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Research Article

Preliminary Phytochemical Screening and GC-MS Analysis of Ethanolic Extract of Mangrove Plant-Bruguiera Cylindrica (Rhizho) L.

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ABSTRACT
The aim of the present study is to screen the phytochemicals present in the leaf extract of *Bruguiera cylindrica* (*B.Cylindrica*) and further analysis of the bioactive components present in it by Gas chromatography-Mass spectrometry (GC-MS) analysis. Phytochemical screening of the sequential extracts of *B.Cylindrica* leaves revealed the presence of various bioactive components of which Carbohydrate, Protein, Amino Acids, Lipids, Fatty acids, Fibre, Alkaloids, flavonoids, Tannin, Tri terpenoids, Saponins, Phenols /Gallicacid equivalent, Glycosides, Cardiac Glycosides, Lignin, Volatile Oil and Steroids In this work, forty eight phytochemicals were analysed with their fragmentation pattern, molecular formula, molecular weight, structure of phytocomponants by GC-MS technique. The compounds identified such as: 2-Cyclopenten-1-one, 2-hydroxy , 1,1,4-Trimethyl-3-pyrazalone, Phenol,1-Amino-2,6-dimethylpiperidine: , 5,9-Dodecadien-2-one, 6,10-dimethyl(E,E), 6-Desoxy-l-altritol, 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl, Ethyl hydrogen succinate, 3,4-Altrosan, 2-Coumaranone, Benzofuran, 2,3-dihydro, 8-Azabicyclo[3.2.1]octan-3-ol, 8-methyl, endo, D-Allose, 2-Cyclohexen-1-one, 4-(3-hydroxybutyl) -3,5,5-trimethyl-,Tetradecanoic acid,3,7,11,15-Tetramethyl-2-hexadecen-1-ol, n-Hexadecanoic acid, 3-O-Methyl-d-glucose, Phytol, 9,12-Octadecadienoic acid, methyl ester, 9-Octadecenal, (Z)-, Octadecanoic acid, 5-Naphthalen-1-yl-2H-pyrazol-3-ol-etc. From the results, it could be concluded that *B.Cylindrica* contains various bioactive compounds have various biological activities. Therefore, it is recommended as a plant of phytopharmaceutical importance.

Keywords: phytochemical components, *Bruguiera cylindrica,* gas chromatography-mass spectrometry (GC-MS) analysis.

INTRODUCTION

Medicinal plants are of great importance to the health of individuals and communities in general. About 3.4 billion people in the developing world depend on plant-based traditional medicines. This represents about 88 per cent of the world's inhabitants, who rely mainly on traditional medicine for their primary health care. The World Health Organization (WHO) supports the use traditional medicine provided they are proven to be efficacious and safe (WHO 1985). Hence there is a need to validate the ethnomedicinal use of herbal medicine and subsequently isolate and characterize the compounds which are likely to be added to the potential list of drugs[1]. The primary benefits of using plant derived medicines are that they are relatively safer than synthetic alternatives, offering profound therapeutic benefits and more affordable treatment. According to WHO, a medicinal plant is any plant which, in one or more of its parts contains substances that can be used for therapeutic purposes, or which are precursors for chemo-pharmaceutical semi synthesis. Such a plant will have its parts including leaves, roots, rhizomes, stems, barks, flowers, fruits, grains or seeds, employed in the control or treatment of diseases/ailments and therefore contains chemical components that are medically active[2]. These non-nutrient plant chemical compounds or bioactive components are often referred to as phytochemicals ('phyto-'from Greek – *phyto* meaning 'plant') or phytoconstituents[3,4]. The phytochemical analysis of the plants is important commercially and has great interest in pharmaceutical companies for the production of new drugs for curing various diseases[5].

Mangrove vegetations constitute approximately eighty species belonging to twenty families (Duke,1992)[6]. Mangroves are distributed circumtropically, occurring in 112 countries and territories. A total number of 34 true mangrove species are present in the mangroves of India, including both east and west coasts and Andaman and Nicorbar islands. Among them, 31 species are present in the wetlands of Orissa followed by 27 species in Sunderbans of West Bengal and 24 species are present in Andaman and Nicobar Islands. Along the east coast, the least number of 14 species are present in Tamil Nadu

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Table 1: Extractive values of leaves extract of *B. cylindrica*

S.No	Types of Extract	Extraction value(in grams)
1	Hexane	40
2	Chloroform	32
3	Ethanol	5.4
4	Aqueous extract	6

mangroves[7].

Tamil Nadu has a coastline of about 950 km. along the coastline major mangrove wetlands are present in two areas: one at Pichavaram in Cuddalore District and the other in the Muthupet region in Thiruvarur-Thanjavur Districts. Small patches of mangroves are also present along the Palk Bay, particularly in the Devipattinam region and also in some of the islands of the Gulf of Mannar in Ramanathapuram District. Pichavaram Mangroves is situated in the southeast coast of India, located about 240 km south of Chennai City and about 45km south of Cuddalore. According to recent botanical survey conducted by M.S.Swaminathan Research Foundation, a total number of 12 true mangrove plant species are present in this Pitchavaram mangrove wetland[8]. The better known taxa are the members of Rhizophoraceae namely Rhizophora, Bruguiera, Ceriops and Kandelia[6].

