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Research Article

ESSENTIAL OIL EXTRACTION, CHARACTERIZATION AND ANTIMICROBIAL STUDY OF BLUMEA OXYDONTA D.C. FROM KONKAN REGION

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ABSTRACT

The present study of Essential Oil of Blumea Oxyodonta DC was designed to evaluate essential oil extraction, phytochemical composition and Anti Microbial Study of essential oil from Blumea Oxyodonta DC collected from Bhramhanwadi, Murud, in Konkan Region. The Essential Oil was extracted by Hydro distillation using Clevenger's type Apparatus and characterization of essential oil was done by IR, GC-MS and Anti Microbial Activity. Total 81 compounds were identified using Gas Chromatography with Mass Spectroscopy. The main components of essential oil are E,E,Z-1,3,12-Nonadecatriene-5,14-diol (9.11%), 1-(+)-Ascorbic Acid 2,6-dihexadecanoate (8.76%), Cyclopropanecarboxylic acid, 2-methyl-, 2,6-di-t-butyl-4-methylphenyl ester (5.89%), Phytol (5.26%), Propanoic acid, 2-methyl-, 2-[3-[(acetyloxy)methyl]oxiranyl]-5-methylphenyl ester (4.02%), 2-Pentadecanone, 6,10,14-trimethyl (3.96%), Propanoic acid, 2-methyl-, 2-[3-[(acetyloxy)methyl]oxiranyl]-5-methylphenyl ester (3.73%), 1-Methyl-1-(m-methylphenyl)-1-silacyclobutane (3.08%), etc. IR confirms the identification of components. Chemical Investigation was done to determine its molecular formula and structure.

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INTRODUCTION

Asteraceae or Compositae commonly referred as the aster, daisy, composite (1) or sunflower family. It is very large and wide spread family of flowering plants (Angiospermae) (2) (3). The family currently has 32,913 accepted species names in 1,911 genera and 13 subfamilies (4). Most members of Astreaceae are herbaceous but a significant number also shurbs, vines and trees. The species of this families are generally observed from the polar regions to the tropics. Colonizing a wide variety of habitats. It is most common in the arid and semiarid regions of subtropical and lower temperate latitude (5). Asteraceae is an economically important family providing product such as cooking oils, lettuce, sunflower, seeds, artichokes, sweetening agents, coffee substituents and herbal teas (6). The ability to utilize oxygen has provided humans with the benefit of metabolizing carbohydrates, fats and proteins for energy, however it does not come without a cost. A contradiction in metabolism is that, while the vast majority of complex life on earth requires oxygen for its existence. Oxygen is highly reactive. Atom that is capable of becoming part of potentially damaging molecules commonly called "Free Radical". Free radical are capable of attacking the healthy cells of the body causing them to lose their structure

and function (7)(8). Cell damage caused by free radicals leads to major contributor to aging and degenerative diseases such as cancer, cardiovascular diseases, immune system decline and brain dysfunction (7)(9). Overall free radicals have been implicated in the pathogenesis of at least 50 diseases (7,10,11). Almost all studies on essential oil research focus on their extraction, chemical composition and wide application in the food and cosmetic industries and traditional medicines (12,13). Therefore it is often believed that essential oils are completely safe as they are natural in origin. Essential Oils are rich blend of highly concentrated, volatile and fat soluble in nature. Therefore mainly differ from the water soluble whole herb extracts used in herbal medicines (14). The toxicity of essential oils can also be entirely different to that of the herb as they are lipophilic in nature and hence can pass across the membranes very efficiently (15). As these properties are beneficial for their medicinal effects, this may also lead to their toxicity. Some of the major areas of concern about essential oil hazards include allergic contact dermatitis, photosensitization, neurotoxicity, carcinogenicity (16). Hence it is very important to study interaction of essential oils and their constituents in vivo to know their efficiency as well as toxicity. As the fragrance of essential oils are complicated and difficult to characterize, there are very few reports available about their in vivo interactions in

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body fluids (17,18,19). Therefore it is more important to find out right methodology for identification of constituents of essential oil from easily available aromatic plants.

