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A STUDY ON PHYTOCHEMICAL ANALYSIS OF SOME AROMATIC MEDICINAL FLORA

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ABSTRACT

Aromatic Medicinal plants have been very popular from time immemorial since many more years in the Indian culinary, having antimicrobial and insect repellent properties as well. All these plants were biologically active and were used as traditional medicine and their essential oil also being used as perfumery ingredients and aroma enhancers in food products. In the present investigation, five aromatic medicinal plants including *Cymbopogon, Lantana, Mentha, Murraya, Ocimum* which are available in J.M. Patel College Botanical Garden were used. The main objectives of this research was to determine some bioactive phytochemicals like alkaloids, steroids, phenols, terpenoids, flavonoids, seponins, tannins, glycosides and essential oils in dried leaves of five species by following Soxhlet extraction method. Keeping in view their importance, this work was carried out to investigate the qualitative determination of different types of secondary metabolites and their role in the aroma of these aromatic plants was also discussed in the present study.

KEYWORDS: Aromatic plants, Insect repellent, Phytochemicals, Secondary metabolites, Soxhlet.

INTRODUCTION

The aromatic medicinal plants are a group of unique plants containing certain chemicals having antimicrobial and insect repellent properties and these chemicals acts as biocides. The aromatic plants and their secondary metabolites take part in increasingly in Agriculture, cosmetics and pharmaceutical products. Recently plant protection measures have been developed by the use of safer biocides which are environmentally and ecologically safe and could be exploited for commercialization.

These plants which possess specific odour due to the presence of some special types of secondary metabolites such as essential oils which are concentrated with volatile aromatic compounds like terpenoids, benzenoids - the easily evaporated essences that give plants their wonderful scents (aroma). Each of these complex precious liquids is extracted from a particular part of plant life. Essential oils are generally fragrant volatile materials consisting of complex mixtures of terpene hydrocarbons and oxygenated materials biogenically derived from them hence essential oils are used in flavorings, perfumes, in Aromatherapy, as insect & animal repellents, in pharmaceutical preparations, as anti-microbial agents and in many other ways. These phytochemicals are the products of plant metabolism, mainly used by the plants for their defense.

The present study regarding the separation of few classes of aromatic chemical compounds such as terpenoids, phenols, tannins, flavonoids and essential oils in addition to other secondary metabolites such as alkaloids, steroids, glycosides etc. were extracted from the leaves of five aromatic plants to the study of preliminary phytochemical analysis. Hence attempts have been made to use them for therapeutic purposes in future research.

MATERIAL AND METHODOLOGY

Fresh leaves of five aromatic medicinal plants *Cymbopogon, Lantana, Mentha, Murraya* and *Ocimum*, were collected from Botanical Garden of J.M. Patel College, Bhandara (Fig.1). The plant materials were taxonomically identified and authenticated by Flora of Maharashtra -Singh and Karthikeyan (2000), Singh et.al., 2000. The plant materials were washed in running water and shade dried until all the water molecules evaporated and plants material became well dried for grinding. After drying, the plant materials were ground well using mechanical blender into fine powder and transferred into airtight containers with proper labeling for future use.

The types of volatile isolates that are obtained commercially from aromatic plants are essential oils along with other secondary metabolites. Essential oils and other secondary metabolites are isolated from plant material by different solvents.

Preparation of plant extracts by Solvent extraction method

Crude plant extract was prepared by Soxhlet extraction method. About 15 gm of powdered plant material was uniformly packed into a thimble and extracted with 250 ml of different solvents separately. Solvents used were methanol, ethanol, and distilled water. The process of extraction continues for 24 hours or till the solvent in siphon tube of an extractor become colorless. After that the extract was taken in a beaker and kept on hot plate and heated at 30-40ºC till all the solvent got evaporated. Dried extract was kept in refrigerator at 4ºC for their future use in phytochemical analysis.

