gold at margins when reproductive. May be colorless when stressed. Ulva fasciata, also known as limu palahalaha and sea lettuce, is a common green algae.

SIZE:

Cells usually square, 8-20µm wide, 14-40 µm long

HABITATS:

Found on intertidal rocks, in tidepools and on reef flats.

DISTRIBUTION:

Eastern Atlantic, Caribbean, Indian and Pacific Oceans.

USES:

Used for consumption in many parts of the world. High nutrients and fresh water are often indicated by its presence.

SARGASSUM POLYCYSTUM

Kingdom: Plantae

Phylum : Phaeophyta

Class : Phaeophyceae

Order : Fucales

Family : Sargassaceae

Genus : Sargassum

Species : palycystum

DESCRIPTION:

Thallus yellowish brown colour attached with discoid holdfast, main axis cylindrical and rough due to the presence of numerous out growth. Supporting alternatively arranged branches bearing leaves and vesicles in young thalli leaves are longer and broader measuring long including the stalk and wide. Leaves are generally ablong slightly tapered refuse or emerginate at the tip finally serrated throughout the margin mature thalli fewer leaves smaller.

SIZE:

Thallus 35 cm tall, leaves 7-15 cm long including stalk and 17-4 nm.

HABITAT:

Found on coralline rocks in wave exposed low intertidal to shallow subtidal areas.

DISTRIBUTION:

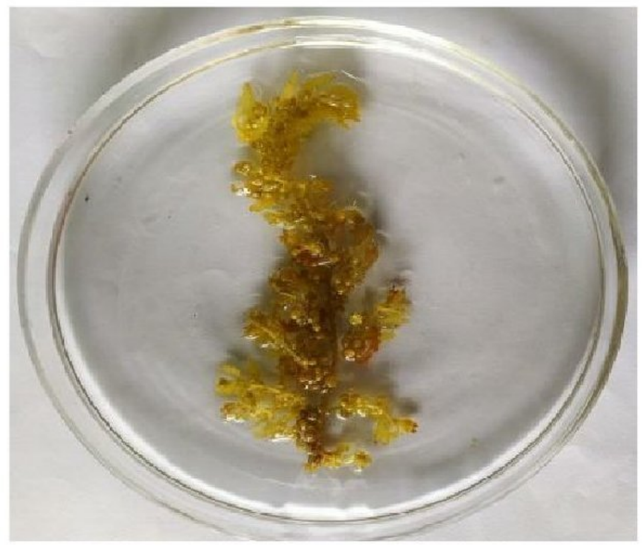
Widely distributed in the tropics including the India, Japan, Indonesia, Vietnam, Srilanka and China.

USES:

Extraction of alginate, fertilizer and vegetables.

Plate 3: Phaeophyceaen members encountered in Manapad of Gulf of Mannar, Thoothukudi District

Spathoglossum asperum



Sargassum cristaefolium

Sargassum swartzii

Sargassum wightii

Sargasso fluitans

cystophyllum muriaticum

Turbinaria ornata

Sargassum polycystum

Turbinaria conoides

Padina tetrastromatica

Stechospermum marginatum

Padina gymnospora

SARGASSUM SWARTZII

Kingdom: Plantae

Phylum : Phaeophyta

Class : Phaeophyceae

Order : Fucales

Family : Sargassaceae

Genus : Sargassum

Species : swartzii

DESCRIPTION:

Main axis short, cylindrical, smooth, bearing radially arranged several primary branches with thick, elongate lanceolate phylloids. Phylloids with asymmetrical base, percurrent midrib, entire or shallowly dentate margins and acute apices.

SIZE:

5-13 mm long

HABITAT:

Found in tropical and subtropical regions

DISTRIBUTION:

Distributed in India, Japan, Indonesia, Vietnam, Srilanka, China.

USES:

Used for its larvicidal, anti-HIV-1, anti inflammatory and analgesic activities

.

SARGASSUM WIGHTII

Kingdom: Plantae

Phylum : Phaeophyta

Class : Phaeophyceae

Order : Fucales

Family : Sargassaceae

Genus : Sargassum

Species : wightii

DESCRIPTION:

It is a brown seaweed contains several bioactive compounds such as fucoidan fucoxanthin along with substantial quantities of polyphenols (phloroglucinol, phlorotannins, phenolic acids etc.) and flavonoids (flavonols and flavonol glycosides).

SIZE:

Size ranging 8-12 nm

HABITAT:

Found in tropical and subtropical regions

DISTRIBUTION:

Distributed in India, China, Japan, Srilanka

USES;

Used as fertilizer, mulch or compost

SARGASSUM FLUITANS

Kingdom: Plantae

Phylum : Phaeophyta

Class : Phaeophyceae

Order : Fucales

Family : Sargassaceae

Genus : Sargassum

Species : fluitans

DESCRIPTION:

It is a type of seaweed, or brown alage. Pelagic plants without a holdfast or a distinct main axis. Branches smooth or with few spiny projections, terete or sometimes compressed 1-1.8 mm diam., ramified several times.

SIZE:

1 to more than 100 meter

HABITAT:

Found in tropical and subtropical regions

DISTRIBUTION:

Western Central Atlantic and Western Indian Ocean

USES:

Used as pharmacological activities.

CYSTOPHYLLUM MURIATICUM

Kingdom: Plantae

Phylum : Phaeophyta

Class : Phaeophyceae

Order : Fucales

Family : Sargassaceae

Genus : Crystophyllum

Species : muriaticum

DESCRIPTION:

It is an abundant brown algae. Leaves are golden brown with dots and a clear spine. Edges of leaves are smooth or spiny with toothlike edges. It can float due to the pneumatocysts found on the leaves.

SIZE:

It can reach upto 30 cm with a flat main branch and wide.

HABITAT:

Found in tropical and subtropical regions.

DISTRIBUTION:

Found in Indian oceans.

USES:

It is usually chopped or ground up and combined with other seaweeds or cooked in soup. Whole leaves are deep fried into chips. It is also eaten fresh at the beach with raw fish or octopus. It is also used for fish bait.

SARGASSUM CRISTAEFOLIUM

Kingdom: Plantae

Phylum : Phaeophyta

Class : Phaeophyceae

Order : Fucales

Family : Sargassaceae

Genus : Sargassum

Species : cristaefolium

DESCRIPTION:

The thalli large, dark brown attached to rocks by a disc shaped holdfast. This seaweed has a short stem, giving rise to 1 or 2 main compressed and smooth primary branches leaves generally oblong, with coarsely toothed outer margins. Distal portions of leaves have thick duplicate margins.

SIZE:

Grows up to a height of 50 cm

HABITAT:

Found on coralline rocks in wave exposed low intertidal to shallow subtidal areas.

DISTRIBUTION:

Widely distributed in the tropics including the India, Japan, Indonesia, Vietnam, Srilanka and China.

USES:

Extraction of alginate, fertilizers and vegetables.

PADINA TETRASTROMATICA

Kingdom: Plantae

Phylum : Phaeophyta

Class : Phaeophyceae

Order : Dictyotales

Family : Dictyotaceae

Genus : Padina

Species : tetrastromatica

DESCRIPTION:

It is brown and yellow in colour when dried they become olive green, basal portion forming a thick rhizomatous disc attached into sand accumulated in pond of mid-littoral region with short stipe. Its thalli are irregularly cleft into narrow lobes, stupose on both surfaces in the basal part of the blade to about one fourth of the length usually divided into a flabellate lobes in young plants and divided to the base blade into many cuneate lobes of about 2-4 cm wide piliferous zones on both surfaces occurring alternately and well developed with glabrous zone between them. Upper portion is broader and forms the fan-shaped and striped frond, apical margins are involute, the lower region of thallus becomes gradually of the thallus and form concentric rows, which render the thallus striped.

SIZE:

Grows up to 10-10 cm is height and 5-7 cm in breadth.

HABITAT:

Grows in intertidal regions

DISTRIBUTION:

Gulf of Mannar, Maharashtra and Kerala coasts.

USES:

Extraction of alginate, fertilizer

STECHOSPERMUM MARGINATUM

Kingdom: Plantae

Phylum : Phaeophyta

Class : Phaeophyceae

Order : Dictyotales

Family : Dictyotaceae

Genus : Stechospermum

Species : marginatum

DESCRIPTION:

Thallus flat, erect, spathulate, dichotomously branched without a midrib, margin entire apex bifid.

SIZE:

Grow upto 20-30 cm long and 8-11 cm broad.

HABITAT:

Commonly found in intertidal zone.

DISTRIBUTION:

Tamil Nadu, Gujarat, Ratnagiri, Malvan, Maharashtra, Goa, Karwar, Honawar, Bhatkal and Karnataka.

USES:

Used as a source of alginate, fertilizer.

SPATHOGLOSSUM ASPERUM

Kingdom: Plantae

Phylum : Phaeophyta

Class : Phaeophyceae

Order : Dictyotales

Family : Dictyotaceae

Genus : Spathoglossum

Species : asperum

DESCRIPTION:

Plants with an indistinct small holdfast. Thallus flat, palmate, sub dichotomously divided into larger and smaller lobes. Lobes elongate, linear, lanceolate, alternated towards base. Apex acute or rounded. Margin sinnate, irregularly dentate with larger or smaller proliferations.

SIZE:

Upto 28 cm height

HABITAT:

Tropical and subtropical regions

DISTRIBUTION:

Found in India, Kasumi, Haygood, Japan

USES:

Used for preparation of liquid fertilizer

GRACILARIA PARVISPORA

Kingdom: Plantae

Phylum : Rhodophyta

Class : Rhodophyceae

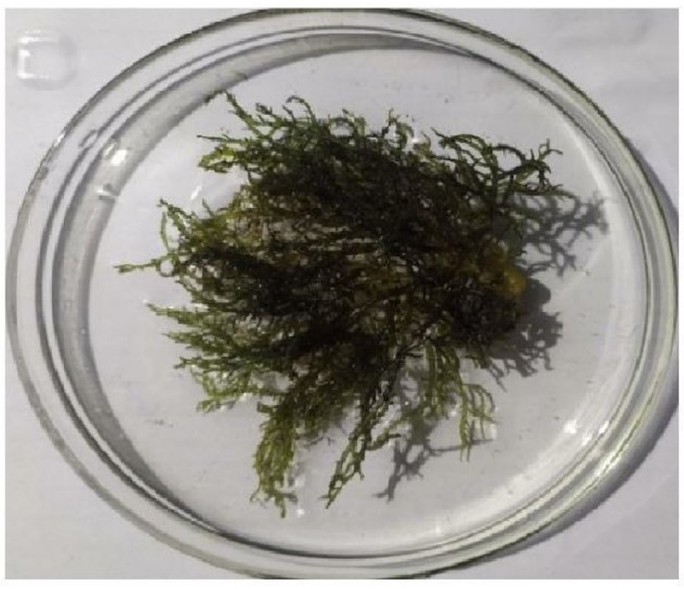
Order : Gracilariales

Plate 4: Rhodophyceaen members encountered in Manapad of Gulf of Mannar, Thoothukudi District

Gracilaria parvispora

Acanthophora spicifera

Gracilaria corticata



Hypnea valentiae

Gracilaria verrucosa

Rhodomenia palmata

Botrycladia occidentalis

Hypnea musiformis

Carynomorpha prismatica

Family : Gracilariaceae

Genus : Gracilaria

Species : parvispora

DESCRIPTION:

It contains erect thallus arise from a small discoid holdfast. The thallus are generally cylindrical, depressed or blade shaped with lateral, alternate or subdichotomous branches. Sometimes different branches may be found in one plant.

SIZE:

The plants grows tall, to 30 cm or more, with a single dominant axis, 0.8-3.5 mm diameter.

HABITAT:

Found in tropical and temperate regions

DISTRIBUTION:

Indian oceans, Hawaii islands and Pacific oceans

USES:

It used as a nutritious food source for various herbivorous fish and invertibrates, including tangs, rabbitfish

ACANTHOPHORA SPICIFERA

Kingdom: Plantae

Phylum : Rhodophyta

Class : Florideophyceae

Order : Ceramiales

Family : Rhodomelaceae

Genus : Acanthophora

Species : spicifera

DESCRIPTION:

It is an erect macroalgae which grows up to 40 cm tall. It has solid cylindrical branches, 2-3 mm wide, branched either sparingly or repeatedly. The main branches have short, determinate branches, irregularly shaped and spinose, with spines numerous and radially arranged.

SIZE:

Thalli are tall 10-25 cm long in height.

HABITAT:

Found in most tropical or subtropical seas of the world

DISTRIBUTION:

Found in Indian oceans and Pacific oceans

USES:

Used in vegetable salads, as soup flavouring and as a thickening agent in the Phillippines, and is reported to contain carragenaans, used as an emulsifying agent.

HYPNEA VALENTIAE

Kingdom: Plantae

Phylum : Rhodophyta

Class : Florideophyceae

Order : Gigartinales

Family : Cystocloniaceae

Genus : Hypnea

Species : valentiae

DESCRIPTION:

It is characterized by membranous or cartilaginous thalli which can be either erect or prostrate. The thallus features a main axis with an apex of varying shapes (straight, curved, tendril or bifurcated)

SIZE:

1.5 to 2.4 mm long

HABITAT:

Found in tropical or subtropical regions

DISTRIBUTION:

Distributed in Indian Ocean: from the Arabian sea south to Madagascar, including the Red Sea, Persian Gulf, Oman Sea, Aldabra Islands, Seychelles, Reunion and Mauritius, east to India and south to the Andaman Sea; in Australia, from Western Australia to Victoria

USES:

Used as animal feed, medicine, and as fertilizer.

BOTRYCLADIA OCCIDENTALIS

Kingdom: Plantae

Phylum : Rhodophyta

Class : Rhodophyceae

Order : Rhodymeniales

Family : Rhodymeniaceae

Genus : Botrycladia

Species : occidentalis

DESCRIPTION:

It is a species of red algae in the family Rhodymeniaceae.

SIZE:

2 cm long

HABITAT:

Tropical and subtropical regions

DISTRIBUTION:

Found in Indian ocean

USES:

Used as cosmetic ingredients

HYPNEA MUCIFORMIS

Kingdom : Plantae

Phylum : Rhodophyta

Class : Florideophyceae

Order : Gigartinales

Family : Cystocloniaceae

Genus : Hypnea

Species : muciformis

DESCRIPTION:

Firm, cartilaginous, highly branched. Branching is variable and irregular, often tendril like and twisted around axes of other algae. The ends of many axes and branches are flattened with broad hooks. Holdfasts are small, inconspicuous or lacking.

SIZE:

10-20 cm tall, 0.5-1.0 cm diameter

HABITAT:

Found in calm intertidal and shallow subtidal reef flats, tidepools and on rocky intertidal benches.

DISTRIBUTION:

Found in Tamil Nadu and coastal of Maui

USES:

It is important for fishermen who had to spend days at sea waiting to catch fish.

RHODYMENIA PALMATA

Kingdom : Plantae

Phylum : Rhodophyta

Class : Florideophyceae

Order : Rhodymeniales

Family : Rhodymeniaceae

Genus : Rhodymenia

Species : palmate

DESCRIPTION:

Reddish brown, membranous or leathery, flattened fronds,arising from a discoid base, usually with a small stipe expanding gradually to form simple or dichotomously and palmately divided fronds, often with characteristic marginal leaflets.

SIZE:

15 cm long and 1.5 cm widw thallus

HABITAT:

Tropical regions

DISTRIBUTION:

Grows on the northern coasts of the Atlantic , Pacific Oceans and Indian Oceans

USES:

Used in soups, chowders, sandwiches, and salads or added to bread or pizza dough

CORYNOMORPHA PRISMATICA

Kingdom : Plantae

Phylum : Rhodophyta

Class : Rhodophyceae

Order : Cryptonemiales

Family : Siphonodentaliidae

Genus : Corynomorpha

Species : prismatica

DESCRIPTION:

It contain long erect thallus

SIZE:

2-10 cm long

HABITAT:

Found in tropical regions

DISTRIBUTION:

Found in Indian oceans.

GRACILARIA VERRUCOSA

Kingdom: Plantae

Phylum : Rhodophyta

Class : Rhodophyceae

Order : Gracilariales

Family : Gracilariaceae

Genus : Gracilaria

Species : verrucosa

DESCRIPTION:

Erect thallus arise from a small discoid holdfast

SIZE:

1-4 mm in diameter

HABITAT:

Tropical and temperate regions

DISTRIBUTION:

Found in Indian Oceans

USES:

Used as anticancer agents

PHYTOCHEMICAL ANALYSIS:

Phytochemicals are chemical compounds that are naturally found in seaweeds. They are responsible for the colour and organoleptic properties of the seaweeds. It is also reoffered to as those chemicals that may have biological significance but are not established as an cuentials nutrient in sea weeds. Phytochemicals could be available as dietary supplements, but the parential health benefits of phytochemicals are derived from consumption of the whole part of the seaweed. Several phytochemicals have a wide range of activities, which helps to give immunity against long term disease.

Phytochemical constituents in seaweed samples are considered to be biologically active compounds with a variety of functions including antioxidant and antimicrobial properties. The phytochemical analysis of different seaweed extracts (ethanol, methanol, acetone, hexane and aqueous) of Turbinaria ornata, Gracilaria corticata and Halimeda tuna were found to contain alkaloids, flavonoids, phenols, quinines, carbohydrates, saponins, terpenoids, steroids, tannins, glycosides, proteins and coumarin (Table 4, 5 and 6).

Phytochemistry takes into account the structural compositions of these metabolites, the biosynthetic pathways, functions, mechanisms of actions in the living systems and it's medicinal, industrial, and commercial applications. The proper understanding of phytochemical is essential for drug discovery and for the development of novel therapeutic agents against major diseases.

The study revealed that the seaweed extracts of Turbinaria ornata was showing the maximum presence of phenols, steroids, coumarin, alkaloids and carbohydrates in all solvents.