Numerous mangrove plants are being used in folklore medicine, and recently, extracts from mangroves and mangrove-dependent species have proven activity against human, animal and plant pathogens, but only limited investigations have been carried out to identify the metabolites responsible for their bioactivities as well as cytotoxic and scavenging activities[9,10,11]. Commercial use of mangroves as source of timber, fuel has long been recognized in tropical coastal zones. Besides, mangroves also provided many non-timber products such as tannin, fish poison, medicine, food, fodder, etc. They have been used as traditional medicine in South Asian countries including India[12]most yet to be explored[13].

A number of mangroves and associates are a rich source of steroids, triterpenes, saponins, flavonoids, alkaloids and tannins. Extracts from different mangrove plants are reported to possess diverse medicinal properties such as antibacterial and antihelminthics[12]. Previous studies on mangrove plant parts and its major chemical classes displayed various levels of biological activities such as antifungal, antibacterial, antifeedant, molluscicidal and pesticidal[12] antibacterial, antifungal, antiplasmodial, cytotoxic, antifouling, hepatoprotective, ichtyotoxic and free radical scavenging activities which are attributable to the chemical components of mangrove plants such as flavonoids, alkaloids, triterpenes, saponins and tannins[14]. Recently, the medicinal value of mangroves and associated plants persist to provide priceless therapeutic agents, both in modern medicines and in traditional systems[15]. Hence scientists are veering in search of effective remedies from mangroves for diseases such as diabetes, asthma, cancer, ulcer, wounds and AIDS[12].

Bruguiera cylindrica (L.) (*B. cylindrica* Blume) (family: Rhizophoraceae) is a rare tree mangrove found along the western coast of India. Traditionally *B. cylindrica* used to treat hepatitis (Fr, L, R)[16]. The bark of *B.cylindrica* is used to stop haemorrhage and applied to malignant ulcers. Also tested for antiviral and larvicidal activity, biotoxicity on tobacco mosaic virus and fingerlings of fish[9, 10]. The stem bark of *B.cylindrica* has been studied for its antioxidant effect[17]. *B.cylindrica* has been used as traditional medicine for years with variety of biological activity such as anti-oxidant, antibacterial, antifungal, antiulcer, anticancer and antiplasmodial and properties[18].

B. cylindrica were tested against antibiotic resistant pathogens (ARB) viz. Staphylococcus aureus, Streptococcus pneumonia, Klebsiella pneumoniae, Pseudomonas aeruginosa and eye pathogens (EP) viz. E. coli, Proteus, Acinetobacter and Staphylococcus epidermidis (Ravikumar, et al., 2011)[11]. Leaf and bark of *B. cylindrica* were studied for antimicrobial effect[19]and antiviral activity[20] against NDV, VV, EMCV, SFV, HBV[11,21] Newcastle disease, Vaccinia and Hepatitis B viruses[11]. The extracts of the seeds of *B. cylindrica* showed antiviral activity against TMV. Leaves were used for treating blood pressure and used as fodder for livestock[22]. Pentacyclic triterpenoid esters from the fruits and brugine alkaloid from stem and bark have been isolated[11, 23, 24]. Recent studies are involved in the identification and isolation of new therapeutic compounds of medicinal importance from higher plants for specific diseases. Natural products derived from plants are being tested for presence of new molecules with new modes of pharmacological action. A special feature of higher plants is their capacity to produce a large number of secondary metabolites[25, 26].

Gas chromatography-mass spectrometry (GC-MS) is a method that combines the features of gas liquid chromatography and mass spectrometry to identify different substances within a test sample. GC-MS can provide meaningful information for components that are volatile, non-ionic, and thermally stable and have relatively low molecular weight. The aim of the present study is to screen the phytochemicals present in the leaves extract of *B. cylindrica* and analyse of the bioactive components present in it by GC-MS analysis.

METERIALS AND METHODS

Plant Collection: The leaves of the *B. cylindrica* were collected from Pichavaram mangrove forest near Chidambaram, Cuddalore district, Tamilnadu after obtaining permission from the District Forest Office and taxonomically identified by botanist of Centre of Advanced Study in Marine Biology, Annamalai University, Parangipettai, India.