The extract was tested for the presence of bioactive compounds by using following different standard methods.

Each extract was tested for presence of alkaloids, flavonoids, saponines, steroids, terpenoids and tannins using different methods. The tests were performed in triplicates to ensure the accurate results (Fig. 2).

Alkaloids

The occurrence of alkaloids was tested using method reported by Sabri *et al.,* (2012). 10 ml of the extract were evaporated to dryness. Two ml of 2% HCL acid were added to the dry residue.

Few drops of Wagner's reagent were added to the solution. The presence of alkaloids was confirmed when reddish brown precipitate occurred.

Flavonoids

The test was based on method described by Pamar *et al.,* (2012). Few drops of NaOH were added to two ml of the extract and intense yellow color appeared. Few drops of dilute HCL were added and the solution turned to colorless as indicator of presence of flavonoids.

Saponins

Precipitation and foam test method was used as described by Devmurari, (2010). Few drops of (1%) lead acetate solution were added to one ml of the extracts. Intense white precipitate appeared due to presence of Saponins.

Tannins

The test method was described by Ugochuhwu *et al.,* (2013). One ml of 3% of Ferric chloride was added to one ml of the extract. Brownish green color development indicated presence of tannins.

Glycosides

To 2 ml of plant extract, 1 ml of glacial acetic acid and 5% ferric chloride was added. To these, 3 drops of concentrated sulphuric acid was added. Presence of greenish blue colour indicates the presence of glycosides.

Terpenoids

2 mL of extracts was treated with 2 mL of chloroform and concentrated sulphuric acid was carefully added to form a layer. A reddish brown colour formation at the interface confirms the presence of terpenoids.

Phenols

To 1ml of the extract, 2 ml of distilled water followed by 5 drops of 10% ferric chloride was added. Formation of blue or green colour indicates presence of phenols.

Steroids

The test was performed based on method described by Solihah *et al.,* (2012). Two ml concentrated sulphuric acid were added to two ml of the extract. Formation of red precipitate indicated presence of steroids.

Essential oils: Pure essential oils are mixtures of more than 200 components, normally mixtures of terpenes or phenylpropanic derivatives.

Extraction of Essential oils by distillation: A hydrocarbon solvent is mixed with the plant material to help dissolve the essential oil. When the solution is filtered and concentrated by distillation. Finally pure alcohol is used to extract the oil. When the alcohol evaporates, the oil is left behind. Harborne J.B. (1973).

RESULT AND DISCUSSION

I. In the present investigation, locally available five aromatic plants were selected and studied the morphological and ethno botanical characters and enumerated as follows for further phytochemical analysis.

Botanical Name: *Lantana camara* L.
Common Name: Tantani
Family: Verbenaceae

Lantana camara is a perennial, erect sprawling shrub which typically grows to around 2 m tall. The stems and branches are angular, bearing curved spines, arranged along the edges. The leaves are broadly ovate, opposite, and simple and have a strong odour when crushed dense thickets in a variety. It has small tubular shaped flowers, which each have four petals and are arranged in clusters in terminal are as of the stems. Flowers come in many different colours, including red, yellow, white, pink and orange, which differ depending on location and age. Its leaves exhibit antimicrobial, antifungal and insecticidal activity. It is used for treating various disease such as tumor, cancer, ulcer, eczema, fever, cold, asthma etc.

Botanical Name: *Ocimum sanctum* L.
Common Name: Tulsi
Family: Lamiaceae

Ocimum sanctum is a perennial herb with a height of 2-3 feet. Leaves have petiole and are ovate, up to 2-5 cm long, usually somewhat toothed, oblong or elliptic oblong, obtuse or acute, entire or subserrate, hairy on both surfaces and minutely dotted. Racemes slender, 5 to 10 cm long, bracts not exceeding the calyx, broadly ovate or cordate-ovate, acuminate, ciliate; pedicles slender, as long as or longer than the calyx, 4 mm long, purplish-pink. Plant is used for treatment of common cold, cough, tonsilites, asthema, diabetes, earche and in preparation of many Ayurvedic formulations. The leaf oil is used in dental cream, mouth wash and toothpaste. It is also possess insecticidal, antibacterial and mosquito repellent properties.