Table 4: Preliminary phytochemical screening of different extracts of Halimeda tuna

S.No	Compounds	Ethanol	Methanol	Acetone	Hexane	Aqueous
1	Alkaloids	+	-	-	-	+++
2	Flavonoids	+	+	+	+++	-
3	Phenols	+	-	+	-	++
4	Quinines	++	++	++	+++	-
5	Carbohydrates	+++	+++	+++	-	-
6	Saponins	-	-	-	-	-
7	Terpenoids	+++	-	+++	+	-
8	Steroids	-	++	+++	-	-
9	Tannins	-	-	-	-	-
10	Glycosides	-	-	-	-	-
11	Proteins	-	-	-	-	-
12	Coumarins	-	-	-	-	-

Most abundant '+++', Abundant '++', Less abundant '+', Absent '-'

Table 5: Preliminary phytochemical screening of different extracts of Turbinaria ornata

S.No	Compounds	Ethanol	Methanol	Acetone	Hexane	Aqueous
1	Alkaloids	+	+++	-	+++	++
2	Flavonoids	++	-	++	-	+++
3	Phenols	+++	+++	+	++	++
4	Quinines	-	-	-	-	+++
5	Carbohydrates	++	++	+++	+	-
6	Saponins	-	-	-	-	-
7	Terpenoids	-	-	+++	+++	+
8	Steroids	+++	++	++	++	++
9	Tannins	-	-	+	-	-
10	Glycosides	++	+	-	-	+++
11	Proteins	-	-	-	-	-
12	Coumarins	+++	++	++	+	++

Most abundant '+++', Abundant '++', Less abundant '+', Absent '-'

Table 6: Preliminary phytochemical screening of different extracts of Gracilaria corticata

S.No	Compounds	Ethanol	Methanol	Acetone	Hexane	Aqueous
1	Alkaloids	+++	++	+	-	+
2	Flavonoids	-	-	+	++	+
3	Phenols	+	++	++	-	+++
4	Quinines	-	-	-	-	-
5	Carbohydrates	-	+++	+++	-	-
6	Saponins	-	-	-	-	-
7	Terpenoids	+++	-	+++	++	-
8	Steroids	++	+++	+++	-	+++
9	Tannins	-	-	-	-	-
10	Glycosides	-	-	-	+	-
11	Proteins	-	-	-	-	-
12	Coumarins	-	-	-	+++	+

Most abundant '+++', Abundant '++', Less abundant '+', Absent '-'

Steroids in modern clinical studies have supported their role as anti inflammatory and analgesic agents (Perumal, 2012). Saponins and proteins are completely absent in all extracts of Turbinaria ornata. Flavonoids and Glycosides were highly present in ethanol and aqueous extracts of Turbinaria ornata. Terpenoids were highly present in acetone, hexane and aqueous extracts of Turbinaria ornata.

Seaweed extracts of Gracilaria corticata showing the maximum presence of alkaloids, phenols and steroids in all solvents, but absent in hexane. Quinines, Saponins, tannins and proteins are completely absent in all extracts of Gracilaria corticata. Flavonoids and Terpenoids were highly present in acetone and hexane solvents of Gracilaria corticata. Flavonoids are a group of plant metabolites thought to provide health benefits through cell signalling pathways and antioxidant effects. Carbohydrates was present in methanol and acetone solvents of Gracilaria corticata.

Halimeda tuna seaweed extracts showing the maximum presence of Flavonoids and quinines in all solvents, but absent in aqueous solution. Saponins, tannins, glycosides, proteins, Coumarins are completely absent in all extracts of Halimeda tuna. Phenols, carbohydrates, terpenoids, steroids are highly present in acetone solvent of Halimeda tuna. Terpenoids are known to posses a wide range of biological activities including antimicrobial, antifungal, antiphrastic, antiviral, antimalarial, antispasmodic, antihyperglycemic, anti-inflammatory, immunomodulatroy properties. Phenols, carbohydrates and terpenoids are present in ethanol extract of Halimeda tuna. Alkaloid was adequately present in ethanol and aqueous extracts of Halimeda tuna.

The seaweeds are rich in secondary metabolites which include alkaloids, flavonoids, saponins and related active metabolites which are of great medicinal value and have been active metabolites which are of great medicinal value and have been extensively used in the drug and pharmaceutical industry. These secondary metabolites are reported to have many biological and therapeutical properties.

ANTIBACTERIAL ACTIVITY:

Seaweeds are considered as source of bioactive compounds and produce a great variety of secondary metabolites characterized by a broad spectrum of biological activities.

Table 7: Antibacterial Activity of Halimeda tuna extracted with different solvent against human pathogen

S.No	Organism	Zone of inhibition (mm)					
		Ethanol	Methanol	Acetone	Hexane	Aqueous	Streptomycin
1	E.coli	21	16	20	4	4	23
2	S.aureus	15	5	5	10	5	20
3	B.subtilis	6	5	5	6	NS	15

Control = streptomycin (100µg/ ml)

Seaweed extract = 2.5 mg/ ml (effective concentration)

NS = No sensitivity

Table 8: Antibacterial Activity of Turbinaria ornata extracted with different solvent against human pathogen

S.No	Organism	Zone of inhibition (mm)					
		Ethanol	Methanol	Acetone	Hexane	Aqueous	Streptomycin
1	E.coli	18	17	19	5	4	20
2	S.aureus	15	16	8	9	7	30
3	B.subtilis	5	4	4	5	NS	25

Control = streptomycin (100µg/ ml)

Seaweed extract = 2.5 mg/ ml (effective concentration)

NS = No sensitivity

Table 9: Antibacterial Activity of Gracilaria corticata extracted with different solvent against human pathogen

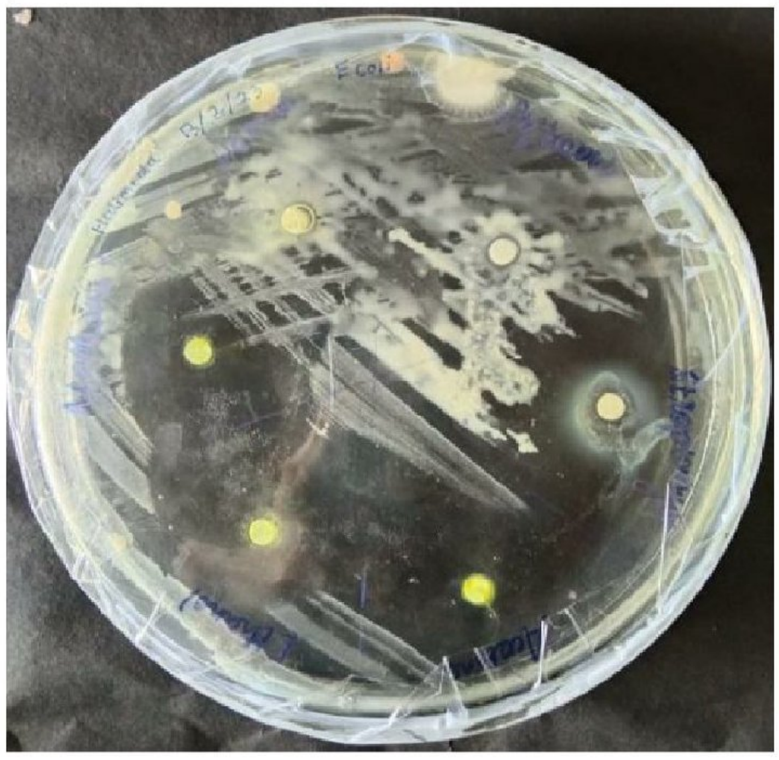
S.No	Organism	Zone of inhibition (mm)					
		Ethanol	Methanol	Acetone	Hexane	Aqueous	Streptomycin
1	E.coli	20	12	7	5	5	5
2	S.aureus	8	5	5	5	NS	21
3	B.subtilis	5	5	5	5	NS	23

Control = streptomycin (100µg/ ml)

Seaweed extract = 2.5 mg/ ml (effective concentration)

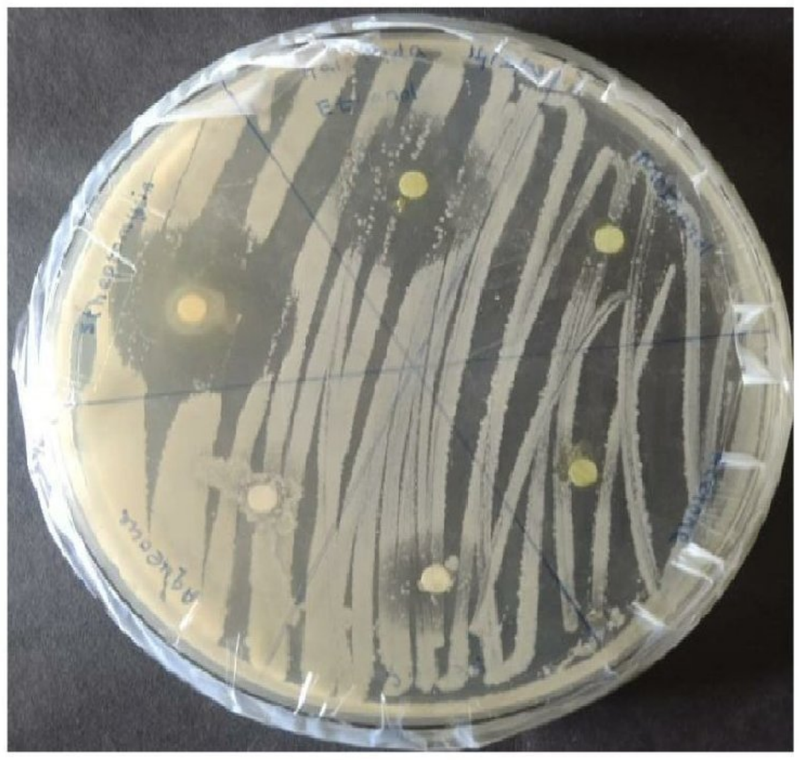
NS = No sensitivity

Plate 5: In vitro Antibacterial activity of Halimeda tuna seaweed extracts against human pathogens



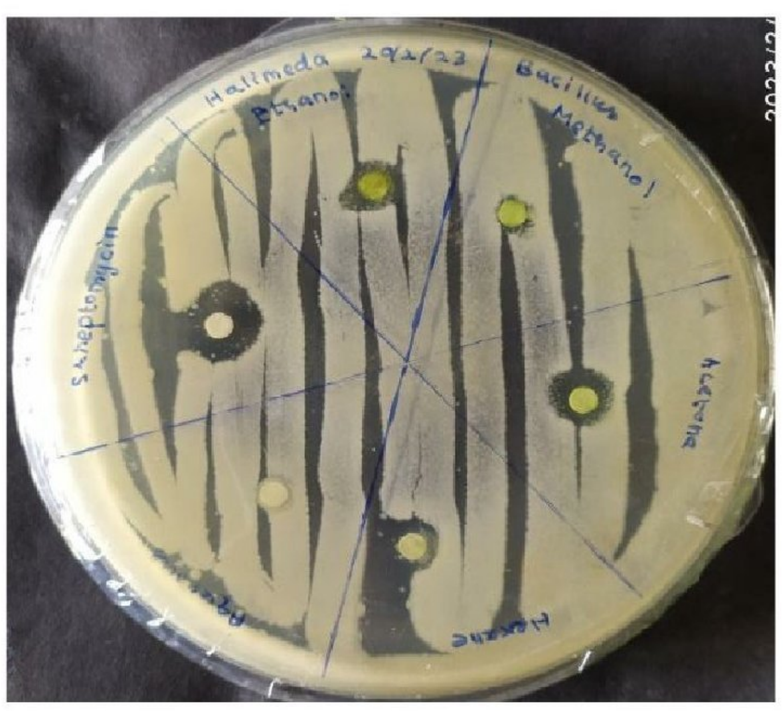
Halimeda tuna extract against

Escherichia coli



Halimeda tuna extract against

staphylococcus aureus

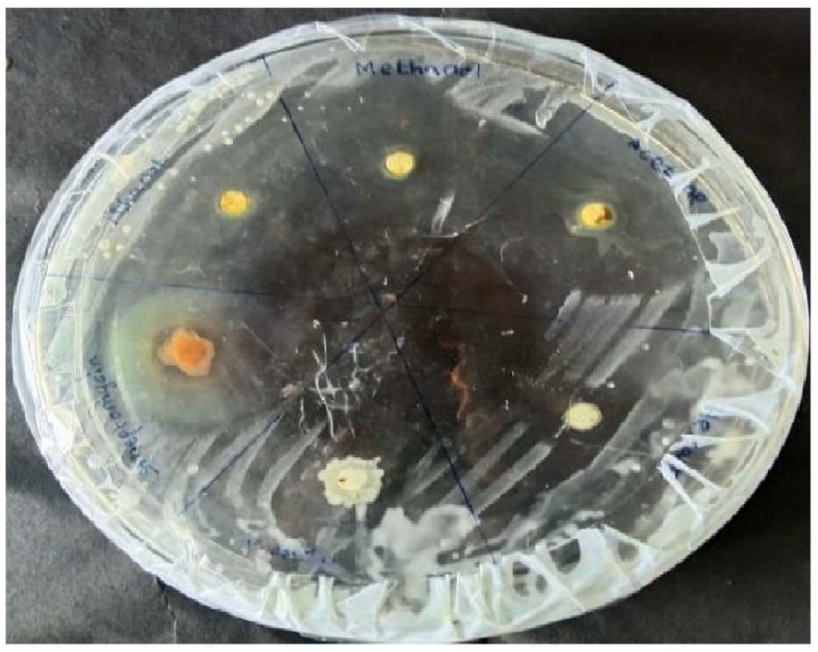


Halimeda tuna extract against

Bacillus subtilis

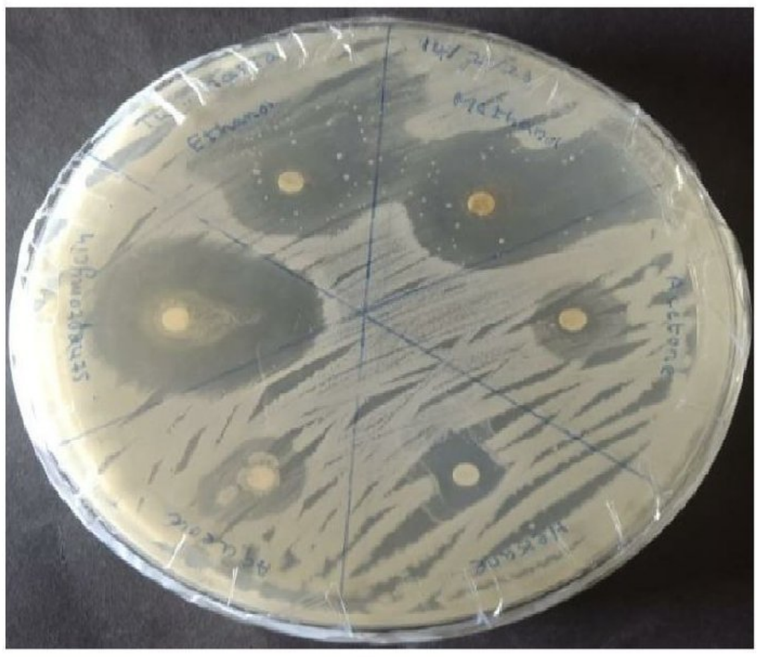
Antibacterial activity is revealed as clear zone around the disc and is represented as zone of inhibition

Plate 6: In vitro Antibacterial activity of Turbinaria ornata seaweed extracts against human pathogen



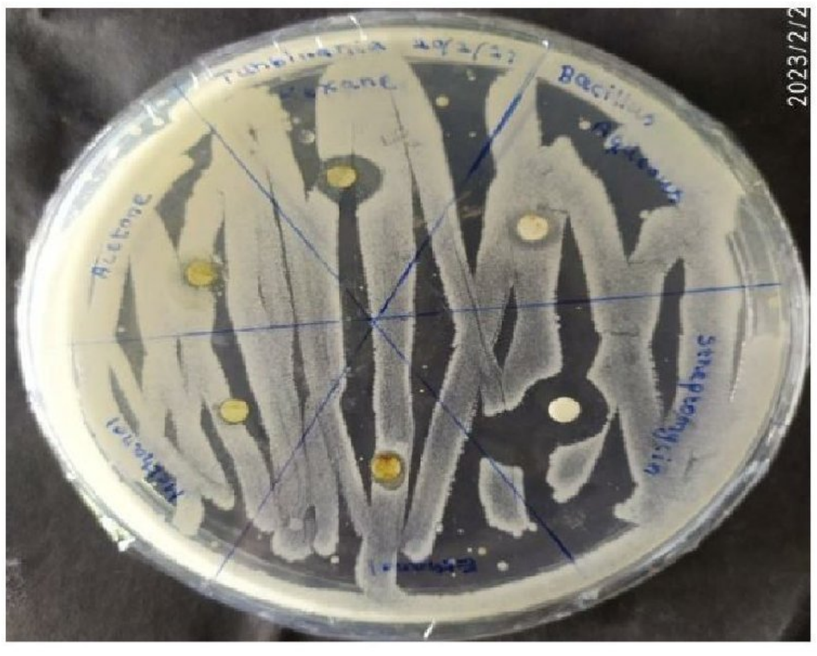
Turbiaria ornata extract against

Escherichia coli



Turbinaria ornata extract against

Staphylococcus aureus

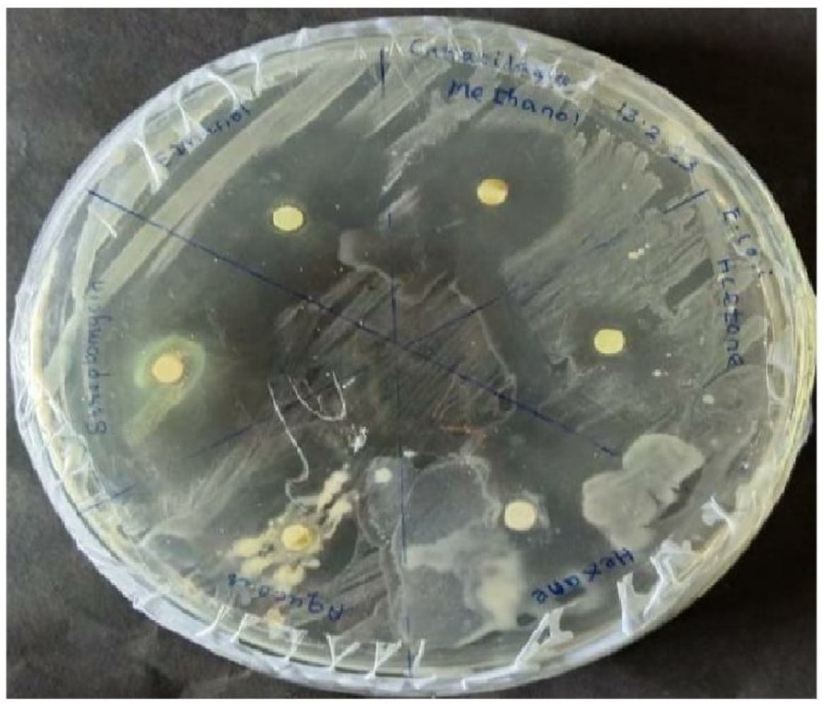


Turbinaria ornata extract against

Bacillus subtilis

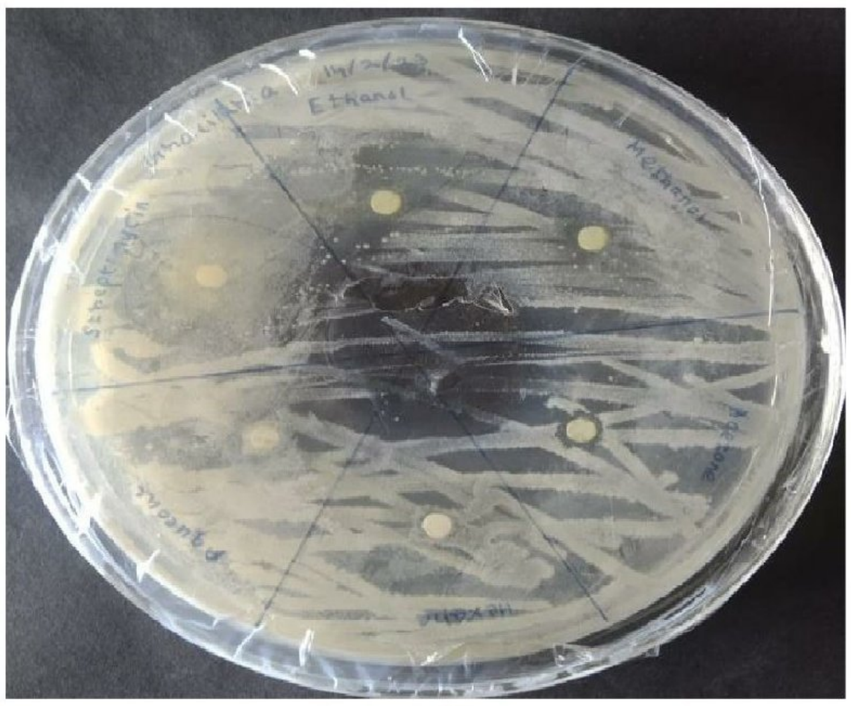
Antibacterial activity is revealed as clear zone around the disc and is represented as zone of inhibition