Plant Extraction

Sequential Extraction of leaf of *B. cylindrica*: The *B. cylindrica* leaves were air-dried and powdered with the help of mixer grinder. The powdered leaf were then extracted with soxhlet apparatus using sequential solvents that were Hexane, Chloroform and Ethanol for 16 h. Aqueous extract was also obtained by soaking sequential plant part powder in double distilled water for 2-3 days and then filtered with cheese cloth. The extracts were than concentrated on a rotary evaporator below 50°C and were

Table 2: Qualitative Phytochemical screening of sequential extracts of *B. cylindrica* leaf

S.No	Phytochemical screening	Aqueous Extract	Hexane Extract	Ethanol Extract	Chloroform Extract
1	Carbohydrate	+	+	+	+
2	Protein	+	+	+	+
3	Amino Acids	+	+	+	+
4	Lipids	+	+	+	+
5	Fatty acids	+	+	+	+
6	Fiber content	+	+	+	+
7	Alkaloids	+	+	+	+
8	Flavonoids	+	+	+	+
9	Tannin	+	+	+	+
10	Triterpenoids	+	+	+	+
11	Saponins	+	+	+	+
12	Phenols/Gallicacid equivalent	+	+	+	+
13	Glycosides	+	+	+	+
14	Cardiac Glycosides	-	-	+	-
15	Lignin	+	+	+	+
16	Volatile Oil	-	+	-	+
17	Steroids	-	+	-	+

Negative (-) absent, Positive (+) present

stored in airtight containers at 4°C temperature for further studies.

Qualitative preliminary phytochemical analysis: The raw material, Aqueous, Hexane, Ethanol and Chloroform extracts were chemically tested for phytochemical constituents using standard procedures recommended by Sofowora, (1994)[27] and Trease and Evans (1989)[28]. (For each test 5ml of each of extracts were used). The phytochemical tests carried out in this work include carbohydrate, protein, amino acids, lipids, fatty acids, fibre, alkaloids, flavonoids, tannin, triterpenoids, saponins, phenols/gallicacid equivalent, glycosides and lignin[29-40].

GC –MS Analysis: GC-MS plays a key role in the analysis of unknown components of plant origin. 2µl of the ethanolic extract of *B.Cylindrica* was employed for GC-MS for analysis of different compounds. Instruments and chromatographic conditions GC-MS analysis was carried out on a GC clarus 500 Perkin Elmer system comprising a AOC-20i auto sampler and gas chromatograph interfaced to a mass spectrometer (GC-MS) instrument employing the following conditions: columnElite-1 fused silica capillary column (30 ×0.25 mm × ID x 1µm of capillary column, composed of 100% Dimethyl poly siloxane), operating in electron impact mode at 70 eV; helium (99.999%) was used as carrier gas at a constant flow of 1ml/min and an injection volume of 0.5 EI was employed (split ratio of 10:1) inject or temperature 250°C; ion-source temperature280°C. The oven temperature was programmed from 110°C (isothermal for 2min), with an increase of 10°C/min, to200°C/min, then 5°C/min to 280°C/min, ending with a 9 min isothermal at 280°C.Mass spectra were taken at 70 eV; a scan interval of 0.5 s and fragments from 45 to 450Da.The eluted component is detected in the mass detector. The spectrum of the unknown component is compared with the spectrum of the known components stored of the NIST library and determines the name, molecular weight and fragrances, floral fragrances, pesticide residues. Terpenes, steroids, alkaloids, phenols, flavonoids and fatty acids are some of the useful components analysed in the GC-MS study.

RESULTS AND DISCUSSION

The highest yield of extract of *B.Cylindrica* leave was found with Hexane and the lowest with ethanol and the details are provided in Table-1

Phytochemical screening of the sequential extracts of *B.Cylindrica* leaves revealed the presence of various bioactive components of carbohydrate, protein, amino Acids, lipids, fatty acids, fibre, alkaloids, flavonoids, tannin, triterpenoids, saponins, phenols /gallicacid equivalent, glycosides, cardiac glycosides, lignin, volatile oil and steroids as presented in Table-2.

The phytochemical screening of leaves of *B.Cylindrica* showed the presence of Carbohydrate, Protein, Amino Acids, Lipids, Fatty acids, Fibre, Alkaloids, flavonoids, Tannin, Tri terpenoids, Saponins, Phenols/Gallicacid equivalent, Glycosides and Lignin are present in all the four extracts. Cardiac Glycosides were present only in ethanol extract, and Volotile oil and steroids were present in hexane and chloropharm extract which are known to exhibit medicinal activities.

The GC-MS analysis showed the presence of forty eight compounds in the leaves of *B.Cylindrica* by comparing their retention times and by interpretation of their mass spectra. The compounds identified and their retention time, molecular formula, molecular weight (MW),and concentration (peak area%)are presented in Table-3.The results pertaining to GC-MS analysis of the phytocomponents showed fourteen peaks - Fig -1&2. The fragmentation pattern and structure of the various phytochemicals which contribute to the medicinal activity of the plant ethanol leaf extract of *B.Cylindrica* are listed in Table 4.