Asteraceae family consists of genera *Blumea* which is vast in its species. From these *Blumea* species, *Blumea Oxyodonta* D.C. is collected first time from the konkon Region. To find out phytochemical constituents present in this plant with the help of Anti Microbial Study of essential oil obtained by different extraction processes.

Antimicrobial resistance is a major global problem with resistant strains of *Staphylococcus aureus* (18) and *Pseudomonas Aeruginosa* (19) and other micro organisms being responsible for much morbidity and mortality.

Medicinal Plants have the ability to inhibit the growth of wide range of pathogenic micro organisms due to presence of essential oil (20). Essential oils are natural, volatile liquid, complex compound characterized by strong odor, rarely colored, soluble in lipids and organic solvents. It could be synthesized by all plant organs i.e. buds, flowers, leaves, stems, twigs, seeds, fruits, roots, wood or bark and are stored in secretory cells, cavities, canals, epidermic cells or glandular trichomes (21).

The Konkan region is rich biodiversity. *Blumea Oxyodonta* D.C. is easily available species in paddy fields in Konkan Region. In India no one done work on *Blumea Oxyodonta* D.C. This is the first research article which is contributing in an identification of phytochemical constituent present in *Blumea Oxyodonta* D.C. Essential Oil generally contains terpenes, sesquiterpenes, alkaloids, flavonoids, etc. Essential Oil of *Blumea Oxyodonta* D.C. consisting of total 81 components which are identified by GC-MS. This indicates how much this species is showing variety in constituents. Antimicrobial study of essential oil of this species shows very efficient against *Staphylococcus Aureus* and *Pseudomonas Aeruginosa* bacteria's. It means that essential oil of *Blumea Oxyodonta* D.C. inhibits the growth of these microorganisms very efficiently. Essential Oil of this species is an alternative to the diseases caused by micro organisms.

Most importantly *Blumea Oxyodonta* D.C. is easily available and people around here throw away this species but they don't know the medicinal use of species. This research article showcasing the medicinal use of *Blumea Oxyodonta* D.C. in the Konkan region.

About *Blumea Oxyodonta* D.C

Asteraceae is a very large and widespread family of flowering plants. The family has 32,913 accepted species names in 1911 genera, 13 subfamilies. *Blumea Oxyodonta* D.C. is a very common weed in India. *Blumea Oxyodonta* D.C. is an annual herb having a strong odor like turpentine. In an Indian System of Traditional Medicines i.e. Ayurveda, *Blumea Oxyodonta* D.C. is used as bitter, astringent, acrid, thermogenic, emmenagogue, anti-inflammatory, styptic, ophthalmic, digestive, anthelmintic, liver tonic, expectorant, febrifuge, antipyretic, diuretic, deobstruent and stimulant⁽²⁾. Taxonomy of Genus *Blumea* L. is as follows:

Kingdom: Plantae
Order: Asterales

Family: Asteraceae
Tribe: Astereae
Genus: *Blumea*

Blumea Oxyodonta D.C. belongs to the genus *Blumea*. *Blumea Oxyodonta* D.C. in Marathi is commonly known as a Bhamurda. *Blumea Oxyodonta* D.C. is commonly called as a spiny leaved *blumea*. *Blumea Oxyodonta* D.C. is an annual erect herb with a slender whitish hairy stem, which is often forked. The branches are spreading or prostrate. Alternately arranged obovate leaves, 5-5.5 X 1.5-2 cm, have spinous toothed margin, and a spiny tip. Leaf stalks are up to 1 cm long. Leaves on branches 1 X 0.5 cm, nearly stalkless, densely white woolly. Yellowish flower heads, to 6 mm across, on long peduncles, arise in leaf axils, either solitary or in corymb-like cymes. Flowering period of *Blumea Oxyodonta* D.C. starts from January.