Plate 7: In vitro Antibacterial activity of Gracilaria corticata seaweed extracts against human pathogens



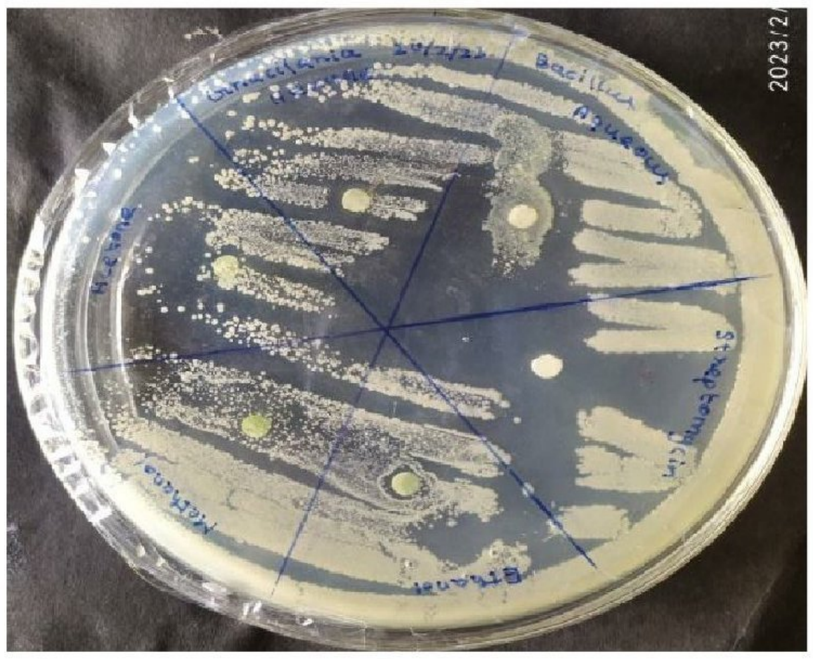
Gracilaria corticata extract against

Escherichia coli



Gracilaria corticata extract against

Staphylococcus aureus



Gracilaria corticata extract against

Bacillus subtilis

Antibacterial activity is revealed as clear zone around the disc and is represented as zone of inhibition

Compounds with cytostatic antiviral, anthelminthic, antifungal and antibacterial activities have been detected in green, brown and red seaweeds (Ganesan et al., 2008).

In the present study, the antibacterial activity of three seaweeds extracts (Halimeda tuna, Turbinaria ornata and Gracilaria corticata) in five different solvents (ethanol, methanol, acetone, hexane and aqueous) was measured against three human pathogenic bacteria (E.coli, B.subtilis and S.aureus) and the results were presented in (Table: 7, 8 and 9). The inhibition zones against these species ranged in size from 4 to 30mm in diameter. All of the extracts examined prevented the growth of all of the pathogens in the sample. Ethanolic extract of Halimeda tuna had the highest activity against E.coli (21mm). Hexane and aqueous extract of Halimeda tuna had lower inhibitory activity against all the pathogens tested in the study. The methanol and acetone extracts of Halimeda tuna assessed to be more antagonistic against E.coli. Methanol was the most effective solvent for extracting antibacterial compounds from selected seaweeds (Vlachos et al., 1996)

As shown as Table 8, acetone extract of Turbinaria ornata exhibited maximum activity against, E.coli (19mm). Similarly ethanol (18mm), methanol (17mm) and hexane (5mm) extracts of E.coli showed the inhibition zone. All the extracts of Turbinaria ornata show the lower inhibitory activity against B.subtilis. Ethanol and methanol extracts of Turbinaria ornata shows the higher inhibitory activity against S.aureus, orderly 15mm and 16mm inhibition zone.

Ethanolic extract of Gracilaria corticata interacted with E.coli and resulted into growth inhibition (20mm) closed to streptomycin. Similarly E.coli was arrested by G.corticata in methanol extracts (12mm). However Staphylococcus aureus was found to be resistant to Gracilaria corticata when compared to other organisms. Aqueous extract of G.corticata was observed to be less effective.

The antibacterial activity of these seaweeds may be possibly due to the alkaloids and phenolic compounds such as phlorotannins, amines and antioxidants that have strong bacterial activity (Nagayama et al., 2002) Variation in antibacterial activity may be due to the method of extraction, solvent and season at which samples were collected. Therefore it is concluded all the seaweeds used in the present study could be effectively processed to be utilized as a source for antibacterial therapeutic drug preparations.

Table 10 : Anthelmintic Activity of Halimeda tuna

Treatment	Group	Concentration mg/ml	Time taken for paralysis	Time taken for death (min)
Control (Saline)	I	10	---	---
Ethanol	II	10	2	3
Methanol	III	10	2	3
Acetone	IV	10	2	3
Hexane	V	10	15	25
Aqueous	VI	10	20	40
Albendazole	VII	10	13	17

Control = Albendazole (10mg/ml)

Seaweed extract = 10mg/ml

Table 11 : Anthelmintic Activity of Turbinaria ornata

Treatment	Group	Concentration mg/ml	Time taken for paralysis	Time taken for death (min)
Control (Saline)	I	10	---	---
Ethanol	II	10	2	3
Methanol	III	10	2	3
Acetone	IV	10	2	4
Hexane	V	10	19	28
Aqueous	VI	10	23	39
Albendazole	VII	10	12	15

Control = Albendazole (10mg/ml)

Seaweed extract = 10mg/ml

Table 12: Anthelmintic Activity of Gracilaria corticata

Treatment	Group	Concentration mg/ml	Time taken for paralysis	Time taken for death (min)
Control (Saline)	I	10	---	---
Ethanol	II	10	2	3
Methanol	III	10	2	3
Acetone	IV	10	2	4
Hexane	V	10	15	25
Aqueous	VI	10	25	38
Albendazole	VII	10	14	16

Control = Albendazole (10mg/ml)

Seaweed extract = 10mg/ml

Plate 8: Anthelmintic activity of Halimeda tuna

Plate 9: Anthelmintic activity of Turbinaria ornata

Plate 10 : Anthelmintic activity of Gracilaria corticata

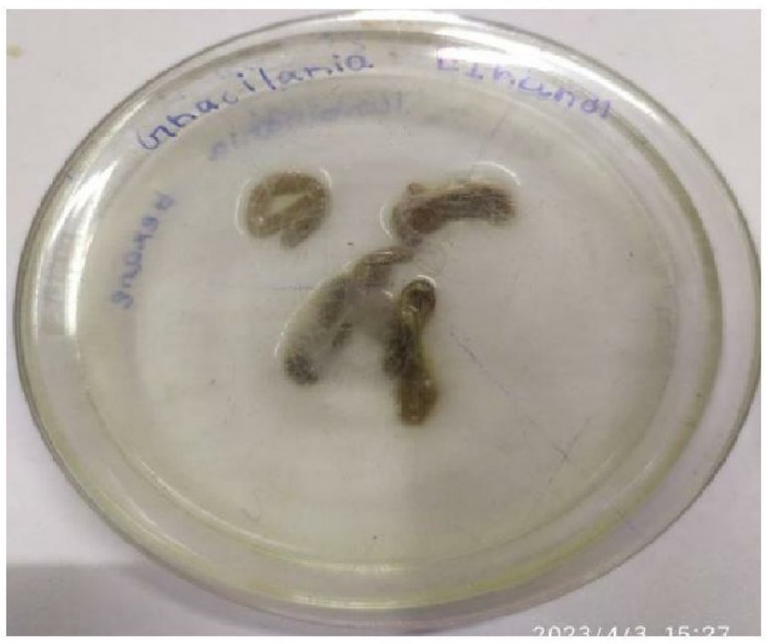
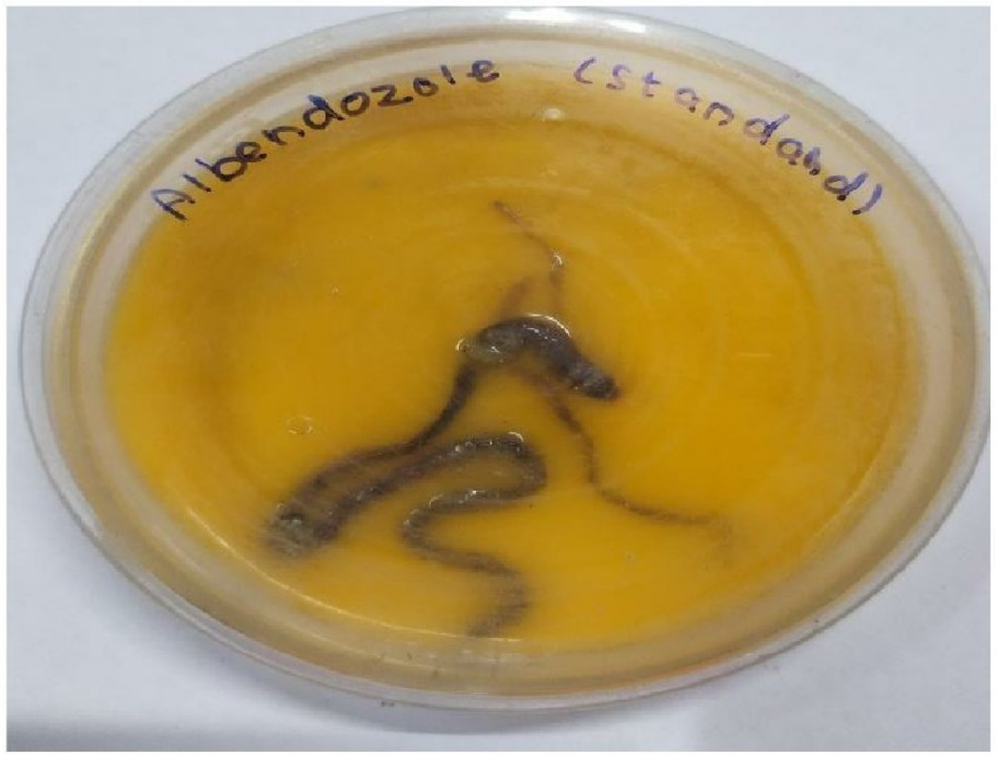


Plate 11: Anthelmintic activity in Saline water and Albendozole



ANTHELMINTIC ACTIVITY:

The word "helminth" comes from the greek word, "helminths", which means "worm". Helminth is a general word that refers to a variety of parasitic worm that live with the body. According to the World Health Organisation (WHO), over two billion people are infected with parasitic worms. By 2025, it is projected that 57% of the population in developing countries will be affected. Anthelmintics are drugs that work locally to remove worms from the GIT or systemically to eliminate adult helminths or development forms that infect organs and tissue. Abdominal pain, loss of appetite, nausea, vomiting, headache and diarrhoea are common side effects of current anthelmintics. Nature anthelmintics can play an important role in the treatment of these parasite infection.

As shown in table (10,11 and 12) the ethanol, methanol and acetone extracts of Halimeda tuna , Turbinaria ornata and Gracilaria corticata shows more potent than hexane and aqueous extracts. The hexane extract of Halimeda tuna at 10 mg/ml concentration shows paralysis at 15 minutes and death at 25 minutes. The hexane extract of Turbinaria ornata at 10 mg/ml concentration was taken at 19 minutes for paralysis and death at 28 minutes. The hexane extract of Gracilaria corticata caused paralysis at 15 minutes and time of death at 25 minutes.

In aqueous extract of Halimeda tuna showed paralysis at 20 minutes and death at 40 minutes. Aqueous extract of Turbinaria ornata at 10 mg/ml concentration showed paralysis at 23 minutes and death at 39 minutes. In Gracilaria corticata aqueous extract caused paralysis at 25 minutes and time of death at 38 minutes. The control (saline solution treated) earthworms were observed for 24 hrs in which no paralysis and death was noticed. So this selected seaweeds play an natural anthelmintic in the treatment of Parasite infection.

FT-IR SPECTROSCOPY

The FTIR analysis was carried out to predict the functional groups present in the seaweed extracts of Halimeda tuna, Turbinaria ornata and Gracilaria corticata. The results of FTIR spectral were presented in the Table 13,14 and 15.

Table 13 : FT-IR Spectroscopy analysis of Halimeda tuna

S.No	Peak value	Bond	Functional group
1	469.63	Alkyl halides	C-Br
2	523.64	Alkyl halides	C-Br
3	712.65	Alkyl halides	C-CL
4	855.37	Alkyl halides	C-CL
5	910.34	Aliphatic nitro group	C-N
6	1027.02	Sulfonic acids	S-O
7	1081.03	Primary alcohol	C-O
8	1163.96	Poly fluorinated compound	C-F
9	1491.84	Alkanes	H-C-H
10	1658.67	Alkene	C=C
11	1786.92	Acid halide	C=O
12	2521.75	Carboxylic acid	O-H
13	2923.88	Amine salt	N-H
14	3392.55	Aliphatic primary amine	N-H

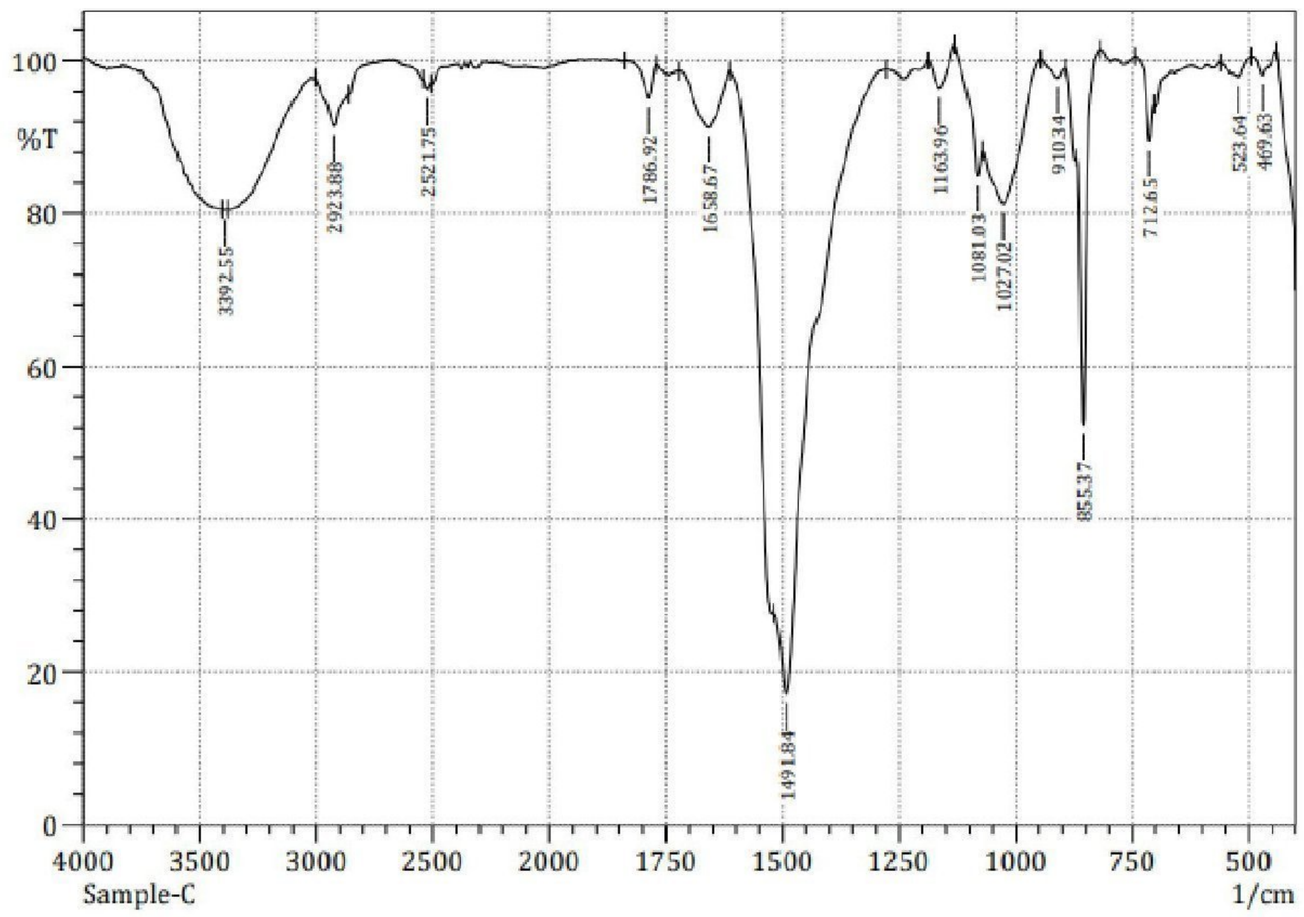
Table 14 : FT-IR Spectroscopy analysis of Turbinaria ornata

S.No	Peak value	Bond	Functional group
1	469.63	Alkyl halides	C-Br
2	624.89	Halo compound	C-Br
3	707.83	Alkene	C=C
4	821.62	Alkene	C=C
5	902.62	Aldehyde	C-H
6	1032.81	Sulfoxide	S=O
7	1054.99	Sulfoxide	S=O
8	1166.85	Carboxylic acid	O-H
9	1232.43	Alkyl aryl ether	C-O
10	1423.37	Carboxylic acid	O-H
11	1511.12	Nitro compound	N-O
12	1654.81	Alkene	C=C
13	2311.53	Primary amine salt	N-H
14	2853.49	Amine salt	N-H
15	2922.92	Amine salt	N-H
16	3340.48	Aliphatic primary amine	N-H
17	3502.49	Alcohol	O-H