Plant cells produce two types of metabolites. Primary metabolites are involved directly in growth and metabolism (sugars, proteins, fats, vitamins, crude fiber carbohydrates, lipids and proteins)[41]. Most natural

Table 3: Components identified in ethanol leaf extract of B. cylindrica

S.N	Peak Name	Formula	MW	Retention Time	Peak Area	% Peak area
1	1H-Tetrazole-1,5-diamin	CH_4N_6	100	2.76	1710986	0.4899
2	Ethanamine, 2-chloro-N,N-dimethyl	$C_4H_{10}ClN$	107	5.72	503260	0.1441
3	3-Buten-1-amine, n-butyl-N-methyl	$C_9H_{19}N$	141	6.06	170201	0.0487
4	2-Cyclopenten-1-one, 2-hydroxy	$C_5H_6O_2$	98	6.76	2505079	0.7173
5	2-Furancarboxaldehyde, 5-methyl	$C_6H_6O_2$	110	7.21	1625430	0.4654
6	1,1,4-Trimethyl-3-pyrazalone	$C_6H_{12}N_2O$	128	7.92	5046222	1.4450
7	N,N-Dinitropiperazine	C4H8N4O4	176	8.09	2039084	0.5839
8	2,5-Pyrrolidinedione, 1-methyl	$C_5H_7NO_2$	113	8.09	2039084	0.5839
9	Phenol	C_6H_6O	94	8.42	3491572	0.9998
10	1-Amino-2,6-dimethylpiperidine:	$C_7H_{16}N_2$	128	8.91	19049844	5.4550
11	N,N-Dimethyl-1-propanamine	$C_5H_{13}N$	87	9.32	704148	0.2016
12	5,9-Dodecadien-2-one, 6,10-dimethyl(E,E)	$C_{14}H_{24}O$	208	9.65	14047165	4.0225
13	6-Desoxy-l-altritol	$C_6H_{14}O_5$	166	10.81	4948005	1.4169
14	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl	$C_6H_8O_4$	144	11.15	8576939	2.4560
15	Octanoic Acicd	$C_8H_{16}O_2$	144	11.51	1952704	0.5592
16	Ethyl hydrogen succinate	$C_6H_{10}O_4$	146	11.89	3571152	1.0226
17	Benzenecarboxylic acid	$C_7H_6O_2$	122	12.11	977829	0.2800
18	3,4-Altrosan	$C_6H_{10}O_5$	162	12.23	3871035	1.1085
19	3,4-Dihydroxy-5-methyl-dihydrofuran-2-one	$C_5H_8O_4$	132	12.57	937659	0.2685
20	2-Coumaranone	$C_8H_6O_2$	134	12.68	5375912	1.5394
21	Benzofuran, 2,3-dihydro	C_8H_8O	120	13.03	1693331	0.4849
22	Caprolactam	$C_6H_{11}NO$	113	13.37	1781170	0.5100
23	2-Methoxy-4-vinylphenol	$C_9H_{10}O_2$	150	14.00	2612899	0.7482
24	1,2-Octadecanediol	$C_{18}H_{38}O_2$	286	14.72	1363925	0.3906
25	n-Decanoic acid	$C_{10}H_{20}O_2$	172	14.83	1525207	0.4367
26	1-Dodecanone, 1-cyclopropyl-	$C_{15}H_{28}O$	224	15.70	209268	0.0599
27	8-Azabicyclo[3.2.1]octan-3-ol, 8-methyl-, endo-	$C_8H_{15}NO$	141	15.90	2013700	0.5766
28	Ecgonine	$C_9H_{15}NO_3$	185	16.10	1245106	0.3565
29	8-Azabicyclo[3.2.1]octan-3-ol, 8-methyl, endo	$C_8H_{15}NO$	141	16.37	5646912	1.6170
30	2-Nitrotoluene-à,à-diol diacetate	$C_{11}H_{11}NO_6$	253	17.44	1164128	0.3334
31	Benzeneacetonitrile, 4-hydroxy-	C_8H_7NO	133	17.62	1124667	0.3221
32	Dodecanoic acid	$C_{12}H_{24}O_2$	200	17.92	541960	0.1552
33	D-Allose	$C_6H_{12}O_6$	180	18.03	15422049	4.4162
34	2H-Inden-2-one, 1,4,5,6,7,7a-hexahydro-7a-methyl-, (S)-	$C_{10}H_{14}O$	150	18.78	3438399	0.9846
35	Octadecane, 1-(ethenyloxy)-	$C_{20}H_{40}O$	296	19.82	2437649	0.6980
36	1,6-Anhydro-á-D-glucofuranose	$C_6H_{10}O_5$	162	20.04	2801334	0.8022
37	2-Cyclohexen-1-one, 4-(3-hydroxybutyl)-3,5,5-trimethyl-	$C_{13}H_{22}O_2$	210	20.21	9936330	2.8453
38	Tetradecanoic acid	$C_{14}H_{28}O_2$	228	20.74	7061333	2.0220
39	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	$C_{20}H_{40}O$	296	21.36	8489220	2.4309
40	Ethyl 2-ethyl-3-hydroxyvalerate	$C_9H_{18}O_3$	174	22.10	1573462	0.4506
41	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	23.82	26250920	7.5170
42	3-O-Methyl-d-glucose	$C_7H_{14}O_6$	194	24.62	137626416	39.4098
43	Phytol	$C_{20}H_{40}O$	296	27.76	1643698	0.4707
44	9,12-Octadecadienoic acid, methyl ester	$C_{19}H_{34}O_2$	294	26.72	3006831	0.8610
45	9-Octadecenal, (Z)-	$C_{18}H_{34}O$	266	27.87	18659844	5.3433
46	Octadecanoic acid	$C_{18}H_{36}O_2$	284	28.34	5238770	1.5001
47	5-Naphthalen-1-yl-2H-pyrazol-3-ol	$C_{13}H_{10}N_2O$	210	30.90	2768748	0.7928
48	Bicyclo[2.2.1]heptan-2-one, 1,4,7,7-tetramethyl	$C_{11}H_{18}O$	166	31.01	836993	0.2397