Botanical Name: *Murraya koenigii* (L.) Sprengel.
Common Name: Meetha Neem
Family: Rutaceae

It is a small tree, growing 4–6 metres tall, with a trunk up to 40 cm diameter. The aromatic leaves are pinnately compound. The plant produces small white flowers which can self-pollinate to produce small shiny-black drupes containing a single, large viable seed. The berry pulp is edible, with a sweet flavor The fresh leaves are an indispensable part of Indian traditional medicines. They are most widely used in southern and west coast Indian cooking to make chutney, rasam, kadhi etc.

Botanical Name: *Cymbopogon citratus* (DC.) Stapf.
Common Name: Olecha (Lemon Grass)
Family: Poaceae

Cymbopogon citratus is part of the grass family, contain simple, bluish-green leaves with entire margins and are linear in shape. The blades tend to be 18–36 inches long. Like other grasses, the leaves also have parallel venation. The leaves of *Cymbopogon citratus* have been used in traditional medicine and are often found in herbal supplements and teas. Citronellal is an essential oil constituent from *Cymbopogon* was used as a remedy for the treatment of oral thrush in HIV/AIDS patients and acts as antioxidant, antibacterial, and antifungal agent.

Botanical Name: *Mentha spikata* L.
Common Name: Pudina
Family: Lamiaceae

Mint is an aromatic perennial herb. It has wide-spreading underground and over ground stolons and erect, square, branched stems. The leaves are arranged in opposite pairs, from oblong to lanceolate and with a serrate margin. Leaf colors range from dark green and gray-green to purple. The flowers are white to purple and produced in false whorls called verticillasters. The corolla is two-lipped with four sub-equal lobes, the upper lobe usually the largest. The fruit is a nut-let, containing one to four seeded. It has numerous applications in the kitchen, being used all over the world as flavoring as well as key ingredients to foods and drinks. Mint oil is used in pharmaceuticals. Mint leaves also repel ants and cockroaches

II. Qualitative estimation for important chemical constituents was carried out and results were presented in table.
Phytochemical analysis revealed that ethnolic, methnolic and aqueous extracts of all the five aromatic plants contained rich source of bioactive compounds such as alkaloids, flavonoids, phenol, tannins, steroids and glycoside. Aqueous extract do not contain saponin, tannins & glycosides with few exceptions. Also observed the positive result for the presence of Flavonoids, Phenols and terpenoids in almost all samples. From this analysis methanolic and ethanolic leaf powder extracts were found to have more chemical constituents compared to aqueous extracts.