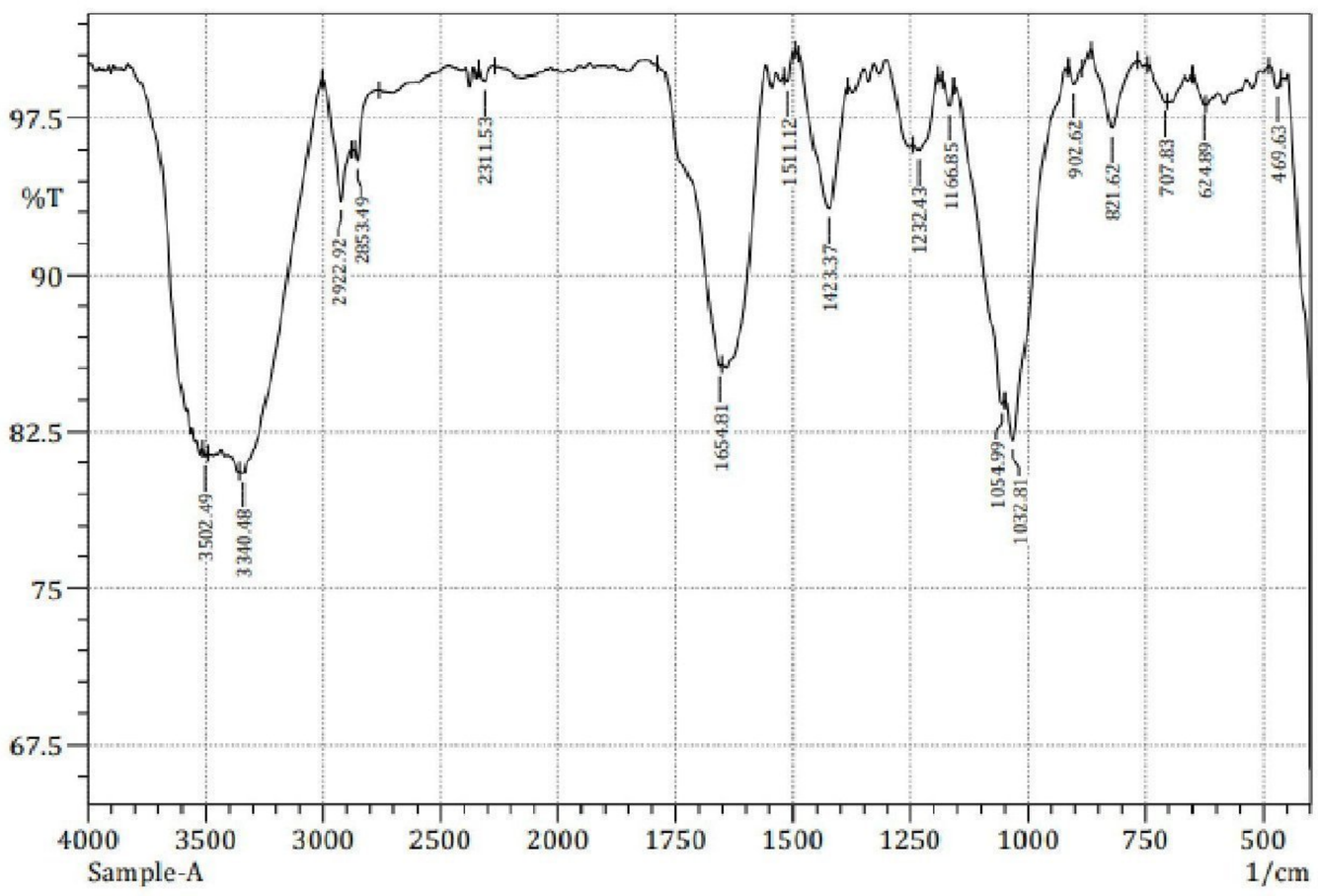
Table 15: FT-IR Spectroscopy analysis of Gracilaria corticata

S.No	Peak value	Bond	Functional group
1	467.71	Alkyl halides	C-Br
2	525.57	Alkyl halides	C-Br
3	603.68	Alkyl halides	C-Br
4	657.68	Alkyl halides	C-Br
5	712.65	Alkane	C=C
6	772.44	Alkyl halides	C-Cl
7	857.3	Aromatic nitro group	C-N
8	929.63	Carboxylic acids	O-H
9	1026.06	Amines	C-N
10	1104.17	Amine	C-N
11	1154.32	Tertiary alcohol	C-O
12	1194.82	Tertiary alcohol	C-O
13	1259.43	Alkyl aryl ether	C-O
14	1340.43	Aromatic Amine	C-N
15	1474.48	Carboxylic acid	O-H
16	1658.67	Aromatic compound	C-H
17	1869.86	Aromatic compound	C-H
18	2136.02	Isothiocyanate	N=C=S
19	2313.46	Primary amine salts	N-H
20	2519.82	Carboxylic acid	O-H
21	2921.96	Carboxylic acid	O-H
22	3325.05	Alcohols	O-H

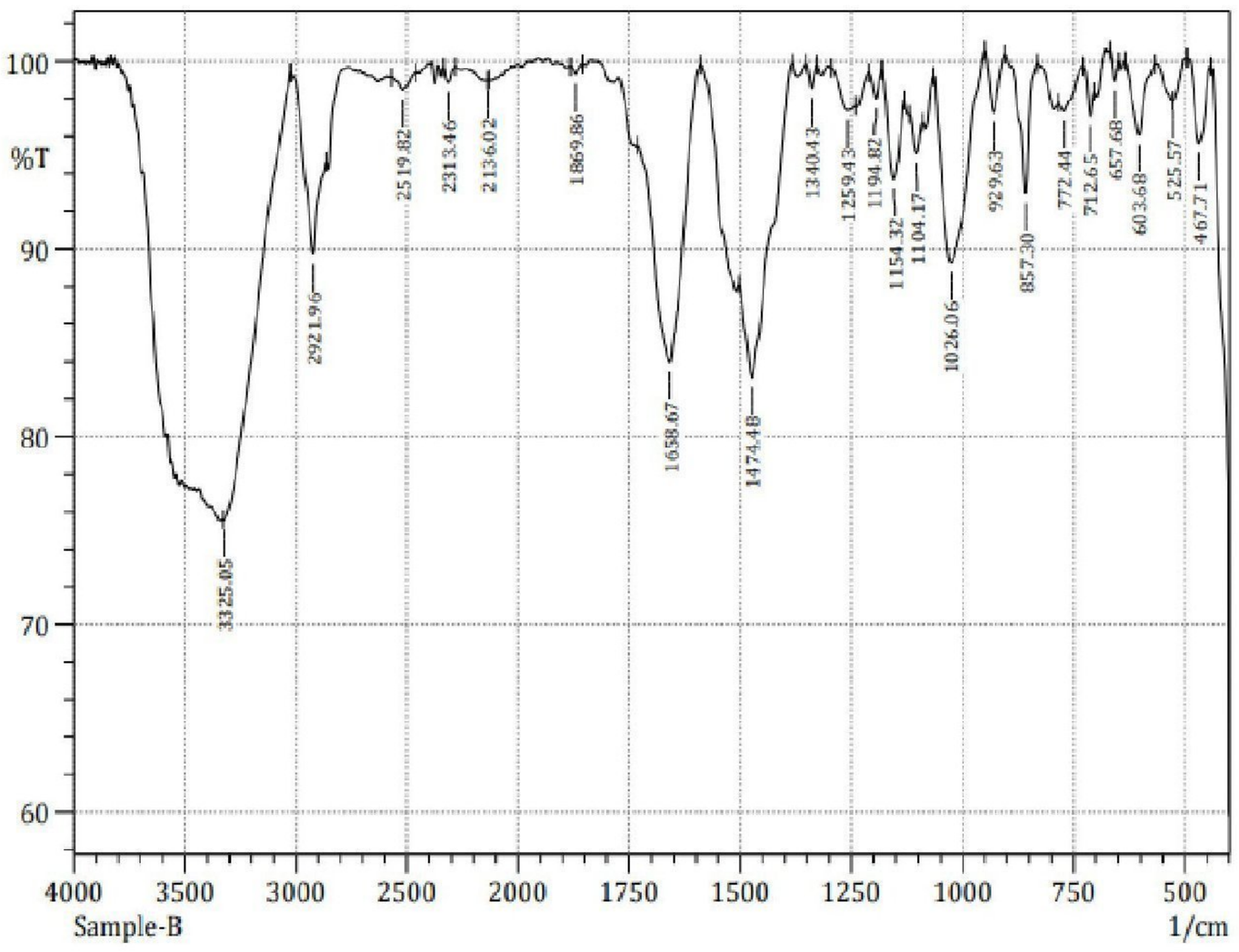
FT-IR SPECTRUM OF SAMPLE HALIMEDA TUNA



FT-IR SPECTRUM OF SAMPLE TURBINARIA ORNATA



FT-IR SPECTRUM OF SAMPLE GRACILARIA CORTICATA



From the spectral data, presence of C-Br, C-CL, C-N, S-O, C-O, C-F, H-C-H, C=C, C=O, O-H and N-H were identified. These bonding are responsible for the presence of alkyl halides, aliphatic nitro group, sulfonic group, primary alcohol, poly fluorinated compound, alkane, alkene, acide halide, carboxylic acid, amine salt and aliphatic primary amine in the seaweed of Halimeda tuna

From the spectral data, presence of C-Br, C=C, C-H, S=O, O-H, C-O and N-H were identified. These bonding are responsible for the presence of alkyl halide, halocompounds, alkene, aldehyde, sulfoxide, carboxylic acid, alkyl aryl ether, nitro compound, primary amine salt, aliphatic primary amine and alcohol in the seaweed of Turbinaria ornata.

From the spectral data, presence of C-Br, C=C, C-CL, O-H, C-H, C-O, C-N, C-C, N=C=S and N-H were identified. These bondings are responsible for the presence of alkyl halides, alkane, aromatic nitro compounds, carboxylic acids, amines, tertiary alcohol, alkyl aryl ether, aromatic amine, carboxylic acids, aromatic compound, isothiocyanate, primary amine salts and alcohols in the seaweed of Gracilaria corticata.

Fourier transform infrared spectroscopy helps analysis the functional groups. The spectral data of FTIR revealed the presence of aliphatic constituents containing alkanes, ketones, alkyl halides, hydroxyl groups etc. (Agardh, 2020). The main functional groups involved in the seaweeds uptake are Carboxyl, Sulfydryl and Hydroxyl which are the prime constituents of the seaweeds. The estimation of trace elements in the seaweeds is of great value so that the seaweeds can be treated with necessary nutrient materials in order to ensure proper growth, yields and make them resistant to disease and toxicity. (Kannan,2014)

SUMMARY AND CONCLUSION

SUMMARY AND CONCLUSION

Sea vegetables, which are commonly referred to as marine algae or seaweeds, have been a stable food since ancient times in countries located by the sea, viz., U.K, Ireland, Norway, the Pacific Islands, African countries and American countries. In modern days, they have become primarily associated with Asian cuisine, Japan which has the world's largest seaweed consumption per capita with 10-15%, is also accorded with significantly lower rate of cancer, thyroid diseases, heart diseases and dementia (Fitz Gerald et al., 2011). Today China, Japan and the republic of Korea are the largest consumer of seaweeds as food. Seaweeds are considered to be the food supplement for 21st century and as source for proteins, lipids, polysaccharides, minerals, vitamins and enzymes. In India, seaweeds are generally being used as raw material for the production of agar, alginates and seaweed liquid fertilizer, in spite of their great potential as therapeutic health booster, beauty enhancer and the source of nutrition. Hence it becomes essential to popularise seaweeds as health food which will help to feed undernourished people in India. For this purpose it requires a thorough study on the occurrence, availability and diversity of seaweeds both locally and regionally, their neutraceutical and therapeutical value which provide a baseline data for further exploration. So in the present study an attempt has been made picturise the diversity and availability status of seaweeds, found along the coast of Manapad, Gulf of Mannar, Thoothukudi.

Seaweeds were surveyed during four months viz., December, January, February and March at unique and distinct stations during low- tide in the year 2022-2023 by random sampling method. The study indicated that, Manapad was endowed with numerous taxa belonging to Chlorophycea (9 species), Phaeophyceae (12 species) and Rhodophyceae (9 species). Nine taxa were found as most abundant in this area. In Manapad region the post monsoon season (February to May) was noted for proliferous growth of different members of Chlorophyceae, Phaeophyceae and Rhodophyceae. The supply of nutrients from Manapad after the short rainy season, and the associated climatic conditions would probably favoured the growth and establishment of these seaweed species during post- monsoon season. Distributional diversity and abundance of species were high in the coast of Manapad. So this area is suitable for seaweed cultivation and harvest.

Phytochemical constituents in seaweed extracts contain biologically active compounds of antioxidant, antibacterial and anti inflammatory properties. Among them alkaloids, flavonoids, phenols, carbohydrates, terpenoids, steroids, glycosides and coumarins were present in Turbinaria ornata and Gracilaria corticata.

The study revealed that the seaweed extracts of Halimeda tuna was showing maximum presence of alkaloids, flavonoids, phenols, quinines, carbohydrates, terpenoids and steroids. Saponins, tannins, glycosides, proteins and coumarins were completely absent in all extracts of Halimeda tuna. Phenols, steroids and coumarins were found in all extracts of Turbinaria ornata. Quinines, saponins, tannins and proteins are completely absent in Gracilaria corticata. Flavonoids and Glycosides were highly present in ethanol and aqueous extracts of Turbinaria ornata. Terpenoids were highly present in acetone, hexane and aqueous extracts of Turbinaria ornata.

Phytochemistry takes into account the structural compositions of these metabolites, the biosynthetic pathways, functions, mechanisms of actions in the living systems and it's medicinal, industrial, and commercial applications.

Ethanolic extract of Halimeda tuna had the highest activity against E.coli (21mm). Acetone extract of Turbinaria ornata exhibited maximum activity against, E.coli (19mm). Similarly ethanol (18mm), methanol (17mm) and hexane (5mm) extracts of E.coli showed the inhibition zone. Ethanolic extract of Gracilaria corticata interacted with E.coli and resulted into growth inhibition (20mm) cloned to streptomycin. Similarly E.coli was arrested by G.corticata in methanol extracts (12mm).

Ethanol, methanol and acetone extracts of Halimeda tuna , Turbinaria ornata and Gracilaria corticata shows more potent than hexane and aqueous extracts. The hexane extract of Halimeda tuna at 10 mg/ml concentration shows paralysis at 15 minutes and death at 25 minutes. The hexane extract of Turbinaria ornata at 10 mg/ml concentration was taken at 19 minutes for paralysis and death at 28 minutes. The hexane extract of Gracilaria corticata caused paralysis at 15 minutes and time of death at 25 minutes.

From the spectral data, presence of C-Br, C-CL, C-N, S-O, C-O, C-F, H-C-H, C=C, C=O, O-H and N-H were identified. These bonding are responsible for the presence of alkyl

halides, aliphatic nitro group, sulfonic group, primary alcohol, poly fluorinated compound, alkane, alkene, acide halide, carboxylic acid, amine salt and aliphatic primary amine in the seaweed of Halimeda tuna

From the spectral data, presence of C-Br, C=C, C-H, S=O, O-H, C-O and N-H were identified. These bonding are responsible for the presence of alkyl halide, halocompounds, alkene, aldehyde, sulfoxide, carboxylic acid, alkyl aryl ether, nitro compound, primary amine salt, aliphatic primary amine and alcohol in the seaweed of Turbinaria ornata.

From the spectral data, presence of C-Br, C=C, C-CL, O-H, C-H, C-O, C-N, C-C, N=C=S and N-H were identified. These bondings are responsible for the presence of alkyl halides, alkane, aromatic nitro compounds, carboxylic acids, amines, tertiary alcohol, alkyl aryl ether, aromatic amine, carboxylic acids, aromatic compound, isothiocyanate, primary amine salts and alcohols in the seaweed of Gracilaria corticata.

It is advocated that Halimeda tuna, Turbinaria ornata and Gracilaria corticata can be sustainably utilized for the extraction and purification of bioactive compounds which would open a new door in the field of drug development.

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EVALUATION OF PHYTOCHEMICAL , ANTIBACTERIAL ACTIVITY AND SYNTHESIS OF NANOPARTICLES OF *ACHYRANTHES ASPERA* (L.) AND *ALTERNANTHERA SESSILIS* (L.) R.Br.

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affiliated to **MANONMANIAM SUNDARANAR UNIVERSITY**

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Master of Science in Botany.

By

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2022-2023

CERTIFICATE

This is to certify that this dissertation entitled, **Evaluation of phytochemical, antibacterial activity and synthesis of sliver nanoparticles of *Achyranthes aspera* (L.) and *Alternanthera sessilis* (L.) R.Br. submitted by S.SNOWBA SERAPHINE Reg.No. 21APBO15,** to ST. MARY'S COLLEGE (Autonomous), THOOTHUKUDI in partial fulfilment for the award of the degree of " **Master of Science in Botany"** is done by her under my supervision. It is further certified that this dissertation or any part of this has not been submitted elsewhere for any other degree.

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DECLARATION

I do here by that this entitled **Evaluation of phytochemical, antibacterial activity and synthesis of sliver nanoparticles of *Achyranthes aspera* (L.) and *Alternanthera sessilis* (L.) R.Br.** By me in partial fulfillment for the award of the degree of **Master of Science in Botany,** is the result of my original and independent work carried out under the guidance of **Dr.A. Jacintha Tamil Malar,** Assistant Professor of Botany, St.Mary's College (Autonomous), Thoothukudi and it has been submitted for the award of any other degree.

Station: Thoothukudi

S. Snowba Seraphine

Date: **(S.SNOWBA SERAPHINE)**

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CONTENT

INTRODUCTION

INTRODUCTION

Nature has been a source of medicinal agents for thousands of years and since the beginning of mankind. Medicinal plant is an integral part of human life to combat the sufferings from the dawn of civilization. It is estimated that more than 80,000 of total plant species have been identified and used as medicinal plants around the world. Over the past twenty years, interest in medicinal plants has grown enormously from the use of herbal products as natural cosmetics and for self-medication by the general public to the scientific investigations of plants for their biological effects in human beings. Therefore, people are encouraging indigenous production and processing of these medicinal plants to use in different cultures and religion for the treatment of various diseases. Nowadays, there is a revival of interest with herbal-based medicine due to the increasing realization of the health hazards associated with the indiscriminate use of modern medicine and the herbal drug industries is now very fast-growing sector in the international market. There is great demand for herbal medicine in the developed as well as developing countries like India, because of their wide biological activities, higher safety of margin than the synthetic drugs and lesser costs.