MATERIAL AND METHODS

Plant Material

The entire plant including leaves, stem, aerial part, flowers of *Blumea Oxyodonta* D.C. were collected from the paddy fields of Bhamanwadi, Murud, Dapoli region, Maharashtra, India between the months of January to May. After collection of *Blumea Oxyodonta* D.C. particular species was submitted to The Botanical Survey of India Western Region Pune Maharashtra for identification and certification. They have certified and identified this species as *Blumea Oxyodonta* D.C.

Essential oil Extraction

Essential oil extraction of *Blumea Oxyodonta* D.C. was carried out by hydro distillation by using Clevenger's apparatus as well as steam distillation.

Essential Oil Extraction by Hydro Distillation with Clevenger's Apparatus Method

Essential oil extraction of *Blumea Oxyodonta* D.C. was carried out with hydro distillation by using Clevenger's Apparatus Method. The fresh plant material including aerial part, stem, leaves and flowers gets chopped into small pieces. 50 gm of fresh plant material was subjected to hydro distillation using Clevenger type apparatus of capacity 1 liter. Only 70 ml of water was added just to wet the fresh plant material. The mixture was heated on heating mantle at 85^o C. The distillation was continued for about 3 hours. As the essential oil obtained was in very less quantity i.e. 0.1 ml that's why I have to carry out the hydro distillation process number of times till I get the desired quantity for characterization of essential oil. After obtaining desired quantity it was dried over by anhydrous sodium sulphate and stored in sealed vials in refrigerator until analysis.

Essential Oil Extraction by Steam Distillation Method

Essential oil extraction of *Blumea Oxyodonta* D.C. was carried out with hydro distillation by using Clevenger's Apparatus Method. The fresh plant material including aerial part, stem, leaves and flowers gets chopped into small pieces. 50 gm of fresh plant material was subjected to steam distillation using steam distillation apparatus of capacity 1 liter. 400 ml of water was added to develop vapors to pass from the fresh plant material. The water was heated on heating mantle at 85^o C. The distillation was continued for about 3

hours. As the essential oil obtained was in quantity i.e. 0.12 ml that's why I have to carried out the Steam distillation process number of times till I get the desired quantity for characterization of essential oil. After obtaining desired quantity it was dried over by anhydrous sodium sulphate and stored in sealed vials in refrigerator until analysis.

Characterization of Essential Oil

Essential Oil characterization of BlumeaLacera DC was done by the techniques like Gas Chromatography with Mass Spectroscopy, Infrared Spectroscopy, Nuclear Magnetic Resonance Spectroscopy. Procedures for these techniques are given as follows:

Infrared Spectroscopy

Infrared Spectroscopy (Figure 1) gives information on the vibrational and rotational modes of motion of a molecule and hence an important technique for identification and characterization of a functional group. The infrared spectrum of an organic compounds a unique fingerprint which is readily distinguished from the absorption patterns of all other compounds. An IR analysis was accomplished using Bruker, 3000 Hyperion Microscope with vertex 80 FTIR system equipped with focal plane array of 128 X 128 and ranges from 4000 – 900 cm⁻¹. It does have single point detector ranging from 7500 – 450 cm⁻¹. It is having analysis area 128 X 128 in 2D format on the sample plane 300 X 300 μm. This instrument is having spatial resolution with 15 times objective is 2.7 μm, temperature controlled sample stage and spectral resolution of FTIR is 0.2 cm⁻¹. (Figure No. 1)