MW: Molecular weight

products are compounds derived from primary metabolites such as amino acids, carbohydrates and fatty acids and are generally categorized as secondary metabolites. Secondary metabolites are considered products of primary metabolism and are generally not involved in metabolic activity (alkaloids, phenolics, essential oils and terpenes, sterols, flavonoids, lignins, tannins, etc.). These secondary metabolites are the major source of pharmaceuticals, food additives, fragrances and pesticides[42].

The qualitative phytochemical studies of *B.Cylindrica* extract of naked the presence of carbohydrate, protein, amino acids, lipids, fatty acids, fibre, alkaloids, flavonoids, tannin, tri terpenoids, saponins, phenols/gallicacid equivalent, glycosides, cardiac glycosides, lignin, volatile oil and steroids. Phytochemical constituents in crude leaf extracts of *B.Cylindrica* leaves has been studied in one study showed alkaloid, terpenoid, flavonoid, phenol, steroid, saponin, tannin, protein, carbohydrate and aminoacid[11]. Phytochemical analysis of different parts of *B.Cylindrica* in another study showed Collar contains protein and tannin, Hypocotyls – sugar, protein, phenolyic group and catachin, Bark – sugar, phenolic group and tannin. Chemical compounds of *B. cylindrica* contain fatty acids, hydroxy, secondary alcohols, diketones, Hydrocarbons Esters & ketones, unidentified compounds [43,44].

Primary metabolites like carbohydrate, protein, amino acids, lipids, fatty acids and fiber present in ethanolic extract of *B.cylindrica.* Plants containing carbohydrates and glycosides are known to exert a beneficial action on immune system by improving body strength and hence are valuable as dietary supplements[45]. Proteins contributed to the structure and functions of the living cell, they occur as independent units as well as in combination with lipids, nucleic acids, carbohydrates and many other compounds[46]. The protein content in leaves could have supplementary effect for the daily protein requirement of the body. The symptoms of protein energy malnutrition such as kwashiorkor and marasmus. The presence of these important nutrients like carbohydrate, protein in the leaves means that leaves could be used as a nutritionally valuable and healthy ingredient to improve poultry health and growth performance[47]. The non –essential amino acids also play a vital role in human nutrition and metabolism, especially arginine and histidine which are referred to as semi – essential amino acid, which do not actually play a role in nitrogen balance but needed for growth and development in infants[48]. Many body functions depend on lipids. Lipids provide excellent source of energy and enhance transport of fat soluble vitamins, insulate and protect internal tissues and contribute to vital cell processes[49,50,51].

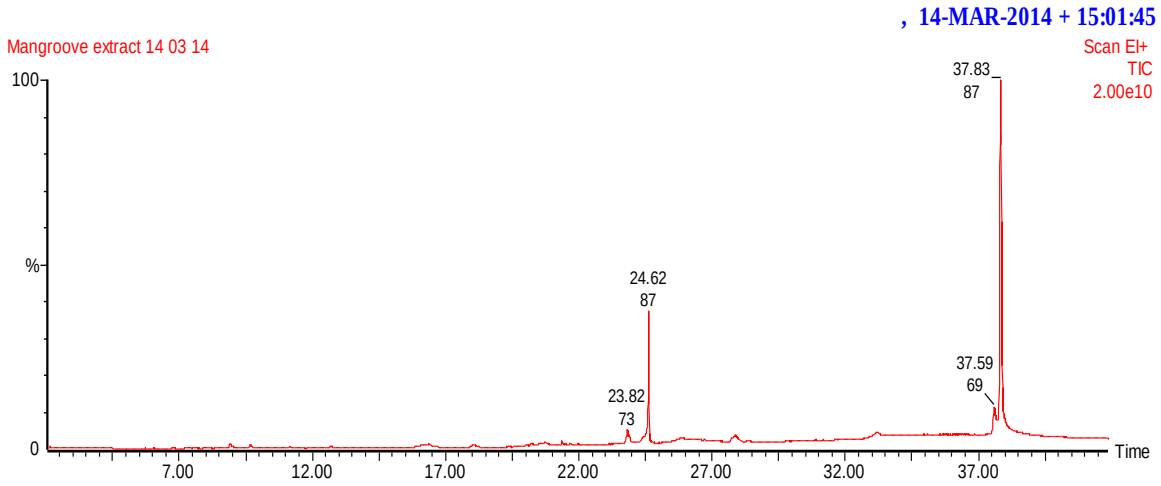


Fig. 1: Analysis of Compounds present in the Mangrove sample by GC-MS Chromatogram-GC-MS analysis shows fourteen peaks