Ever since ancient times, in search for rescue for their disease, the people looked for drugs in nature. The beginnings of the medicinal plants' use were instinctive, as is the case with animals. In view of the fact that at the time there was not sufficient information either concerning the reasons for the illnesses or concerning which plant and how it could be utilized as a cure, everything was based on experience. In time, the reasons for the usage of specific medicinal plants for treatment of certain diseases were being discovered; thus, the medicinal plants' usage gradually abandoned the empiric framework and became founded on explicatory facts. Until the advent of iatrochemistry in 16th century, plants had been the source of treatment and prophylaxis. Nonetheless, the decreasing efficacy of synthetic drugs

and the increasing contraindications of their usage make the usage of natural drugs topical again.

Nature has been a source of food and medicine since ancient times, especially wild plants that are edible and have therapeutic properties due to the presence of different nutrients and active phytochemicals. Globally, people rely on wild edible plants, including weeds to supplement their staple food, compensate food shortage, and as medicine, particularly those in rural areas and developing countries who are living in poverty (Ulian *et al.*, 2020).

Knowledge of herbs has been handed down from generation to generation for thousands of years. Herbal drugs constitute a major part in all traditional systems of medicines. Herbal medicine is a triumph of popular therapeutic diversity. Plants above all other agents have been used for medicine from time immemorial because they have fitted the immediate personal need, are easily accessible and inexpensive. In the recent past there has been a tremendous increase in the use of plant based health products in developing as well as developed countries resulting in an exponential growth of herbal products globally. An upward trend has been observed in the research on herbals. Herbal medicines have a strong traditional or conceptual base and the potential to be useful as drugs in terms of safety and effectiveness leads for treating different diseases.

World Health Organization has made an attempt to identify all medicinal plants used globally and listed more than 20,000 species. According to the WHO more than 80 % of the world's population relies on traditional herbal medicine for their primary health care. Plants continue to serve as possible sources for new drugs and chemicals derived from various parts of plants. In recent time there has been a marked shift towards herbal cures because of the pronounced cumulative and irreversible reactions of modern drugs. However, due to over population, urbanization and continuous exploitation of these herbal reserves, the natural resources along with their related traditional knowledge are depleting day by day. In the

present era of drug development and discovery of newer drug molecules many plant products are evaluated on the basis of their traditional uses.

There exists a plethora of knowledge and information and benefits of herbal drugs in our ancient literature of Ayurvedic and Unani medicine. One of the earliest treatises of Indian medicine, the Charaka Samhita (1000 B.C.) mentions the use of over 2000 herbs for medicinal purpose. According to the WHO survey 80% of the populations living in the developing countries rely almost exclusively on traditional medicine for their primary health care needs. Exploration of the chemical constituents of the plants and pharmacological screening may provide us the basis for developing the leads for development of novel agents. In addition, herbs have provided us some of the very important life saving drugs used in the armamentarium of modern medicine.

Plants are rich in a wide variety of secondary metabolite, such as tannins, terpenoids, alkaloids and flavonoids, which have been found in vitro to have antimicrobial properties. Among the various medicinal and culinary plants, some indigenous species contain a wide variety of free radical scavenging molecules, such as flavonoids, anthocyanins, cartenoids, dietary glutathionine, alkaloids, tannins, saponins, steroids, terpenoids, and rotenoids which are in antioxidant activities (Pham-HuyLA and Pham-Huy, 2008). Therefore, the intake of natural antioxidants from plants has been associated with low incidence of cancer, cardiovascular diseases associated with aging (Kho *et al.*, 1999).Considering the potential role in food industry and human health, antioxidants are gaining popularity all across the globe.

With the novel approach of nanotechnology, we have witnessed tremendous changes with the emergence of nanoparticles with unique functions and size dependent physiochemical properties (Arya *et al.*, 2019). Research on nanomaterials mainly emphasizes the synthesis of nanoparticles of various size, shape and structure for desired applications.

Nanoparticles shows varying characteristics including differences in shape, size and also higher area than that of their bulk material and are used for wide range of applications (Schexnailder and Schmidt, 2009). The synthesis of nanoparticles are classified mainly into three categories mainly physical, chemical and biological method. For the physical method of synthesis of nanoparticles temperature, pressure, and energy will be maintained. Chemical methods of nanoparticles synthesis are carried out by methods such as laser pyrolysis, atomic or molecular condensation, sol-gel processes, chemical etching, sputtering, spray pyrolysis (Valodkar *et al.*, 2011). The shape, size parameters of nanoparticles are often altered with different concentrations of chemicals and reaction conditions. When the synthesized nanoparticles are subject to a specific application they will face challenges of bioaccumulation, toxic nature, modeling factors, regeneration, reuse and recycle (Albanese *et al.*, 2012).

Carbon-based nanoparticles like fullerenes, carbon nanofibers, carbon nanotubes, lampblack graphene have a different application. Inorganic nanoparticles include metals like gold, silver and metal oxides like titanium dioxide, iron oxide, Organic nanoparticles include dendrimers, micelles, liposomes and polymer nanoparticles are generally focused on targeted drug delivery systems and so on; Composite based nanoparticles include combination of carbon based, metal based or organic based nanoparticles with any sort of metal, ceramic, or polymer bulk materials also are widely used in various sectors (Jeevanandam *et al.*, 2018). Among these, inorganic nanoparticles are currently explored worldwide in drug delivery systems, biomedical devices, cosmetics, electronics and energy sector(Kumaresan *et al.*, 2018).

Achyranthus is a genus of medicinal and ornamental plant in the amaranth family, Amaranthaceae, chaff flower is a common name for plant in this genus. It include 20 species *Achyranthus aspera* is distributed throughout the tropical world. It can be found, in many

places growing as an introduced species and a common weed. However, among the estimated 250,000-400,000 plant species, only 6% have been studied for biological activity, and about 15% have been investigated phytochemically. This shows a need for planned activity guided phyto-pharmacological evaluation of herbal drugs.

Alternanthera is a genus of flowering plants in the family Amanthaceae. It is a widespread genus with most species occurring in the tropical America and Asia plants of the genus may be known generally joy weed. Several species are notorious noxious weeds. *Alternanthera sessilis* is a flowering plant known by several common names, including sessile joyweed, Brazilian spinach, dwarf copperleaf. It is as cultivated as a vegetable worldwide.

Based on these background information, the present study was justifiably designed with the following objectives:

- To carry out the characterization of leaves of *Achyranthes aspera* and *Alternanthera sessilis*
- To evaluate the antibacterial efficiency of different extracts of leaves of selected plants against some important human pathogenic organisms.
- To explore the FTIR bioactive compounds by spectroscopy.
- To synthesize of sliver nanoparticles

REVIEW OF LITERATURE

LITERATURE REVIEW

Plants are source of many potent and strong drugs that are used medicinally in various countries (Srivastva *et al.*, 2008). In various traditional literature, a variety of herbs with significant antimicrobial activity have been published (Jones *et al.*, 1996). Ayurvedic traditions use a variety of medicinal plants on a regular basis. More than 7000 medicinal plants have been identified in medicine meets the primary health care needs of more than 80% of the world's population (Umamaheswari *et al.*, 2008). Because of the lack and high cost of new generation antibiotics, researchers are turning to alternative medicines for anti microbial action (Poovendren *et al.*, 2011).

DISRRIBUTION AND MEDICINAL VALUES OF PLANTS

Goyal *et al.*, 2007 reported that *Achyranthes aspera* is used as an emmenagogue, antiarthritic, purgative, diuretic, antimalarial, oestrogenic, antispasmodic , cardiotonic, antibacterial and antiviral agent.

Veena and Chauduary 2015 reported that antioxidants like alkaloids, terpenoids, saponins etc of various pharmacological properties are present in *A.aspera* many chemical constituents have also been isolated from this plant by various techniques.

Leaves of *Achyranthes aspera* are used to treat a variety of illness, including renadropsy, haemorrhoids, dysentery, promoting labour pain, nose bleeding, snake bites and dilating blood vessels. Alkaloids, saponin, tannins, flavanoids, glycosides, steroids, essential oils and examples of secondary metabolites that have been shown to have higher bioactivity against a number of aliments. The presence of secondary metabolite in particular such as achyranthine, ecdysterone, oleanolic acid, spinasterol, apigenin, achyrantheric acid, ursolic effects .(Senthil kumar raju *et al.*, 2022).

The seeds and leaves of *Achyranthes aspera* can reduce antibiotic resistance and boost the effectiveness of existing medications. Several secondary metabolites found in

Achyranthes aspera leaves are essential for combating resistance. Methicillin-resistant *Staphylococcus aureus, Enterococcus faecalis, Acinetobacter baumannii, Klebsiella pneumoniae* and *Psedomonas aeruginosa* were different bacterial strains that were treated with seven different antibiotics of seven medications Cefoxitin, Pencillin and Co-trimoxazole were resistant against Methicillin resistant *Staphylococcus aureus*. When being combine with plant extracts the zone of inhibition for all three of these antibiotics shifts from the resistant to the susceptible range. Following treatment with the plant extracts, the antibiotics Ciprofloxacin, Levofloxacin, Penicillin, Amoxicillin, Imipenem and Vancomycin proved ineffective against *Enterococcus faecalis* (Hamna *et al.,* 2022).

Varma 2016 reported that the herb is highly used by traditional healers in treatment of cold, cough, asthma, boils, bronchitis, headache, colic, debility, dropsy, bleeding, dysentery, ear problems, leucoderma, pneumonia, kidney issues, snake and scorpion bites and skin conditions.

Ganesh 2021 reported that *Achyranthes aspera* are used for the treatment of various diseases because of their safety and effectiveness. Though almost all of its parts are used in traditional systems of medicines, seeds, roots and shoots are the most important parts which are used medicinally. The major chemical constituents are carbohydrates, protein, glycosides, alkaloids, tannins, saponins, flavonoids, lignin etc.

Rehman *et al.,* 2018 tested the composition of *Achyranthes aspera* and reported that this plant is known to have high caloric and nutritional value owing to the presence of vitamin c, minerals, sodium, calcium, magnesium, potassium chloride and phosphorous. Essential oil extracted from leaves of this plant has number of important chemical constituents that possesses strong prophylactic potentials and anti-cancer, anti-microbial, anti-diabetic, cardio-protective, immune-modulatory and prothyrodic activity used in several medicinal formulations to treat severe illnesses. Plant produce a great number of

secondary metabolites, many of them with antibacterial and antifungal activity. Well known examples of these compounds include flavonoids, phenols and phenolic glycosides, unsaturated lactones, sulphur compounds, saponins, cyanogenic glucosides and glucosinolates (Gomez *et al.*, 1990).

Achyranthes aspera Linn. is a well-known plant drug in Ayurvedic, Unani-Tibbi, Siddha, Allopathic, Homeopathic, Naturopathic & Home Remedies. The pharmacognostic evaluation on the different parts of *Achyranthes aspera* Linn. (Amaranthaceae) revealed the presence protein, glycosides, alkaloids, tannins and phenolic compound, steroid reducing sugars and saponin, glycosides. These observations will help in the Pharmacognostical identification and standardization of the drug in the crude form and also to distinguish the drug from its adulteration (Dhale and Sonal 2013).

Neupane *et al.,* 2022 tested the quantitative phytochemical profiles and determined the free radical scavenging activity of phytochemical constituents of the entire plant of *Achyranthes aspera* Linn and revealed the presence of flavonoids, alkaloids, terpenoids, phytosterols, tannins, saponins, phenolic compounds, and carbohydrates. Total phenolic content, total flavonoids content, and antioxidant activity of the extract were found as 209.007 µg GA/mg, 17.59 µg QE/mg, and 25.12% (100 µg/mL), respectively.

Kota *et al.,* 2017 reported that *Alternanthera sessilis* widely consumed leafy vegetable in different parts of India, and investigated its phytochemical composition, antioxidant, antibacterial and anti-cataract activities using standard procedures. Antioxidant assays were done by *in vitro* methods such as 1,1-diphenyl −2- picrylhydrazyl (DPPH) and reducing power assays. Total phenolic content was determined by Folin-Ciocalteu method. Antibacterial tests were performed by disc/agar well diffusion methods using laboratory and clinical human isolates of chosen micro-organisms. Anti-cataract activities were determined by lipid peroxidation and Na^+- K^+ ATPase assay. Experimental results indicated that all

the organic solvent extracts contained flavonoids, terpenoids, phenols, phytosterols and alkaloids. Highest total phenolic content was found in ethyl acetate extract (67.75 µg GAE/mg) followed by methanolic (44 µg GAE/mg), Chloroform (12.13 µg GAE/mg) and petroleum ether (0.013 µg GAE/mg) extracts.

Almutairi 2015 analysed the phytochemical profile of Amaranthaceae plants comprises essential oils, betalains, phenolic compounds and terpenoids.

The increasing prevalence of multidrug resistant strains of bacteria and the recent appearance of strains with reduced subceptibility to antibiotics raises the specter of untreatable bacterial infections and adds urgency to the search for new infection-fighting strategies (Sieradski *et al.*,1999).

Dhale and Sonal 2013 tested antibacterial activities of *Achyranthes aspera* and *Cassia alata* against *Eschercia coli*, *Bacillus subtilis*, *Vibrio cholerae*, *Salmonella typhi* and *Staphylococus aureus*. While neither the leaf nor stem parts of *A. aspera* in any organic extractions showed antibacterial activity, the methanolic extracts of both the leaf and stem parts of *C. alata* exhibited antibacterial activity, but only to *B. subtilis* and *S. typhi*, and the corresponding MIC values of the leaf extracts were estimated as 1.25 and 1.5 mg/ml respectively. However, the ethanolic extracts of both the stem and leaf parts were found equally effective only to *S. aureus* (MIC= 1.25 mg/ml).

Londonkar *et al.,* 2011 recognized antibacterial and antifungal activities the petroleum ether, Chloroform and Methanol extract of dried leaves of *Achyranthes aspera* against 5 different species of human pathogenic bacteria and 17 fungal strains by the agar-solid diffusion method. Most of the extracts were devoid of antifungal and antibacterial activities, except the methanolic ectracts of leaves of *Achyranthes aspera* obtained by infusion, which has showed a strong inhibitory activity against the Gram-positive bacteria *Staphlyococcus aureus* with a minimal inhibitory concentration (MIC) of 5000 µ l ml-1. The minimal

inhibitory concentration values to dermatophyte strains were 2500 µ l ml-1 against *Trichophyton rubrum* (LM-09, LM-13) and *Microsporum canis*. In conclusion, it appears that *Achyranthes aspera* has non-specific antimicrobial activity

Mohinder Kaur *et al.,* 2005 tested the anti bacterial activity tested of root and shoot extracts of *A. aspera*. While petrolium Ether (60-80°) root extract showed the activity against *B. Substilis*. Only antifungal activity of roots was found in extracts with pet ether, chloroform and methanol against *fusarium* sp. only. Methanol and Aqueous shoot extracts were weakly active against Pencillium.

Yadav *et al.,* 2016 assessed antibacterial activity of the *A. aspera* extract against *Streptococcus mutans*. *A. aspera* extract showed statistically significant zones of inhibition. *A. aspera* showed marked antibacterial activity against *S. mutans*

Khan *et al.,* 2009 evaluated the anti bacterial activity of *Achyranthes aspera*. using disc diffusion method with some Gram-positive and Gram negative bacteria species. The ethanol extraction displayed the highest antibacterial activity against bacteria. The ethanol extract of *A.aspera* is much effective than the petroleum ether.

Kleinowski *et al.,* 2016 evaluated the allelopathic effects and verified the antibacterial properties of extracts from *A. philoxeroides*.

Akbar *et al.,* 2021 investigated the antibacterial activity of alligator weed (*Alternanthera philoxeroides*) organic extracts against three bacterial phytopathogens (*Erwinia carotovora, Ralstonia solanacearum and Xanthomonas axonopodis*). The n-hexane extract of *A. philoxeroides* leaves showed the maximum inhibition zone diameter (IZD)= 28.1 mm against R. solanacearum, while, the corresponding value for the positive control (Penicillin) was 48 mm IZD. There was no antibacterial activity of negative control, dimethyl sulfoxide (DMSO).

Kumar *et al.*, 2021 evaluated the antimicrobial activity ethanol extract of *Alternanthera sessilis* Linn. against medically important gram-positive bacteria such as *Staphylococcus aureus*, *Bacillus subtilis*, *Bacillus polymexia* & *Streotococcus faecalis*, gram negative bacteria such as *Pseudomonas aerugenosa*, *Salmonella typhii*, *Shigella dysenteriae* & *Escherichia coli* and fungi like *Penicillum notatum*, *Aspergillus niger* & *Candida albicans*. antimicrobial activity of the extract against the tested strains of microorganisms between concentration range of 50 and 400 µg/ml. The results of zone of inhibition study revealed concentration dependant nature of the extract with better effectiveness against gram-positive bacteria than gram-negative bacteria.

The SNPs synthesized from bark extracts of *Bowellia ovalifolia* and Shorea *tumbuggaia* showed toxic towards *Klebsiella* and *Aspergillus*; and *Pseudomonas* and *Fusarium* species respectively. Where the growth of *Pseudomonas* and *Rhizopous* species were inhibited maximum by the SNPs synthesized from the leaf extract of *Svensonia hyderobadensis*, the results indicate that the silver nanoparticles may have an important advantage over conventional antibiotics (Savithramma *et al.*, 2011).

Fourier transform infrared spectroscopy (FTIR) is a fast and nondestructive analytical method. Associated with chemometrics, it is a powerful tool for research and industry. Bunaciu *et al.*, (2012). FTIR spectroscopy is proved to be a sophisticated instrument to analyse the components of the plant cells. The cellular constituents in the leaves and stem of these plants *Mimosa pudica* and *Caesalpinia pulcherrima* were monitored for the qualities of medicinal applications. (Mitra and .Basker 2011).

Kalaichelvi and Dhivya 2017 determined the Fourier Transform Infrared Spectral analysis of *Micrococca mercurialis*. The aqueous and organic solvent extracts (petroleum ether, acetone, chloroform, ethanol and aqueous) from the whole plant of *Micrococca mercurialis* (Euphorbiaceae) were tested for the availability of alkaloids, glycosides,

flavonoids, phenols, saponins, steroids, amino acids, tannins, terpenoids, quinones, anthraquinones and coumarin. The UVVIS spectrum showed the peaks at 214, 446 and 472 nm with the absorption of 0.599, 0.655, and 0.550 respectively. The FT-IR spectrum showed the presence of alcohols, phenols, alkanes, alkynes, alkyl halides, aldehydes, aromatics, nitro compounds and amines.