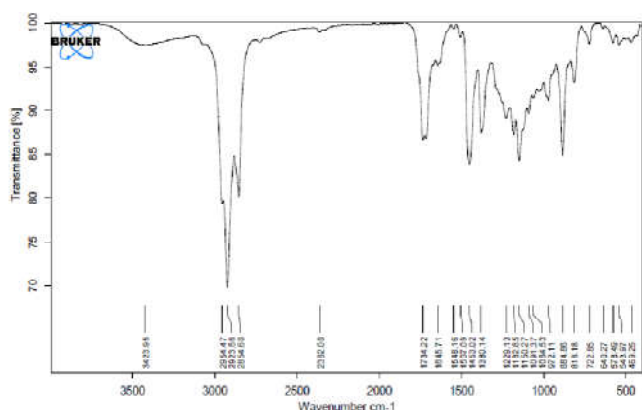


Figure 1 IR Spectra of Essential Oil

Gas Chromatography

Initially GC was used for development of chromatographic method for the selected plant essential oil. The GC analysis was accomplished using Shimadzu GCMS-QP 2010 Ultra gas chromatograph equipped with FID and Rtx®-5 MS capillary column (0.25mm X 30m X 0.25 μm film thicknesses). Following temperature program was optimized for analysis (Table 1)

Table 1 The optimized temperature program for column oven

Minutes	Rate	Final Temperature (0 ^o C)	Hold Time (minute)
0	-----	70.0	0.00
1	20.00	200.0	3.00
2	25.00	300.0	2.00
3	0.00	0.0	0.00
Total Program Time			15.50 minutes

Injector temperature was 240^oC while detector temperature was 225^oC. Helium was used as a carrier gas, at a flow rate 1.53 cm³/ min. Split ratio was 1:25.

Gas Chromatography-Mass Spectrometry (GC-MS) Analysis

The GC method was then transferred to GC-MS with slight modifications for identification of various phytoconstituents of selected plant essential oil. The oil was analyzed by Shimadzu GCMS-QP 2010 Ultrasystem. The system was equipped with fused silica Rtx-1Sil MS silarylene capillary column with dimensions 30m X 0.25mm X 0.25μm. Helium (0.93 ml/min) was used as a carrier gas. The program used for GC oven temperature was 1 minute isothermal at 50^oC, followed by 50-220^oC at a rate of 50^oC/min, then held at 220^oC for 1minute, followed by 220 - 260^oC at a rate of 200C/min, then again held at 260^oC for 15 minutes. The injection port temperature was 266^oC. The ionization of sample components was performed in the E.I. mode (70eV). The Linear Retention Indices (LRI) for all the compounds was determined by co-injection of the sample with a solution containing the homologous series of C8-C29 n-alkanes. Individual constituents were identified by referring to compounds known in the literature data and also by comparing their mass spectra with known compounds and NIST Mass Spectral Library (NIST 05). (Figure 2)

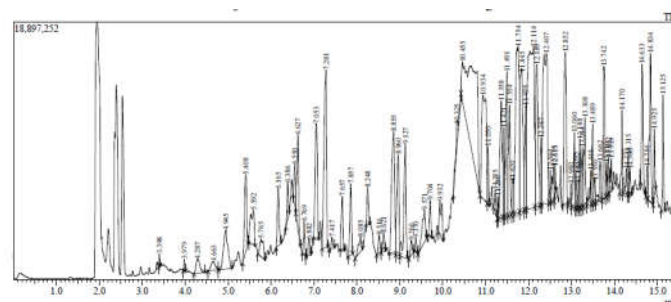


Figure 2 GC-MS of Essential Oil

Chemical Components of Essential Oil of Blumea Oxyodonta Dc (Table No 2)

Table No. 2 Chemical Components of Essential Oil of Blumea Oxyodonta D.C.