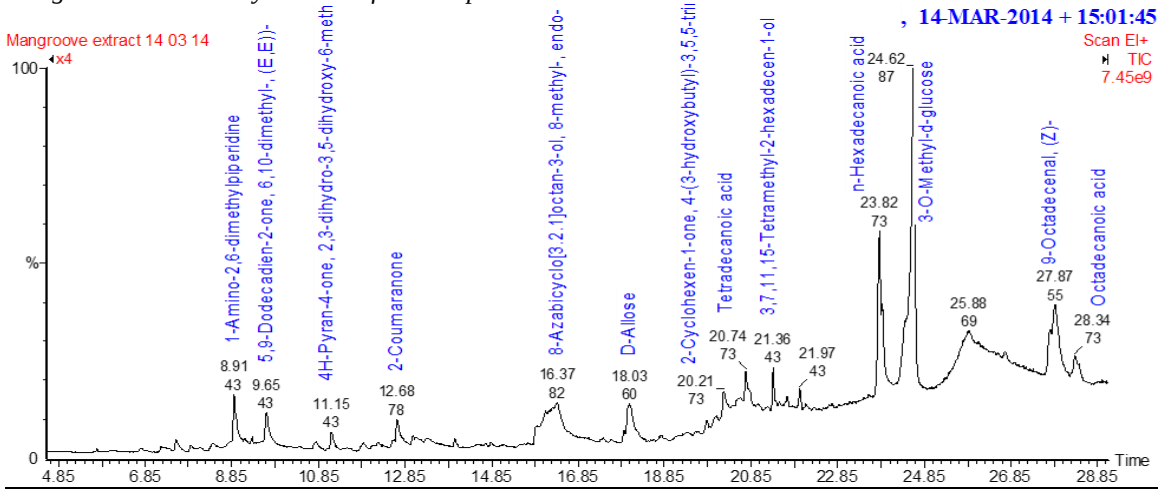


Fig-2:

Table: 4 Activity of phytocomponents identified in the ethanol leaf extract of *B. cylindrica*

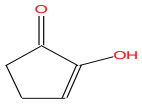
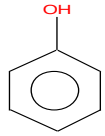
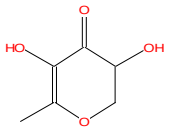
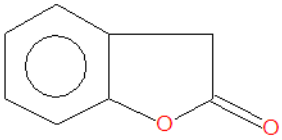
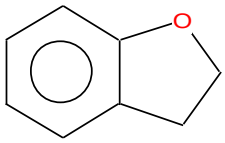
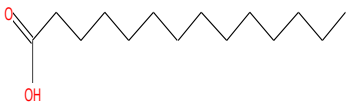
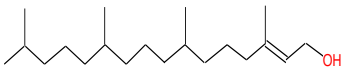
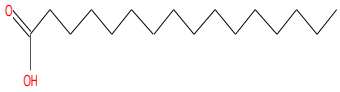
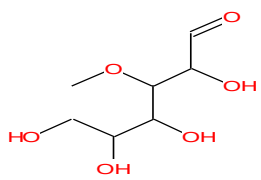
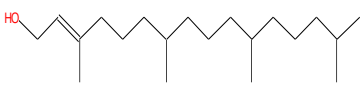
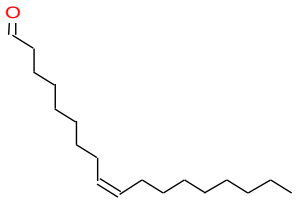
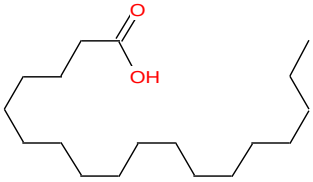
S.No	Peak Name	Nature of Compound	Reference no.	Biological activities
1	2-Cyclopenten-1-one, 2-hydroxy 	Diterpene	83	Antimicrobial, Anti-inflammatory, Anticancer, Dieruretic.
2	Phenol 	Phenol	13, 83, 84, 85	antimicrobial activity antiviral, anti-inflammatory, cytotoxic activity, antimutagenic and anticarcinogenic activities, Antioxidant function in nutrient uptake, protein synthesis, enzyme activity, photosynthesis; structural components and allelopathy in herbs,
3	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl 	Flavanoid	86, 87	Antimicrobial, Anti-inflammatory, Antiproliferative.
4	2-Coumaranone 	Coumaran compound	88	Antioxidant
5	Benzofuran, 2,3-dihydro 	Coumaran compound	89	Antimicrobial, Antiinflamatory
6	2-Methoxy-4-vinylphenol	Phenolic compound	89	Antimicrobial, Antiinflamatory Antioxidant
8	Tetradecanoic acid 	Myristic acid	89, 90	Cosmetics,Antioxidant, Cancer preventive, Nematicide, Lubricant Hypocholesterolemic
9	3,7,11,15-Tetramethyl-2-hexadecen-1-ol 	Terpene alcohol	91,92	Antimicrobial
10	n-Hexadecanoic acid 	Palmitic acid	87, 89, 91, 92, 93	Antioxidant, Hypochloesterolemic, Nematicide, Pesticide, Lubricant, Antiandrogenic, Hemolytic, 5-Alpha reductase inhibitor.
11.	 3-O-Methyl-d-glucose	Sugar moiety	83	Presevative activity