Quershi and Solanki 2022 to identifyed the functional groups present in the crude sample of powdered *A.aspera* leaves. The FT-IR results showed the presence of 11 peaks, which confirmed the presence of essential phytochemicals present in the crude sample of powdered *A.aspera* leaves.

Nanotechnology is now creating a growing sense of excitement in the life sciences especially biomedical devices and Biotechnology (Prabhu *et al., (*2010).

Sliver nanoparticles have found tremendous applications in the field of high sensitivity biomolecular detection and diagnosis, antimicrobials and therapeutics catalysis and microelectronics (Geethalakshimi and Sarada 2010).

Use of plants for the synthesis of nanoparticles does not require high energy, temperatures, and it is easily scaled up for large scale synthesis and it is cost effective too (Mukunthan *et al.,* 2011).

MATERIALS AND METHODS

MATERIALS AND METHODS

Collection and identification of plant material source of plant material:

The fresh leaves of *Alternanthera sessilis* and *Achyranthes aspera* belongs to the family Amaranthaceae are collected from Lionstown and Campus of St.Mary's College, in Thoothukudi district in TamilNadu, during December 2022 to January 2023. After collection the leaves are removed from the branches, sorted out to separate the bad ones from the good ones, washed properly with sterile water. The leaves were dried under shade and coarsely powdered.

Prepartion of extracts:

2.5 grams powdered sample was sequentially extracted with 50 ml of benzene, acetone, ethanol, chloroform and aqueous solution soaked with 24 hours and filtered through the cheese cloth. The filtrate stored in airtight bottles. The prepared extracts were tested for phytochemical screening, anti bacterial activity.

Preparation of leaf flour sample:

Achyranthes aspera and *Alternathera sessilis* leaves are processed using home based processing techniques i.e., shade drying (Iijarotimi *et al.*, 2013).

Raw *Achyranthes aspera* and *Alternanthera Sessilis* leaves flour:

Collected fresh leaves of *Achyranthes aspera* and *Alternanthera sessilis* leaves are spread on a tray and shade dried at room temperature for a period of 15-20 days for proper drying. The dried leaves are homogenized to fine powder in a mechanical pulverizer and stored in airtight bottles.

Phytochemical qualitative analysis:

The phytochemical tests were done for analysising different chemical groups present in the extracts. These were done to find out the presence of bioactive chemical constituents

such as alkaloid, flavonoids, tannins, phenol, terpenoids, glycoside, cardiac glycosides, anthroquinone, steroids and saponins. Detection of active phytochemical constituents was carried out for all the extracts using the standard procedures (Kokatte, 200, Harborne, 1984).

Test for alkaloids:

Mayer's Test:

3 ml of extracts was added to 1% HCL and then allowed to steam bath. Few drops of Mayer's reagent was added to the mixture, Turbidity indicates the presence of alkaloids.

Test for flavonoids:

Lead acetate Test:

To 1 ml of extract, 1ml of 10% lead acetate was added. Formation of yellow precipitate showed the presence of flavonoids.

Test for Tannins:

Ferric chloride Test:

To 1ml of extract, 1 ml of distilled water was taken and stirred, few drops of Ferric chloride solution were added to the mixture bluish green colour precipitate showed the presence of tannins.

Detection of Phenols:

$FeCl_3$ Test:

About 2ml of plant extract was taken and warmed at 45-50°c. Then 2ml of 0.3% $FeCl_3$ was added. Formation of green or blue colour indicated the presence of phenols.

Test for Terpenoids:

Salkowski Test:

About 2ml of chloroform was added to 1ml of the extract. Then 3ml of concentrated H_2SO_4 carefully added to form a layer. A reddish brown coloration of the interface indicated the presence of terpenoids.

Test for Glycosides:

2ml of extract was dissolved in chloroform and 2ml of acetic acid was added to the mixture. The solutions were cooled and then add few drops of sulphuric acid. A colour change from blue to green indicated the presence of glycosides.

Test for Cardiac glycosides:

Keller- Killiani Test:

1ml of extract was dissolved in 5ml of water 2ml of glacial acetic acid containing one drops of ferric chloride solution was added. This was under layer with 1ml of concentrated sulphuric acid. A brown ring at the interface indicated the presence of a deoxysugar characteristics of cardenolides. A violet ring may appear below the brown ring, while in the acetic acid layer a greenish ring may from just above the brown ring and gradually spread this layer.

Test for Anthraquinone:

1ml of the extract was boiled with 10 ml of sulphuric acid and filtered while hot. The filtered was shaken with added to 5m of chloroform. The chloroform layer was pipette into another test tube followed by addition of 1ml dilute ammonia. The resulting solution was observed for colour changes to violet indicated presence of anthraquinone.

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Test for steroids:

Sal kowski Test:

To 2 ml of extract, was dissolved in chloroform, 2ml of concentrated sulphuric acid was added to the mixture. Red colour formation indicated the presence of steroids.

Test for saponins:

2 ml of extract dilute with 5 ml of distilled water and warmed. The formation of stable form indicated the presence of saponins.

ANTIBACTERIAL ACTIVITY:

Bacterial strains used:

The test organism were obtained from the Department of Botany, St. Mary's College (Autonomous), Thoothukudi. The two gram positive bacteria viz , *Bacillus subtilis* G +ve, *Staphylococcus aureus* G+ve were used in the present study. *Bacillus subtilis* is responsible for causing food borne gastroententis. *E.coli., Staphylococcus aureus* cause disease like mastitis, abortion and upper respiratory complications.

Disc diffusion assay (Bauer *et al.*, 1966)

Antibacterial activity was evaluated by agar disc diffusion method. Test solutions were prepared with known weight of different solvent extracts dissolved in 5% dimethyl sulphoxide (DMSO). What man No:1 sterile filter paper discs (5mm) were impregnated with 2o µl of these extracts and allowed to dry at room temperature. The spread plates were prepared by proper concentration of inocula. Each sample loaded disc was placed in the seeded agar plate. After 24-48 hours of 37° c incubation, the diameter of the inhibition zone was measured. For positive control, streptomycin disc (100µg / ml) was used, where as negative control, respective solvents were loaded on the sterile disc.

FT-IR (Fourier transforms infra –red spectroscopy) spectroscopic analysis

(Vijaybasker and Shiyamala, 2012)

Ten milligram of *A.aspera* and *A.sessils* leaf powder was mixed with 100 mg of dry potassium bromide (FT-IR grade) and then compressed into a pellet using hydraulic press (5000-10000 psi). The pellet was immediately put into the sample holder and FT-IR (Systronics 166) spectra were needed in the range of 400 to 4000 cm^{-1}.

SYNTHESIS OF SLIVER NANOPARTICLES:(Hamlata *et al.*, 2020)

Procedure:

For the green synthesis of sliver nanoparticles, we used the aqueous leaf extract of *A.aspera* and *A. sessilis.* For this 9 ml of leaf extract was added 1 ml of 1Mm aqueous sliver nitrate solution, followed by heating at 80° C for 1 hour constant stirring. The formation of the AgNPs was preliminarily detected by the change in color from yellow to dark brown. The final synthesized sliver nanoparticles were denoted as AgNPs , which were freeze dried and then stored at 4° until further use. It showed that aqueous sliver ions could be reduced by aqueous extract of plant parts to generate extremely stable sliver nanoparticles in water.

RESULT AND DISCUSSION

Plate 1: *Achyranthes aspera* (L.)

RESULT AND DISCUSSION

In the present investigation, the phytochemicals, antibacterial activity are evaluated and chemical components are identified by FTIR. Sliver nanoparticles are synthesized from the leaves of *Achyranthes aspera* and *Alternanthera sessilis.*

Systematic position

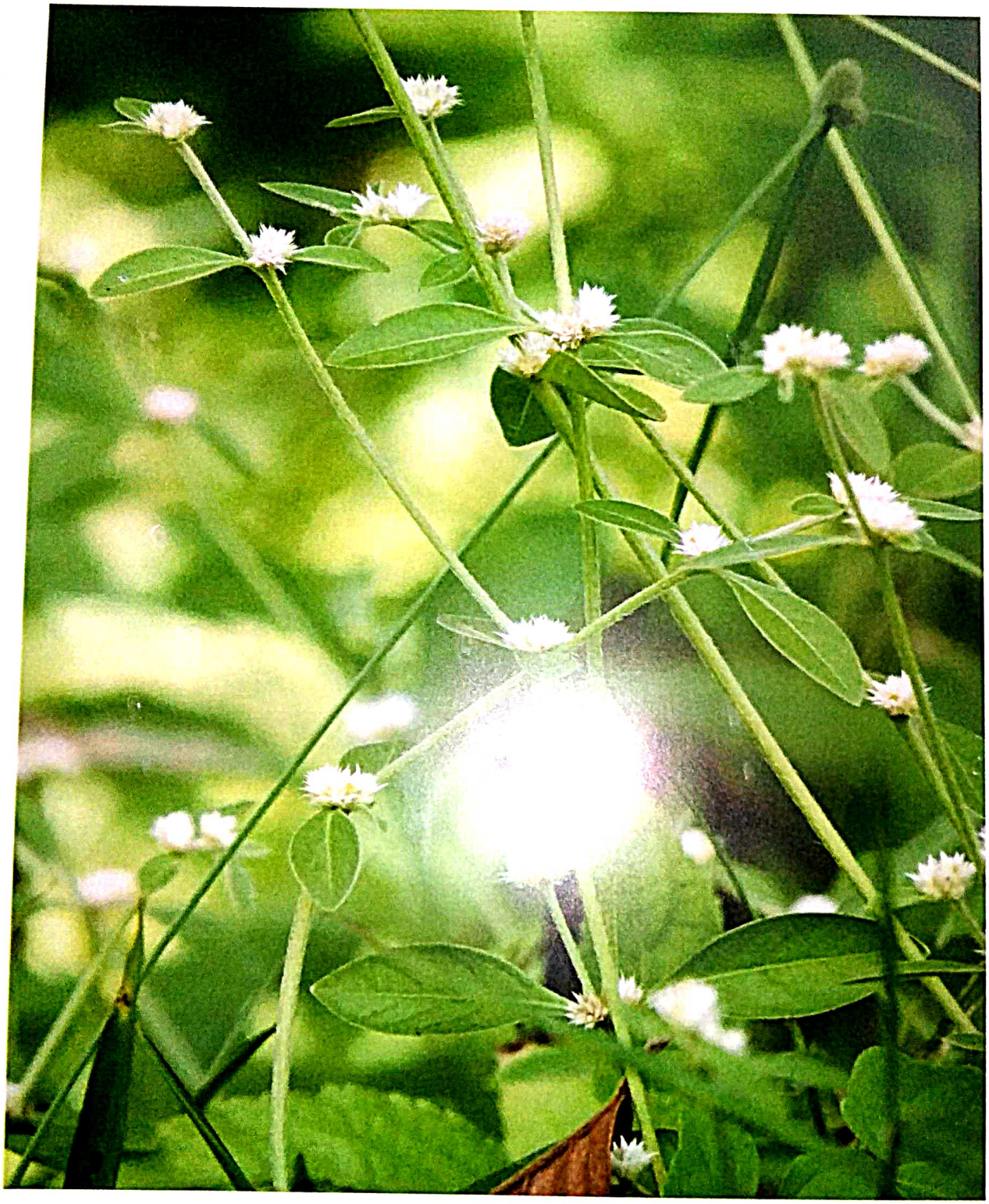
In the Bentham and Hooker system of classification, the systematic position of *Achyranthes aspera* (L.) is as follows,

Kingdom	:	Plantae
Division	:	Tracheophyta
Class	:	Dicotyledons
Sub class	:	Caryophyllidae
Order	:	Caryophyllales
Family	:	Amaranthaceae
Genus	:	*Achyranthes*
Species	:	*aspera*

Description:

Achyranthes aspera is an erect, many branched, spreading and quadrangular herb that lives several years. A wild, perennial, erect herb. Stem is herbaceous but woody below erect and branched, cylindrical, solid, angular, hairy, longitudinally striated, nodes and internodes are prominent. Leaves are ramal and cauline, simple, exstipulate, opposite decussate, petiolate .Inflorescence is spike with reflexed flowers arranged on long peduncle. Flowers are bracteate, shorter than perianth, dry membranous and persistent, sessile. complete, hermaphrodite, actinomorphic, pentamerous and hypogynous. Bract ovate and persistent.

Plate 2: ***Alternanthera sessilis*** **(L.) R.Br.**



Perianth made up of 10 stamens, out of which 5 are fertile and 5 are scale like, both alternating which each other, filaments slightly fused at the base, dithecous or versatile. Fruits are oblong capitates. Seed 2 to 3mm long, 1 to 1.5 mm wide, truncate above, reddish to dark brown and shiny, enclosed by persistent perianth and bracts, detaching easily from rachis.

Ecology:

Commonly found in disturbed areas. Prefers moist soil but can grow well in dry areas.

Distribution:

Native to southern Asia, Australia and some Pacific Islands..

Vernacular name:

Prickly chaff flower

Alternanthera sessilis

Systematic position

In the Bentham and Hooker system of classification , the systematic position of *Alternanthera sessilis* (L.) R.Br. is as follows

Kingdom	:	Plantae
Division	:	Angiosperms
Class	:	Dicotyledons
Sub class	:	Caryophyllidae
Order	:	Caryophyllales
Family	:	Amaranthaceae
Genus	:	*Alternanthera*
Species	:	*sessilis*

Description:

Alternanthera sessilis is a many branched, perennial, sometimes annual herb. Main root is a taproot white or brown. Cylindric, many branched villous in lines and transversely at

the nodes. Leaves are simple, opposite and decussate, paired blades are are generally in same size. The lamina are narrow and elongated, very variable in shape and size. Inflorescence are sessile spikes in the axils of leaves, isolated or grouped. Each flower is sustented by a white scarious bract, 0.75 -1 mm long with a pointed end, and 2 bracteoles similar to the bract. The ovary is compressed sub- orbicular, with a short style

Ecology:

The plant prefers moist soils, along ditches, fallow land. The plant grows in moist soils of the lakes, swamps, irrigation canals and rice paddies dams and navigation channels, up to 1200m altitude.

Distribution:

It is native to tropical and subtropical regions of the world. I t has been introduced to the Southern United States and its origin in Central and South America are uncertain.

Vernacular name:

Joy weed

Phytochemical screening:

The bio-activity of natural products is due to phytochemical having therapeutic, prophylactic, nutrional and antibacterial properties. Phytochemical screening of plant material is thus vital in the knowledge of their therapeutic properties. They have been found to inadvertently confer anti- microbial protections to humans due to compounds synthesized in the secondary metabolism as well as being immunodulatory (Al-Bayati and Al-Mola 2008).

The phytochemical analysis of different extracts of selected medicinal plant showed the presence of secondary metabolites such as alkaloids, flavonoids, tannins, phenols, terpenoids, glycosides, cardiac glycosides, anthroquinone, steroids, saponins (Table 1 and 2).

Table 1: Preliminary phytochemical analysis of *Achyranthes aspera* leaf (+ : present; -: absent)

Phytochemical analysis of *Achyranthes aspera* extracts						
S.NO	Phytochemical	Extracts				
		Ethanol	Acetone	Chloroform	Benzene	Aqueous
1.	Alkaloids	−	+	−	−	+
2.	Flavonoids	−	−	−	+	−
3.	Tannins	+	+	−	−	−
4.	Phenols	+	+	−	+	−
5.	Terpenoids	+	−	+	+	+
6.	Glycosides	+	+	+	+	−
7.	Cardiac glycosides	−	−	+	+	−
8.	Anthroquinone	−	−	−	−	−
9.	Steroids	+	+	+	−	+
10.	Saponins	−	−	−	−	+

Table 2: Preliminary phytochemical analysis of *Alternanthera sessilis* leaf (+ : present; -: absent)

Phytochemical analysis of *Alternathera sessilis* extracts						
S.NO	Phytochemical	Extracts				
		Ethanol	Acetone	Chloroform	Benzene	Aqueous
1.	Alkaloids	–	+	–	–	+
2.	Flavonoids	–	–	–	+	–
3.	Tannins	+	+	–	–	–
4.	Phenols	+	+	–	+	–
5.	Terpenoids	+	+	+	+	+
6.	Glycosides	+	+	+	+	–
7.	Cardiac glycosides	–	–	+	+	–
8.	Anthroquinone	–	–	–	–	–
9.	Steroids	+	+	+	–	+
10.	Saponins	–	+	+	–	+

Tannins had antibacterial activities and they decrease the bacterial proliferation by blocking key enzymes at metabolism (Mungole *et al.*, 2010). The acetone and ethanol extracts of a *Achyranthes aspera* contained tannins.(Table: 1). The chloroform, benzene and aqueous extracts revealed the absence of tannins.(Table:1) Neupane *et al.*, 2022 tested the quantitative phytochemical profiles and determined the free radical scavenging activity of phytochemical constituents of the entire plant of *Achyranthes aspera* Linn and revealed the presence of tannins. The different extracts of *Alternanthers sessilis* showed presence of tannins, flavonoids, phenols , saponins (Table: 2).

Alkaloids have analgesics, antimicrobial and antioxidant activity (Govindappa *et al.*, 2011). Alkaloids interfere with cell division, hence the presence alkaloids in the studied plants could account fot the inhibition of bacterial species in this study. The phytochemical analysis of different extracts of two medicinal plants showed the presence of alkaloids.(Table 1 & 2) Sobha kota *et al.*, 2017 and Dhale and Sonal 2013 reported similar results in Amaranthaceae members.

Saponins have antibiotic and antimicrobial activity (Mandal *et al.*, 2005) and this activity of saponin is due to its ability to cause leakage of protein and certain enzymes from the cell. Aqueous,acetone and chloroform extract revealed the presence of saponins in *Achyranthes aspera* and *Alternanthera sessilis.*

Flavonoids found in benzene extracts of *Achyranthes aspera* and *Alternanthera sessilis* plants. Flavonoids have antibacterial activities and the probably due to their ability to complex with extracellular and soluble proteins and bacterial cell wall (Nioku and Obi 2007).

Further more glycosides were present in all the extracts except aqueous. The result agree with Dhale and Sonal (2013) who reported flavonoids in *Achyranthes aspera*

Antibacterial activity:

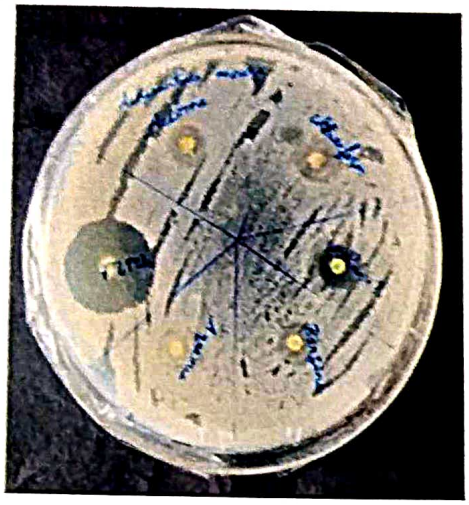
Life threatening diseases and high rate of mortality occur in animal and human population due to bacterial infection. Many bacteria both Gram positive and Gram negative contaminate food, water, air, soil, etc., and cause biological / microbial pollution. *Bacillus subtilis* is responsible for causing food borne gastroenteritis, *Escherichia coli,* cause diseases like mastitis albortion and upper respiratory complication. (Jawetzet *et al.*, 1987).