Relative Retention Indices	Relative concentration of components in Area Percentage (%)	Name of Compound
1.940	0.08	Toluene
2.215	0.08	Cyclopentane, 1-ethyl-3-methyl-, trans-
2.395	0.08	o-Xylene
2.540	0.08	m-Xylene
3.395	0.08	Santolina Epoxide
3.980	0.08	Benzaldehyde dimethyl acetal
4.285	0.44	1H-Indene, 2,3,3a,4,7,7a-hexahydro-2,2,4,4,7,7-hexamethyl-
4.650	0.31	10-12-Pentacosadiynoic acid
4.935	1.44	Aromadendrene oxide-(2)
5.410	1.48	Z-5,17-Octadecadien-1-ol acetate
5.600	1.08	Butyl 6,9,12,15-octadecatetraenoate
5.775	0.53	3a,6-Methano-3ah-inden-5-ol, octahydro-, (3a.alpha.,5.alpha.,6.alpha.,7a.beta.)-
6.170	0.91	1,5,5-Trimethyl-6-(3-methyl-buta-1,3-dienyl)-cyclohexene
6.380	0.44	Ethyl iso-allocholate
6.545	0.48	Eicosanoic acid
6.630	1.37	Methyl 3,5-tetradecadiynoate
6.770	0.39	1,5,5-Trimethyl-6-(3-methyl-buta-1,3-dienyl)-cyclohexene
6.885	0.23	3.beta.-Hydroxyguaia-4(15),10(14),11(13)-

		trien-6,12-olide 8-(.alpha.,.beta.-dihydroxybutyrate)
7.050	2.21	3-Methyl-2-butenic acid, 2,7-dimethyloct-7-en-5-yn-4-yl ester
7.270	3.96	2-Pentadecanone, 6,10,14-trimethyl-
7.425	0.11	1-Heptatriacotanol
7.655	0.79	4,5-Dihydro-5,5,7-trimethyl-6H-[1,2,5]oxadiazolo[3,4-b][1,4]diazepine
7.860	1.00	3-Methyl-2-butenic acid, 2,7-dimethyloct-7-en-5-yn-4-yl ester
8.080	0.34	cis-1-Chloro-9-octadecene
8.255	0.47	Strophanthidin
8.525	0.24	Ethyl iso-allocholate
8.625	0.26	Bicyclo[3.1.1]hept-3-ene, 2-formylmethyl-4,6,6-trimethyl-
8.850	3.08	1-Methyl-1-(m-methylphenyl)-1-silacyclobutane
8.955	1.88	1-(4-Isopropylphenyl)-3-(tetrahydrofuryl-2)propane
9.120	2.22	Hexadecanoic acid, methyl ester
9.265	0.23	1H-Benzocyclohepten-7-ol, 2,3,4,4a,5,6,7,8-octahydro-1,1,4a,7-tetramethyl-, cis-1,5,5-Trimethyl-6-(3-methyl-buta-1,3-dienyl)-cyclohexene
9.375	0.17	Isophytol
9.580	0.86	1,5,5-Trimethyl-6-(3-methyl-buta-1,3-dienyl)-cyclohexene
9.705	0.08	1H-Benzocyclohepten-7-ol, 2,3,4,4a,5,6,7,8-octahydro-1,1,4a,7-tetramethyl-, cis-1-(+)-Ascorbic acid 2,6-dihexadecanoate
10.340	0.48	1-(+)-Ascorbic acid 2,6-dihexadecanoate
10.465	8.76	Propanoic acid, 2-methyl-, 2-[3-[(acetyloxy)methyl]oxiranyl]-5-methylphenyl ester
10.935	4.02	2,6,10,14-Hexadecatetraen-1-ol, 3,7,11,15-tetramethyl-, acetate, (E,E,E)
11.050	0.52	Ethyl iso-allocholate
11.220	0.26	Ethyl iso-allocholate
11.275	0.17	Acetic acid, 3,7,11,15-tetramethyl-hexadecyl ester
11.360	2.23	1-(+)-Ascorbic acid 2,6-dihexadecanoate
11.420	1.02	2-Methylbut-2-enoic acid, 3-hydroxy-3-isopropyl-6,8a-dimethyl-8-oxo-1,2,3,3a,4,5,8,8a-octahydro-azulen-4-yl ester
11.485	2.34	Butyl 9,12,15-octadecatrienoate
11.555	1.51	(2,2,6-Trimethyl-bicyclo[4.1.0]hept-1-yl)-methanol
11.625	0.58	Phytol
11.725	5.26	Propanoic acid, 2-methyl-, 2-[3-[(acetyloxy)methyl]oxiranyl]-5-methylphenyl ester
11.830	3.73	2-Dodecen-1-yl(-)succinic anhydride
11.925	1.43	Furandione, 3-dodecenyl-
12.110	9.11	E,E,Z-1,3,12-Nonadecatriene-5,14-diol
12.190	2.96	Octadecanoic acid
12.280	0.64	1,1,6-trimethyl-3-methylene-2-(3,6,9,13-tetramethyl-6-ethenyl)-10,14-dimethylene-pentadec-4-enyl)cyclohexane
12.360	5.89	Cyclopropanecarboxylic acid, 2-methyl-, 2,6-di-t-butyl-4-methylphenyl ester
12.500	0.33	Ethyl iso-allocholate
12.610	0.19	2-Butenoic acid, 2-methyl-, 2-(acetyloxy)-1,1a,2,3,4,6,7,10,11,11a-decahydro-7,10-dihydroxy-1,1,3,6,9-pentamethyl-4a,7a-epoxy-5H-cyclopenta
12.735	2.85	2-Methylbut-2-enoic acid, 3-hydroxy-3-isopropyl-6,8a-dimethyl-8-oxo-1,2,3,3a,4,5,8,8a-octahydro-azulen-4-yl ester
12.855	2.85	Cyclopropanecarboxylic acid, 2-methyl-, 2,6-di-t-butyl-4-methylphenyl ester
12.980	0.24	1-Heptatriacotanol
13.065	0.82	Phytol, acetate
13.095	0.26	1-Heptatriacotanol
13.160	0.27	1-Heptatriacotanol
13.185	0.61	2H-Pyran-2-one, tetrahydro-6-tridecyl-
13.245	0.72	Hexadecanoic acid, cyclohexyl ester
13.305	1.11	Octadecane, 1-chloro-