Table: 4 Activity of phytocomponents identified in the ethanol leaf extract of *B. cylindrica*

S.No	Peak Name	Nature of Compound	Reference no.	Biological activities
12	Phytol 	Diterpene	84, 85, 92	Anticancer(R26), antioxidant, antiinflammatory and diuretic
13	9,12-Octadecadienoic acid, methyl ester 	Linoleic acid Ester	89, 91, 92,93	Anti-inflammatory, Hypercholesterolemic, Cancer preventive, Hepatoprotective, Nematicide, Insectifuge, Antihistamine, Anti-eczemic, Anti-acne, 5-Alpha reductase inhibitor, Antiandrogenic, Anti-arthritic, Anti-coronary
14	9-Octadecenal, (Z)- 	Fatty acid	93	anti-alopcic, Antitumour, Cholerectic, Dermatitigenic, immunostimulant, Anti leukotriene, anti- androgenic, Haemolytic, Hypercholesterolemic, Lubricant, Nimoticide, Pesticide, irritant, Flavour, 5 α reductase inhibitor, Percutanea-stimulant, Anemiagenic
15	Octadecanoic acid 	Stearic acid	87, 92	Hypocholesterolemic
16	2-Furancarboxaldehyde, 5-methyl	Aldehyde compound	89	Antimicrobial preservative

Crude fibre in foods or plants indicates the presence of non-digestible carbohydrate and lignin[51]. Fiber content forms bulk in diet and it helps to reduce the intake of starch foods, enhances gastrointestinal function, prevents constipation and thereby reduce the development of metabolic disease like maturity onset diabetes mellitus and hypercholesterolemia. High levels of dietary fibre in leafy vegetables are advantageous for their active role in the regulation of intestinal transit, increasing dietary bulk and increasing faeces consistency due to their ability to absorb water[52]. Fiber helps in the maintenance of human health and has been known to reduce cholesterol level of the body. A low fiber diet has been associated with heart disease, cancer of the colon and rectum, varicose veins, phlebitis, obesity, appendicitis, diabetes and even constipation[53]. Studies have shown that crude fibre aids in reducing peaks of blood glucose following a meal due to delayed gastric emptying[49,51].

Medicinal plants play a vital role in preventing various diseases. Flavonoids, alkaloids, terpenoids, phlobatannins and reducing sugars were found in leaves of *Morus alba gars* and it has antidiuretic, anti-inflammatory, anti-analgesic, anticancer, anti-viral, anti-malarial, anti-bacterial and anti-fungal activities, due to the presence of the above mentioned secondary metabolites[5]. Alkaloids generally present in medicinal plants play some metabolic role and control development in living system. They are also involved in protective function of animals and are used as medicine especially the steroidal alkaloids[25]. Alkaloids widely well known to have antidiabetic[54] and antimicrobial[43] activities. Plants with alkaloids may have hypoglycemic effect via mechanism of insulin releasing and insulin-mimicking activity and thus improves postprandial hyperglycemia[55] and antimicrobial effects due to the action of intercalates into cell wall and DNA of organisms, inhibits release of autacoids and prostaglandins, possess anti-oxidating effects, thus reduces nitrate generation which is useful for protein synthesis, suppresses transfer of sucrose from stomach to small intestine, diminishing the support of glucose to the helminthes, acts on CNS causing paralysis[56].

Natural antioxidants mainly come from plants in the form of phenolic compounds, such as flavonoids, phenolic acids, tocopherols etc. The presence of flavonoids has important effects in plant biochemistry and physiology as antioxidants, enzyme inhibitors, precursors of toxic substances[57] and they are recognized to possess antidiabetic[58,59,54] antioxidant[5,60,61], antimicrobial[43,61,62,63], fungicidal, natural antihistaminic[57,61], antiinflammatory,

antiallergic and anticarcinogenic activities[57]. The antioxidative properties of flavonoids are due to several different mechanisms, such as scavenging of free radicals, chelation of metal ions, such as iron and copper and inhibition of enzymes responsible for free radical generation[64]. Flavonoids may exert beneficial effects in antimicrobial, antidiarrhoeal, anthelmintic, antidiabetic and anticancer by complex with cell wall, binds to adhesions, inhibits release of autacoids and prostaglandins, inhibits contractions caused by spasmogens, stimulates normalization of the deranged water transport across the mucosal cells, and inhibits gasterointestinal release of acetylcholine [56], and (i) enhances insulin secretion and reduces apoptosis and promotes proliferation of pancreatic β-cells; (ii) improving hyperglycemia through regulation of glucose metabolism in hepatocytes; (iii) reducing insulin resistance, inflammation and oxidative stress in muscle and fat and (iv) increasing glucose uptake in skeletal muscle and white adipose tissue[65] insulin triggering and/or insulin-like properties [66], cell growth and kinase activity inhibition, apoptosis induction, suppression of the secretion of matrix metalloproteinases and of tumor invasive behaviour respectively[67].