In this present study, the antibacterial activity of two extracts (*Achyranthes aspera* and *Alternanthera sessilis*) in five different solvents (acetone, chloroform, benzene, aqueous and ethanol) was investigated against three human pathogenic bacteria *(E.coli, B. Subtilis* and *S.aureus*) and the result were presented in (Table: 4 & 5). The inhibition zones against these species ranged in size from 3 to 33 mm in diameter. All of extracts examined prevented the growth of all of the pathogens in sample. Ethanolic extract of *Achyranthes aspera* highly inhibit the growth of *B.subtilis* (12 mm). Chloroform and aqueous extract of *Achyranthes aspera* showed lower inhibitory activity against all pathogens tested in the study.

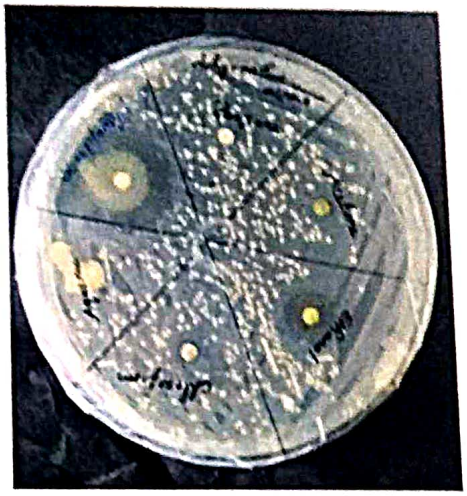
As shown as Table 5, chloroform extract of *Alternanthera sessilis* exhibited maximum inhibitory activity against *B. subtilis* (15 mm) subsequently followed by ethanol and, benzene (5mm) and ethanol(3mm) extracts.

Devi *et al.*, 2009 revealed that methanol extraction showed higher antibacterial activity than other extract. The variation of antibacterial activity among different crude extracts of this investigation might be due to distribution of varied antimicrobial substance. Similarly Owosemi *et al.*, (2010) reported that different extracts of plants show different antimicrobial activities on an organism. Therefore it is concluded that leaf extracts of *Achyranthes aspera* and *Alternanthera sessilis* used in the present study could be effectively processed to be utilized as a source for antibacterial therapeutic drug preparations.

Plate 3 : In *vitro* antibacterial activity of *Achyranthes aspera* leaf against human pathogens



Staphyloccous aureus



E.coli

Bacillus subtilis

Fig : 1 Antibacterial activity of *Achyranthes aspera*

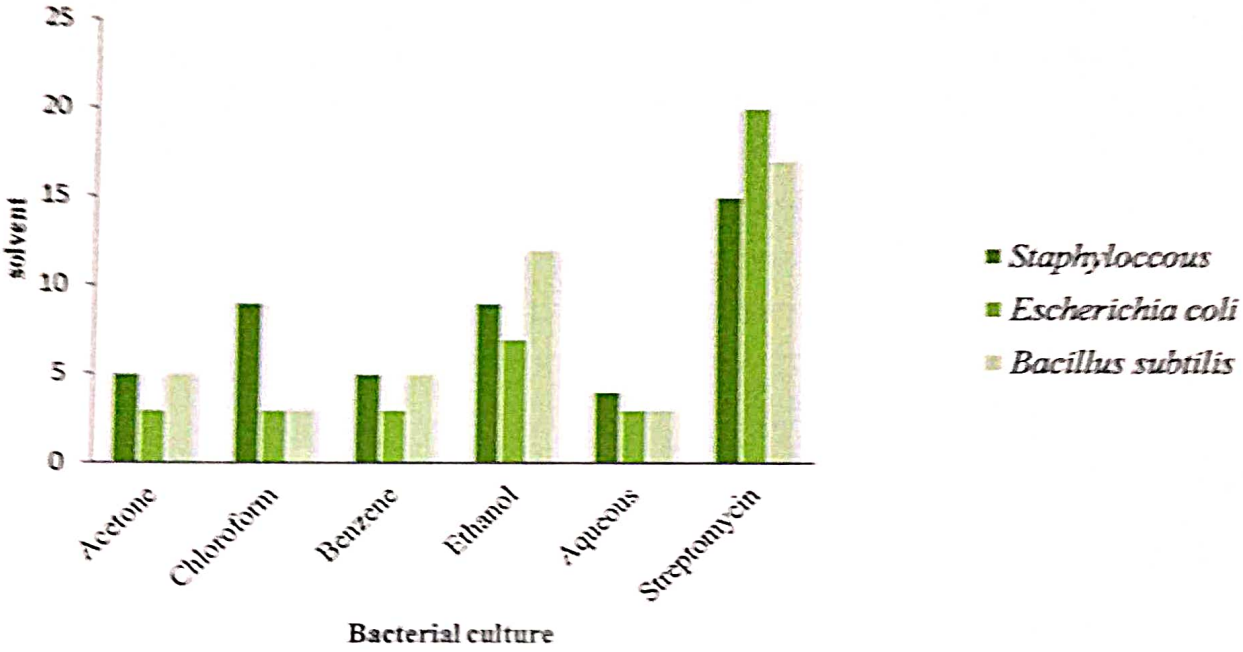
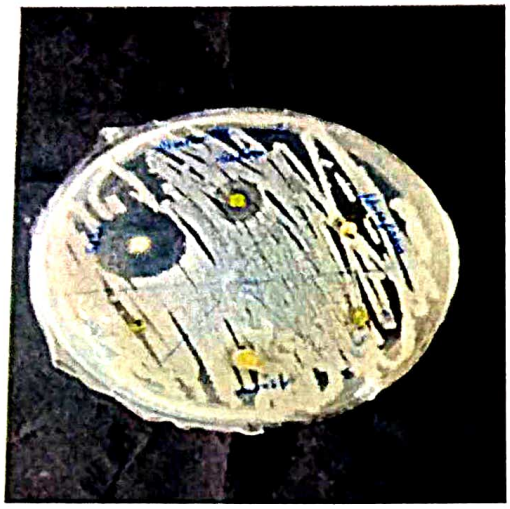
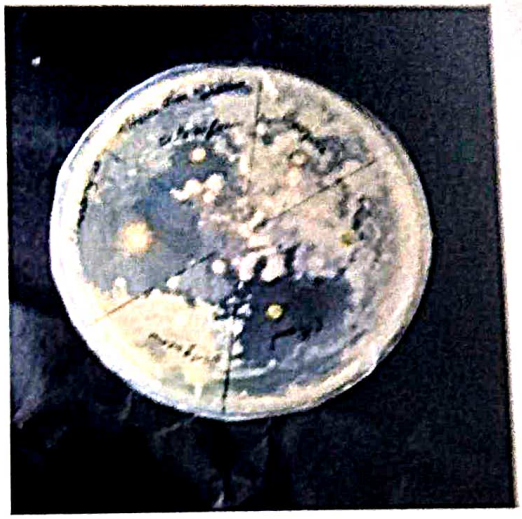


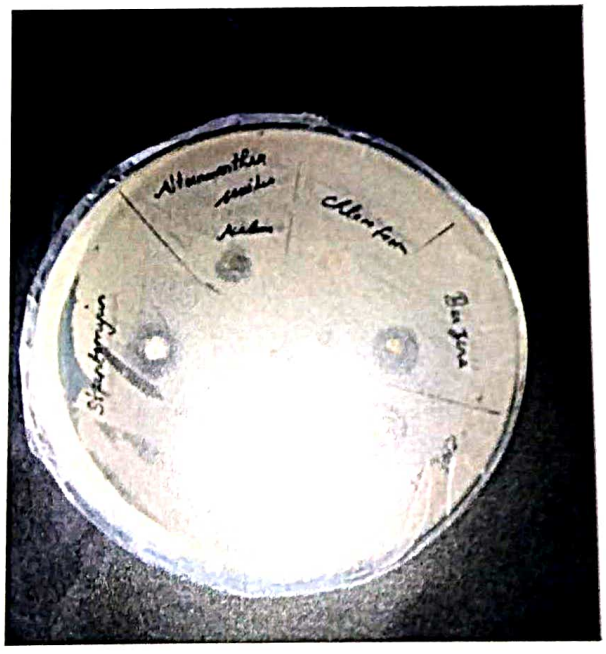
Plate 4 : In *vitro* antibacterial activity of *Alternanthera sessilis* leaf against human pathogens



Staphylococcus aureus



E.coli



Bacillus subtilis

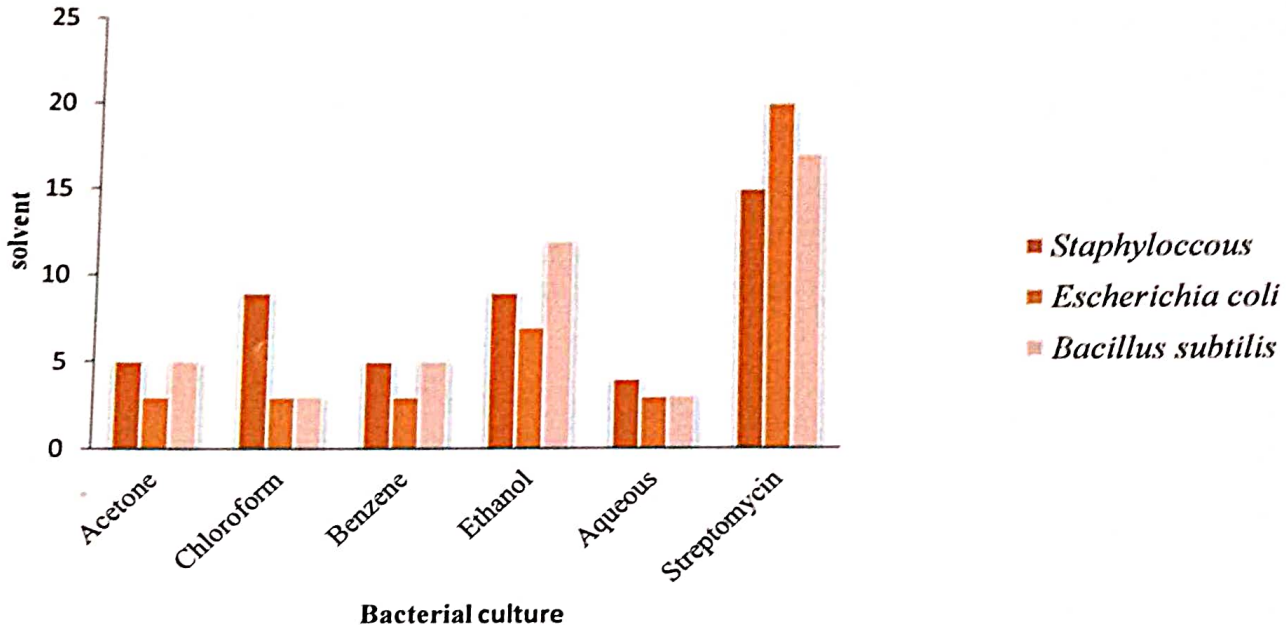
Table : 4 Antibacterial activity of different solvents of extracts of *Alternanthera sessilis* leaves

Bacterial Cultures	Acetone	Chloroform	Benzene	Aqueous	Ethanol	Streptomycin
Staphylo coccus	8mm	4mm	3mm	5mm	3mm	15mm
Escherichia coli	3mm	10mm	5mm	3mm	3mm	25mm
Bacillus subtilis	5mm	15mm	5mm	3mm	3mm	33mm

Control: Sterptomycin- (2.5 mg/ ml)

Leaf extract - (2.5mg/ ml)

Fig : 2 Antibacterial activity of *Alternanthera sessilis*



FI-TR analysis

The FT-IR spectroscopic analysis of the precent study reveals the different characteristics peak values with funtional groups of phytocomponents in stem and leaf extract of *A.aspera* and *A.sessilis*. FT-IR analysis of leaves extract of *A.aspera* confirms the presence of amines, sulphinic, esters, aralkyl and hydroxyl in the major peak vales at 467.71, 517.85, 615.25, 780.15, 831.26, 896.84, 1032.81, 1162.03, 1244.97, 1320.18, 1373.22, 1465.8,1514.98, 1636.49, 1739.67,1861.18, 2916.17, 3408.95 (Table: 4).The result of *A.sessilis* leaf extracts show the presence of amines, amides, sulphinic acid, ether, acetates, urethanes it shows the major peaks at 514.96, 619.11, 671.18, 781.12, 832.23, 894.91, 1019.31, 1108.99, 1160.1, 1238.21, 1320.18, 1387.69, 1638.42,2850.59, 2918.1,3418.59 (Table:5).

FT-IR analysis is proved to be a reliable and sensitive method as it proven a unique finger print for the biomolecules. It has been used as a requisite method to identify the various functional groups responsible for medicinal properties in the herbal drug (Devika *et al.*,2013).

Table: 5 FT-IR Spectroscopy Analysis *of Achyranthes aspera* leaf

S.NO	PEAK	BOND	FUNTIONAL GROUP
1	467.71	WEAK	C-Br
2	517.85	STRONG, AROMATIC	N-H
3	615.25	STRONG, PRIMARY AMINES	N-H
4	675.04	STRONG , PRIMARY AMINES	C-H
5	780.15	SRTONG, HYDRO COMPOUND	S-O
6	831.26	SRTONG, SULPHINIC ACID GROUP	N-O
7	896.84	WEAK NITRATE GROUP	C-O
8	1032.81	VERY STRONG, RING STRETCH	C-O
9	1162.03	VERY STRONG, ESTER GROUP	C-O-C
10	1244.97	STRONG, ARALKYL ASYMMETRIC	C-O
11	1320.18	STRONG, HYDROXYL GROUP	C-O
12	1373.22	STRONG, HYDROXYL GROUP	N=O
13	1465.8	STRONG AMINES	N=O

14	1514.98	AROMATIC, ASYMMETRIC	C=N
15	1636.49	WEAK, STRONG,AROMATIC	C=O
16	1739.67	STRONG , ESTER	C=O
17	1861.18	STRONG, STRETCH	C=O
18	2916.17	MEDIUM, ASYMMETRIC	H-C-H
19	3408.95	MEDIUM, WEAK	O-H

Fig :3 FT-IR spectrum of *Achyranthes aspera*

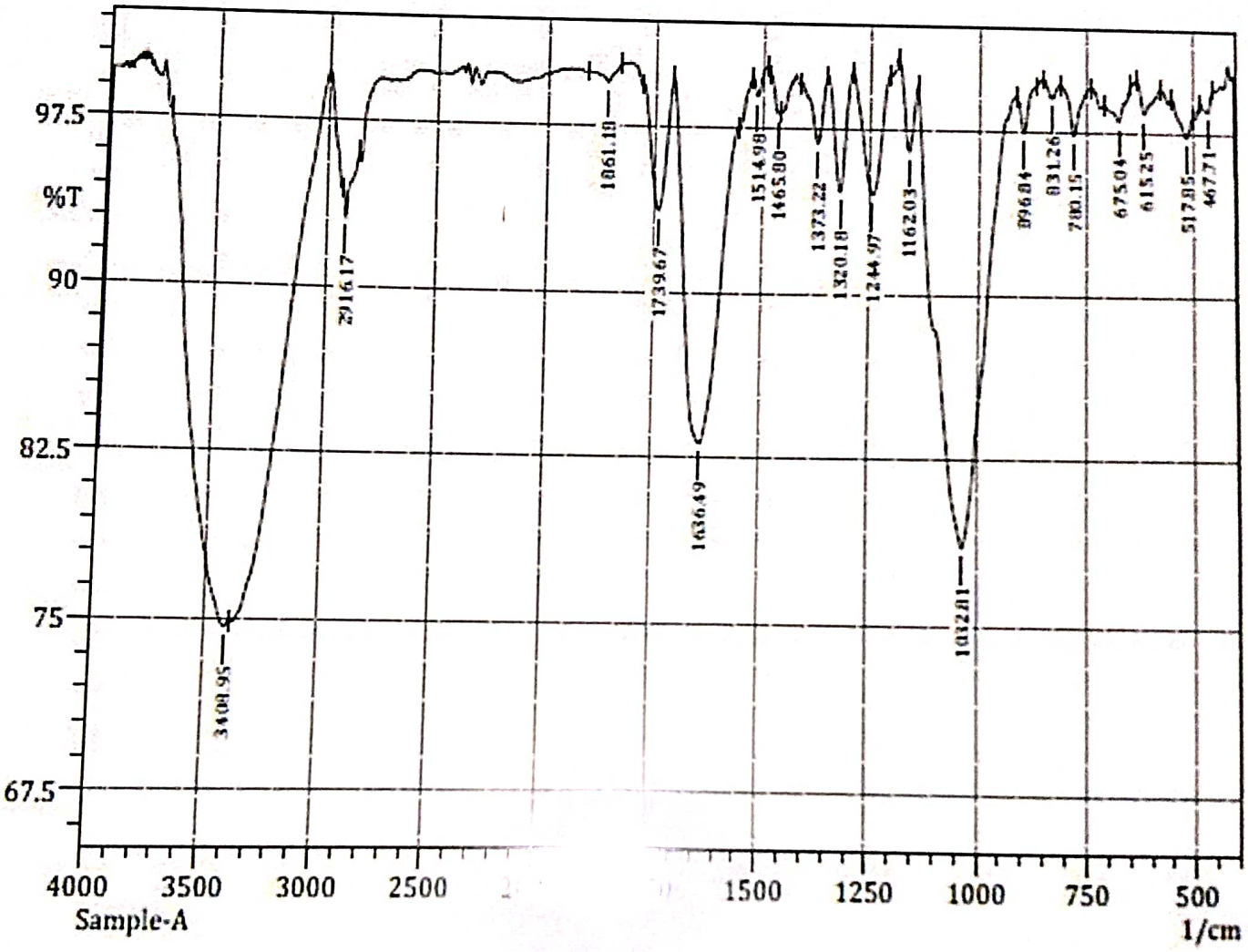
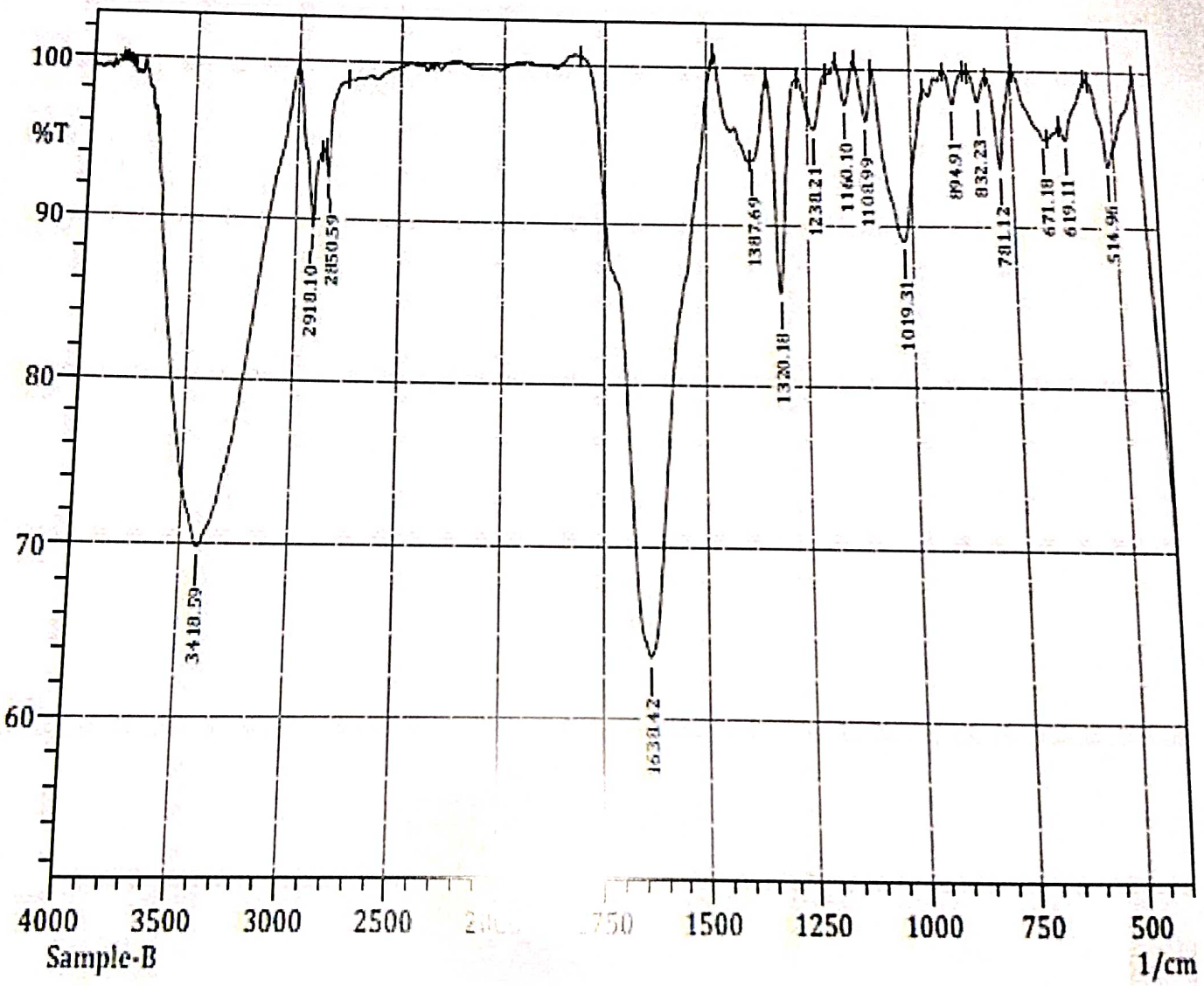