13.490	0.88	1-Heptatriacotanol
13.540	0.31	1-Heptatriacotanol
13.670	0.54	1-Heptatriacotanol
13.745	1.83	Tetrapentacontane
13.805	0.26	Retinal
13.860	0.29	9,19-Cyclolanostan-3-ol, acetate, (3.beta.)-
13.920	0.42	Epiglobulol
14.170	0.81	Octadecane, 1-chloro-
14.270	0.35	9,19-Cyclolanostan-3-ol, acetate, (3.beta.)-
14.310	0.43	9,19-Cyclolanostan-3-ol, acetate, (3.beta.)-
14.360	0.23	1-Heptatriacotanol
14.630	1.72	Tetrapentacontane
14.755	0.27	3,9-Dimethyltricyclo[4.2.1.1(2,5)]decan-9-ol
14.830	2.16	Pentanoic acid, 1,7,7-trimethylbicyclo[2.2.1]hept-2-yl ester, endo-Nerolidyl propionate
14.925	0.54	Tetrapentacontane
15.120	1.09	Tetrapentacontane
15.460	0.23	Viridiflorol

Antimicrobial Activity

Anti-Microbial Activity (Table 3) of Essential Oil from *Blumea oxyodonta* DC was done against five microorganisms. Out of these five microorganisms three microorganisms were Gram negative and remaining two were Gram Positive Bacteria. Agar Cup Method was used to evaluate the results of Anti-Microbial Activity. In this test streptomycin solution having concentration 30 µg/ml was used as a standard against extracted essential oil. Volume for Anti-Microbial Activity Standard Solution and Essential Oil 20 µl/well was used. Anti-microbial activity of Essential oil of *Blumea Oxiodonta* DC was checked by making two different concentrations like 1:4 (Essential Oil : Acetone) diluted concentration of essential oil, undiluted (crude) concentration of essential oil. Concentration of this two samples of essential oil was checked against standard named as streptomycin (20 µg/ml) and pure acetone solution. Results of Inhibition Zones obtained from both were as follows:

Table 3 Inhibition Zones (mm)

Cultures	Gram Character	Diameter of Inhibition Zone (mm)			
		1:4 Diluted (Essential Oil: Acetone)	Undiluted Essential Oil (20 µl/well)	Acetone Control (Pure Solution of Acetone)	Streptomycin (20 µg/ml)
Escherichia Coli		---	---	---	21
Pseudomonas Aeruginosa	Gram Negative	12	17	00	07
Salmonella Typhi		---	---	---	17
Staphylococcus Aureus	Gram Positive	00	26	00	08
Corynebacterium Diphtheriae		---	---	---	19

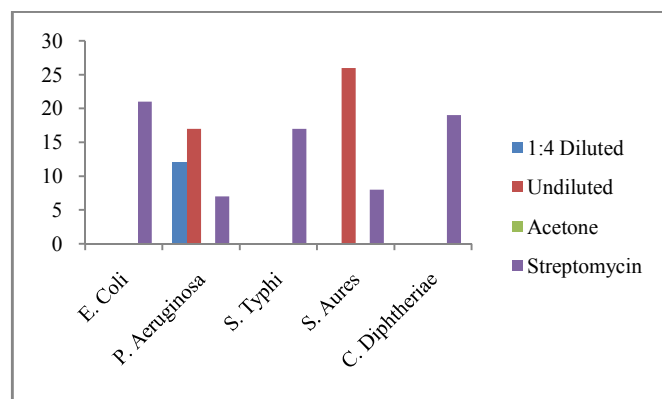


Figure 3 Graph of Inhibition Zones in mm against Cultures of Micro organisms

RESULT AND DISCUSSION

India is one of the richest sources of medicinal plants species which grows in tropical, subtropical regions. But here we are discussing such a type of species which is widely available yet to be ignored till date. We are discussing about the *Blumea Oxyodonta* DC. This species has a flowering period from December to May. *Blumea Oxyodonta* DC is an aromatic plant. It has sweet smell and it shows some therapeutic values through its Antimicrobial study.

The hydro distillation using Clevenger's apparatus in which 100 gm chopped plant material yields 0.10% of brown color essential oil with a sweet smell. In case of steam distillation yield obtained was more than hydro distillation i.e. 0.19%. The oil sample was analyzed by Gas Chromatography with Mass Spectroscopy (GC-MS) and Infrared Spectroscopy (IR) the components were identified on the basis of their Retention Index Values and by comparison of their mass spectra with those reported in literature. The GC-MS analysis of *Blumea Oxyodonta* DC essential oil shows total 81 components were identified shown in Table No. 1. There was presence of Sesquiterpene alcohol, lactones, monoterpenes, diterpenes, triterpenes, terpenoids. The main components of essential oil are E,E,Z-1,3,12-Nonadecatriene-5,14-diol (9.11%), 1-(+)-Ascorbic Acid 2,6-dihexadecanoate (8.76%), Cyclopropanecarboxylic acid, 2-methyl-, 2,6-di-t-butyl-4-methylphenyl ester (5.89%), Phytol (5.26%), Propanoic acid, 2-methyl-, 2-[3-[(acetyloxy)methyl]oxiranyl]-5-methylphenyl ester (4.02%), 2-Pentadecanone, 6,10,14-trimethyl (3.96%), Propanoic acid, 2-methyl-, 2-[3-[(acetyloxy)methyl]oxiranyl]-5-methylphenyl ester (3.73%), 1-Methyl-1-(m-methylphenyl)-1-silacyclobutane (3.08%), etc.