Previous reports revealed that mangroves are rich in polyphenols and tannins[68,69]. Several studies have described the antioxidant properties of different parts of various medicinal plants which are rich in phenolic compounds[13,60,63,70,71] and flavonoids[5,60,61]. The methanol extracts of *B.Cylindrica* bark was yielded the gallicacid equivalent/phenolic contents and flavonoids, it has been studied for its antioxidant and hypolipidemic properties[72]. Phenols are phytochemical compounds that function in nutrient uptake, protein synthesis, enzyme activity, photosynthesis; structural components and allelopathy in herbs. The phenolic compounds, have biological and pharmacological properties especially their antimicrobial activity[43,73], antiviral, anti-inflammatory, cytotoxic activity, antimutagenic and anticarcinogenic activities[59]. Many supportive reports emphasize the positive correlation between phenolic content and antioxidant efficacy[11].

Several studies have described the antidiabetic properties of flavonoids and triterpenoids are due to several different mechanisms, such as stimulate secretion or possess an insulin like-effect (Marles and Farnsworth 1995), on pancreatic β-cells leading to their proliferation and secretion of more insulin Mahesh and Menon (2004) and Sri-Balasubashini, et al., (2004) in diabetic rats[32]. The triterpenoid rich extract from the medicinal plant has both hypoglycemic and hypolipidemic effects that can not only help cure and manage diabetes but also improve insulin resistance (IR)[74]. A novel six new pentacyclic triterpenoid esters (1-6) together with 3α- and 3β-taraxerol were isolated from the fruits of *B. cylindrica*[75], another triterpenoid compound, 3.beta.,30-dihydroxylup-20(29)-en-2-one, for use as a hypoglycemic agent[76] and brugine from *B. cylindrica*[77]. Terpenoids in *Ceriopstagal Perr.* (Rhizophoraceae) exhibited antifouling activity against cyprid larvae expectorant and antitussive and anticancer[63] activities. Terpenoids also exhibit antimicrobial and antidiarrhoeal effects by membrane disruption and inhibits release of autacoids and prostaglandins[56].

Saponins have been reported to possess antidiarrheal[61] piscicidal[78] and for controlling cholesterols[79] and anticancer. Antidiarrhoeal , anticancer and anthelmintic activities may due to inhibits histamine release in vitro, possesses membrane permeabilizing properties and leads to vacuolization and disintegration of teguments respectively[56]. Tannin and steroids present in the medicinal plants are responsible for anti-inflammatory[63], antibcaterial[43,60,63,73], antioxident activities[70,63]. Tannins are astringent, bitter plant polyphenols that either bind and precipitate or shrink proteins, tannins are distributed all over the plant kingdom. Tannins have traditionally been considered antinutritional but it may be employed medicinally as antidiarrheal, hemostatic and antihemorrhoidal compounds. It presence in the plant suggest it to be of medicinal value because tannins have shown potential antiviral, antibacterial and antiparasitic effects[79]. Polyphenols and Tannins present in the medicinal plants are responsible for antimicrobial, antidiarrhoeal and anthelmintic via mechanism binds to adhesions, enzyme inhibition, substrate deprivation, complex with cell wall, membrane disruption, metal ion complexation makes intestinal mucosa more resistant and reduces secretion, stimulates normalization of deranged water transport across the mucosal cells and reduction of the intestinal transit, blocks the binding of β subunit of heat-labile enterotoxin to GM1, resulting in the suppression of heat-labile enterotoxin-induced diarrhoea, astringent action and increases supply of digestible proteins by animals by forming protein complexes in rumen, interferes with energy generation by uncoupling oxidative phosphorylation, causes a decrease in gastro intestinal metabolism[56].

Glycosides have been reported antidiarrhoeal effect may be due to the action of inhibits the release of autacoids and prostaglandins[56]. Steroidal compounds are of importance and interest in pharmacy due to their relationship with such compounds as sex hormones, they also promote immune functions in the skin and also reduce inflammation[53,80,81].

A large number of therapeutic agents in use today have been isolated or derived from plant sources[82]. The bio-activity of phytocomponents identified in the ethanol leaf extract of *B.Cylindrica* presented in Table-4[83-93]. GC-MS analysis of some mangrove plants like *B.Cylindrica*, a plant *Cleome viscosa L.* showed the presence 3-O-Methyl-d-glucose, followed by Benzofuran, 2, 3-dihydro and n-Hexadecanoic acid[94]. n-Hexadecanoic acid and phytol in *Rhizophora mucronata*[95], phytol, hexadecanoic acid, ethyl ester and 3,7,11,15-tetramethyl-2-hexadecen-1-ol compounds of active molecules were found in mangrove plants *Avicennia marina*[96] and *Finlaysonia obovata*[97] were responsible for good antibacterial properties and other biological activities.

CONCLUSION

The results evidently specifies that ethanolic leaf extract of *B.Cylindrica* contains various bioactive compounds have various medicinal properties that can be browbeaten for the

treatment of many diseases. However, isolation of individual phytochemical constituents and subjecting it to the biological activity will definitely give fruitful results. Therefore, it is recommended as a plant of phytopharmaceutical importance.

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