Table: 6 FT-IR Spectroscopy Analysis *of Alternanthera sessilis* leaf

S.NO	PEAK	BOND	FUNTIONAL GROUP
1	514.96	STORNG, AROMATIC	C-Br
2	619.11	STRONG, PRIMARY AMINES	N-H
3	671.18	STRONG, VINYL AMIDES	C-H
4	781.12	STRONG, HYDRO COMPONDS	C-H
5	832.23	STRONG, SULPHINIC ACID GROUPS	S-O
6	894.91	WEAK, NITRATE GROUP	N-O
7	1019.31	STRONG, EHTER	C-O
8	1108.99	STRONG, ESTERS GROUPS	C-O
9	1160.1	WEAK, VERY STRONG	C-O
10	1238.21	STRONG, ACETATES	C-O-C
11	1320.18	STORNG ARYL TERTIARY AMINE	C-N
12	1387.69	STRONG , NITRO GROUP	N=O
13	1638.42	MEDIUM, PRIMARY AMIDES	N-H
14	2850.59	MEDIUM, SYMMETRIC STRETCH	H-C-H
15	2918.1	MEDIUM, URETHANES	N-H
16	3418.59	MEDIUM	O-H

Fig : 4 FT-IR spectrum of *Alternanthera sessilis*



Biosynthesis of nanoparticles:

On mixing the aqueous extract of dried leaves of *Achyranthes aspera* and *Alternanthera sessilis* with 1 ml $AgNO_3$ solution, the colour of the solution changes from pale yellow to yellowish brown colour indicated the presence of silver nanoparticles in the aqueous leaf extract of the selected medicinal plants.(Plate 5&6)

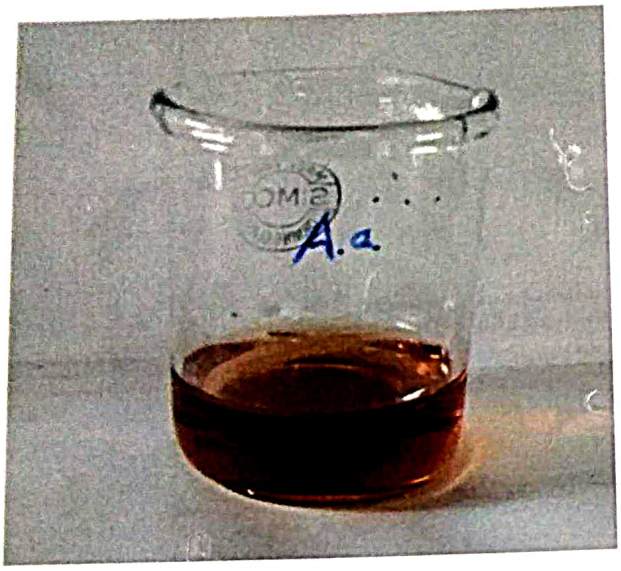
UV-vis spectrometer was utilized to two reacted mixtures containing AgNPs. At first UV-vis spectra did not show any peak in the wavelength region 400-600nm.After 24 hours reaction the peak height was increased as a function of reaction time. This indicated the biosynthesis of AgNPs . UV-vis spectra of the AgNPs and the aqueous leaf extract give a sharp peak at 420 nm after 24 hours incubation.Fig (3). The presence of an absorbance peak at about 420 nm clearly indicates the formation of AgNPs in the solution due to the surface plasmon resonance electrons present on the nanoparticles surface. This peak was caused by the absorption of the plasmonic sliver nanoparticles, so the formation AgNPs was confirmed. The surface plasmon resonance of sliver nanoparticles depended on the free electrons interacting with electromagnetic radiation. Thus the difference in dimension and morphology or sliver nanoparticles could affect on the collective excitation oscillations that might lead to the different absorption peaks singh et al., (2016).

Hemlata *et al.*, 2022 reported UV- visible spectroscopy is one of the most widely used techniques for structural characterization of nanoparticles. The presence of an absorbance peak at about 420 nm clearly indicates the formation of AgNPs in the solution due to surface SPR electrons present on the nanoparticles surface. The intensity of the SPR band increased with reaction time, indicating the synthesis of the AgNPs.

Plate : 5 Synthesis of sliver nanoparticles

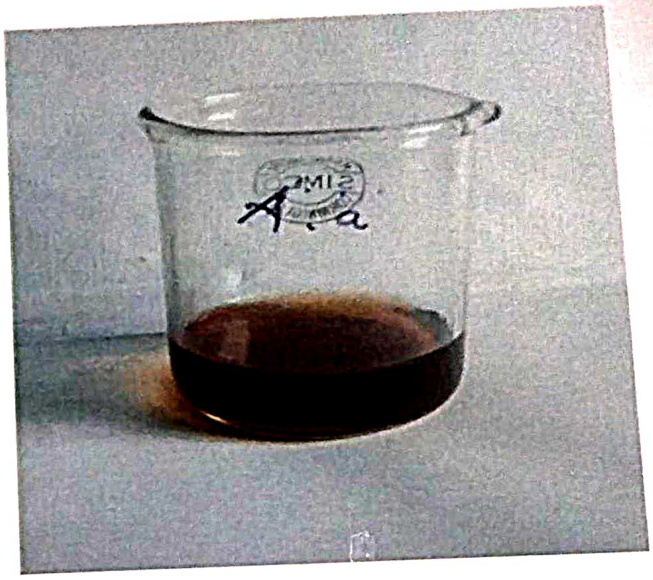
Leaves:

Initial



Achyranthes aspera

Final

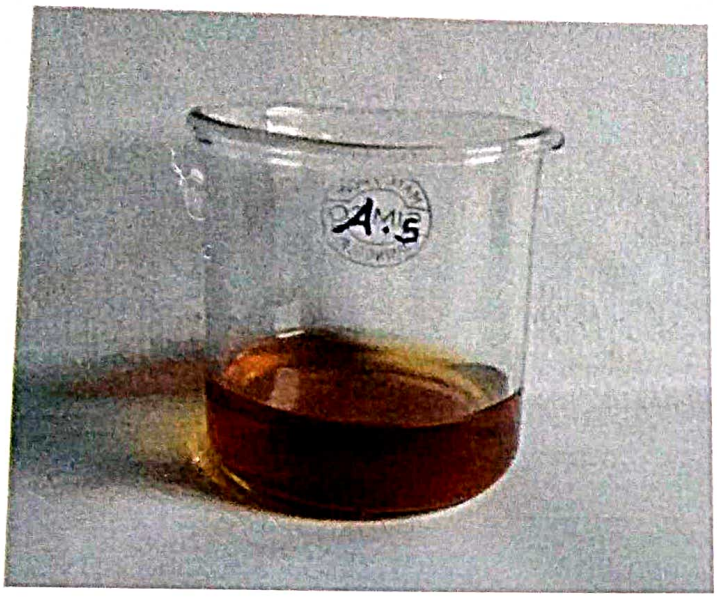


Achyranthes aspera

Plate: 6 Synthesis of sliver nanoparticles

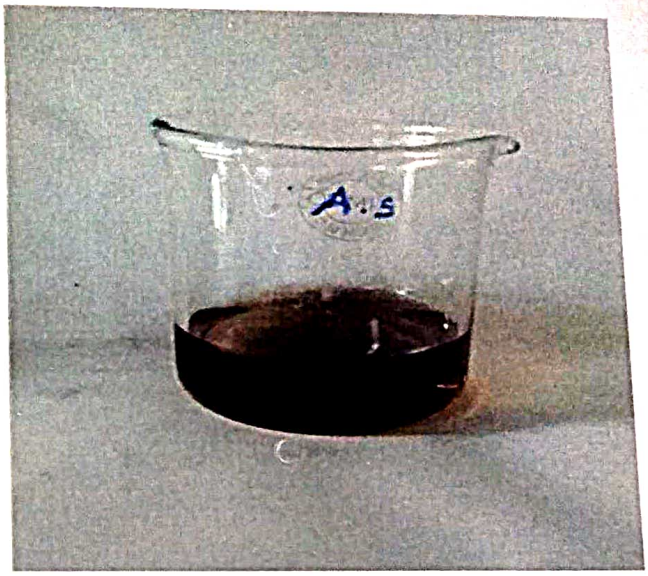
Leaves:

Initial



Alternanthera sessilis

Final

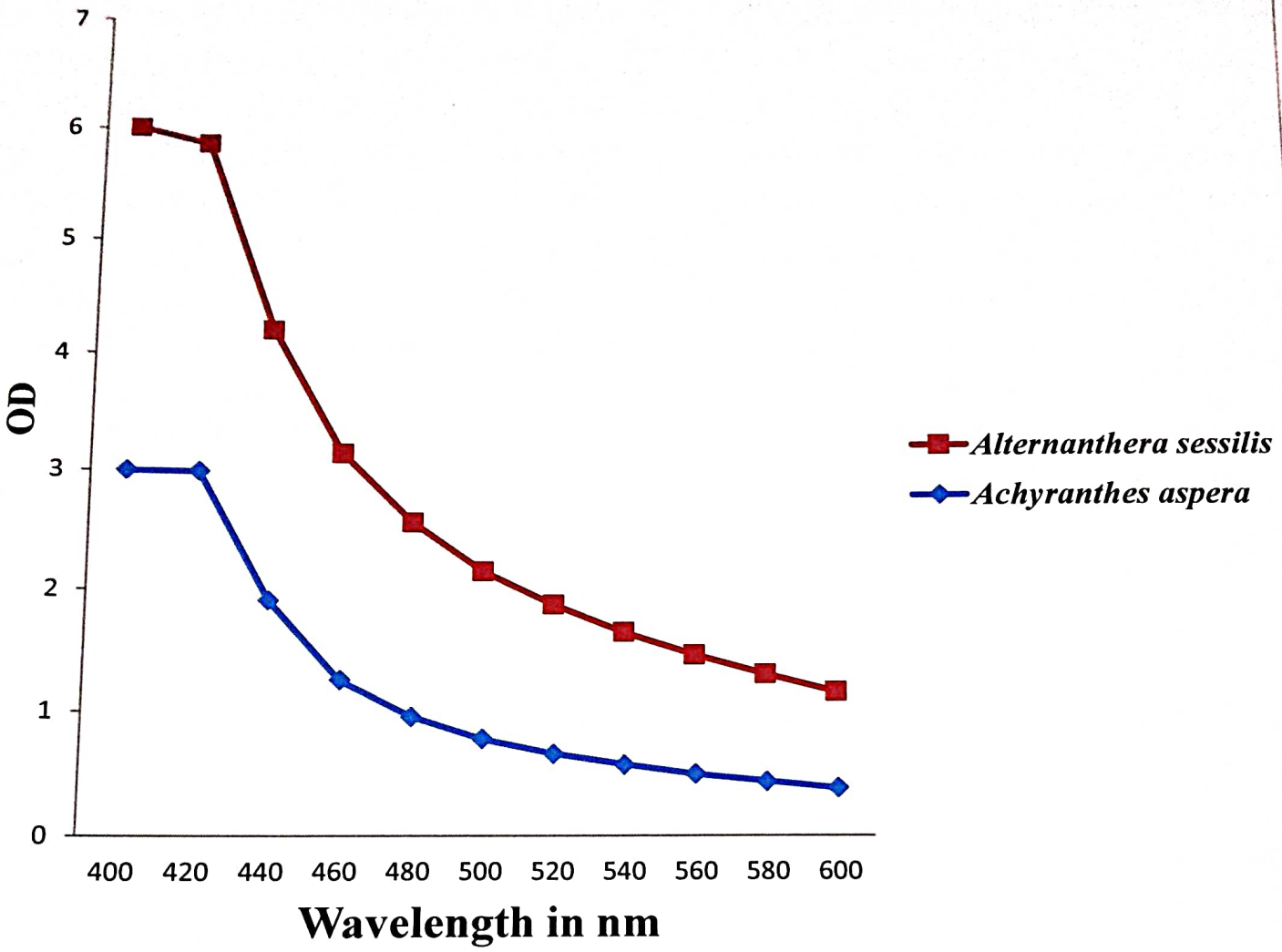


Alternanthera sessilis

Table : 7 Synthesis of sliver nanoparticles

nm	OD (*A.a*)	OD (*A.s*)
400	3.000	3.000
420	3.000	2.853
440	1.924	2.293
460	1.273	1.900
480	0.973	1.621
500	0.789	1.397
520	0.669	1.237
540	0.579	1.096
560	0.504	0.982
580	0.442	0.885
600	0.385	0.793

Fig : 3 Synthesis of Silver nanoparticals



SUMMARY AND CONCLUSION

SUMMARY AND CONCLUSION

The project entitled "Evaluation of phytochemical, antibacterial activity and synthesis of sliver nanoparticles of *Achyranthes aspera* and *Alternanthera sessilis* R.Br" deals with the evalution of phytochemical profile, antibacterial activity, synthesis of sliver nanoparticles from the leaves of *Achyranthes aspera* and *Alternanthera sessilis* of a family Amaranthaceae.

The present work is focused on the following aspects:

- The phytochemical evaluation of dried leaf powder of *Achyranthes aspera* and *Alternanthera sessilis.*
- Identification the chemical compound of *Achyranthes aspera* and *Alternanthera sessilis.*
- Amtibacterial activity of leaf powder of *Achyranthes aspera* and *Alternanthera sessili* by disc diffusion method.
- Synthesis of sliver nanoparticles in *Achyranthes aspera* and *Alternanthera sessilis*

The photochemical analysis shows the presence of alkaloids, flavonoids, tannins, terpenoids, phenols, glycosides, cardiac glycosides, anthroquinone, steroids and saponins. The antibacterial activity is high in *Alternanthera sessilis* than *Achyranthes aspera.*

The biological synthesis of nanoparticles in increasingly regarded on a rapid ecofriendly and easily sealed up technology. Among the biological nanoparticles those produced by medicinal plant have been found to be the most active possibly due to the attachment of several pharmacologically active residues(Sigh *et al.*, 2016). The sliver AgNP present *A.aspera* and *A.sessilis* might have pharmacologically active residues. The biosynthesized nonoparticles are formed by an ecofriendly technique and green chemistry.

This method utilized less energy, minimized the toxic chemicals, simplified the procedure and exploited the natural materials being able to regenerate. So that further investigation will be carried out to characterize the nanoparticles too evaluate their efficacy of antibacterial activity.

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FLORA OF BOTANICAL GARDEN-ST. MARY'S COLLEGE (AUTONOMOUS) THOOTHUKUDI TAMILNADU

A short term field project submitted to

ST. MARY'S COLLEGE (AUTONOMOUS), THOOTHUKUDI

Affiliated to

MANONMANIAM SUNDRANAR UNIVERSITY

In partial fulfillment of the requirement for the degree of

MASTER OF SCIENCE IN BOTANY

BY

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DEPARTMENT OF BOTANY

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APRIL-2023

CERTIFICATE

This is to certify that this project work entitled "**FLORA OF BOTANICAL GARDEN - ST. MARY'S COLLEGE, (AUTONOMOUS), THOOTHUKUDI, TAMIL NADU**" Submitted to Department of Botany **ST. MARY'S COLLEGE (AUTONOMOUS) THOOTHUKUDI TAMILNADU** affiliated to **MANONMANIAM SUNDARANAR UNIVERSITY** in partial fulfillment for the degree of **Master of Science in Botany**, and is a record of work done in the department of Botany, St.Mary's College (Autonomous), Thoothukudi during the year 2022-2023 by the following students.

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