Fourier Transform Infrared Spectroscopy (FT-IR) shows presence of functional groups which were identified in Gas Chromatography with Mass Spectroscopy (GC-MS). FTIR shows various stretching such as 3423.95 cm^{-1} indicates alcohol stretching, 2925 cm^{-1} indicates C-H stretching, 2726 cm^{-1} indicates C-H stretching, $1714-1646\text{ cm}^{-1}$ indicates fingerprint region in which compounds like ketones, esters, etc. were present. The initial spectra exhibit three main peaks in this range: 2954.47 , 2923.56 and 2854.60 cm^{-1} . The 2954.47 and 2923.56 cm^{-1} can be assigned to the anti-symmetric stretching modes of the CH_3 and CH_2 groups. The 2923.56 cm^{-1} is due to corresponding symmetric stretching. The behavior of Essential Oil in IR Spectra indicates a decrease in number of CH_2 groups. There is a dissociation of chains of fatty acid. Further support of this hypothesis negative contribution about fingerprint region in IR spectra where C=O stretching is observed. The spectra suggest that these groups are increasing during aging. This could interpret that there is possible esterification reaction takes place.

Anti-microbial activity of Essential oil is performed against 5 microorganisms *Escherichia Coli*, *Pseudomonas Aeruginosa*, *Salmonella Typhi*, *Staphylococcus Aureus*, *Corynebacterium Diphtheriae*. Out of these five cultures of microorganisms three were gram negative i.e. *Escherichia Coli*, *Salmonella Typhi* and two were gram positive i.e. *Staphylococcus Aureus*, *Corynebacterium Diphtheriae*. An Anti-Microbial Activity was done against the standard solution of streptomycin having concentration of $30\text{ }\mu\text{g/ml}$. Out of these 5 cultures of Micro

Organisms Essential oil showed very good results of inhibition zones for *Pseudomonas Aeruginosa*, *Staphylococcus Aureus*. The most important that Inhibition zones of essential oil were more than the standard streptomycin solution as well as acetone solution both having volume of $20\text{ }\mu\text{l/ml}$. for these two microorganisms. *Staphylococcus Aureus* causes range of illness from minor skin infections, such as pimples, impetigo, boils, cellulitis, folliculitis, carbuncles, scalded skin syndrome and abscesses, to life threatening diseases such as pneumonia, meningitis, osteomyelitis, endocarditis, toxic shock syndrome, bacteremia and sepsis. *Pseudomonas Aeruginosa* causes a diseases like urinary tract infections, respiratory system infections, dermatitis, soft tissue infections, bacteremia, bone and joint infections, gastrointestinal infections and a variety of systemic infections, particularly in patients with severe burns and in cancer and AIDS patients who are immunosuppressed. As an essential oil of *Blumea Oxyodonta* DC shows very good results than standard streptomycin solution against these microorganisms i.e. *Pseudomonas Aeruginosa*, *Staphylococcus Aureus*. This is proposed that essential oil of *Blumea Oxyodonta* DC restrict the growth of diseases caused by *Pseudomonas Aeruginosa*, *Staphylococcus Aureus*.

Therefore, our results revealed the importance of plant extracts when associated with antibiotics, to control resistant bacteria, which are becoming a threat to human health. Furthermore, in a few cases, these plant extracts were active against antibiotic resistant bacteria under very low concentration, thus minimizing the possible toxic effects.

CONCLUSION

Analytical Method has been used for the identification of phytoconstituents of essential oil of *Blumea Oxyodonta* DC from *Astreascea* family. This methodology includes GC-MS, FT-IR for the identification of volatile phytoconstituents. Mass Spectroscopy has been used for the exact mass measurement with identification of phytoconstituents. Anti-Microbial Activity has been used to evaluate the therapeutic use of this essential oil from *Blumea Oxyodonta* DC against standard streptomycin solution. Specifically yield given by dry plant material is much more than yield given by the fresh plant material. This is the most important observation by experimentation. Essential Oil of *Blumea Oxyodonta* DC has great potential as anti-microbial compounds against these microorganisms. Thus they can be used in the treatment of infectious diseases caused by resistant microbes. Most importantly *Blumea Oxyodonta* DC is easily available in konkan region but this is the first research article showcasing the importance of this species.

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