PLATE- I

Fig: 1. Plant Samples Collection from JMPC Botanical Garden

PLATE-II

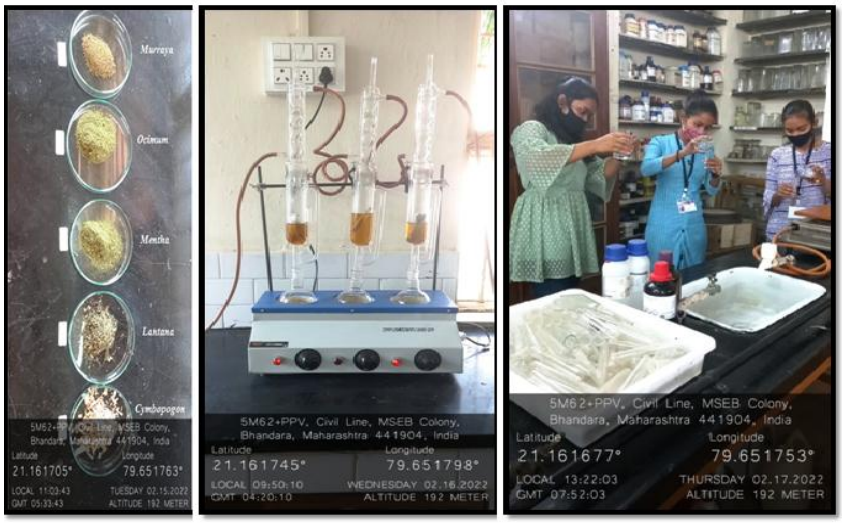

Fig:2 Phytochemical Tests performing in Department of Botany

Table Qualitative Phytochemical Analysis of some Aromatic Plants.

Variabels	*Cymbopogan*			*Lantana*			*Mentha*			*Murraya*			*Ocimum*		
	Ethanol	Methanol	Aqueous	Ethanol	Methanol	Aqueous	Ethanol	Methanol	Aqueous	Ethanol	Methanol	Aqueous	Ethanol	Methanol	Aqueous
Tanins	+	-	+	-	+	-	+	-	-	+	-	-	+	+	-
Saponins	+	+	-	+	-	-	-	-	-	+	-	-	+	-	-
Alkoloids	+	-	-	+	-	-	+	-	-	+	-	-	-	+	-
Steroids	-	-	+	-	-	-	-	-	-	-	+	-	-	-	-
Flavonoid	-	+	+	+	-	-	-	+	-	+	+	-	-	+	+
Terpenoid	+	+	-	+	+	+	+	+	+	+	+	+	+	-	+
Glycosoid	+	+	-	-	+	-	+	-	-	-	+	-	-	+	-
Phenols	+	-	-	+	+	+	+	+	-	+	-	-	+	+	-
Essential Oil	+	+	-	-	+	-	+	+	-	+	-	-	-	+	+

'+' = Present'
'-' = Absent (undetectable)

The major chemical components of the aromatic plants are Volatile oils, which are complex mixtures of variable composition. These essential oils largely comprise different types of terpenoids and benzenoids. The extraction and purification of phytochemical and antioxidant substances from the plant material are generally affected by various factors including time, temperature, solvent concentration and solvent polarity. Depending on chemical nature, various phytochemicals are extracted in solvents of different polarity as no single solvent may be reliable to extract all the phytochemical and antioxidant compounds present in the plant material (Lapornik *et.al.*, 2005). Serial exhaustive extraction method involves the successive extraction with solvents of increasing polarity from non-polar (*n*-hexane) to more polar solvent (water) to ensure the extraction of a wide range of compounds with different polarity **(**Das *et.al.*, 2010**;** Bimakr *et al*., 2011**;** Abdel-Aal *et.al.*, 2015).

The secondary metabolites contribute significantly towards the biological activities of medicinal plants such as hypoglycemic, antidiabetic, antioxidant, antimicrobial, antiinflammatory activities etc. The essential oils are the most popular secondary metabolites of the plants, used for thousands of years regarding the variety of objectives, principally for their health benefits Sharif-Rad J et.al., 2017. Essential oil or essence is referred to by this name due to their primary principle. Briefly, essential oils are secondary metabolites biosynthesized in different plant organs (Rassem HHA et.al., 2016) obtained by mainly hydrodistillation from almost all parts of the aromatic plants of the secretary special elements with volatile properties along with characteristic fragrances and soluble, in organic solvents (Nieto G. 2017). The chemical composition of the essential oils is quite complex including mostly terpenes (monoterpenes and partly sesqui-terpenes formed by isoprene) and aromatic compounds derived from phenylpropane and phenolic constituents (Bakkali F et.al., 2008 & Rassem HHA et.al., 2016). These essential oils known as fragrant oils, steam volatile liquids, or semi-liquids, ethereal oils are concentrated hydrophobic aromatic oil.

Generally the phenolic compounds exhibit aromatic smell, as usual in the present investigation also all the five selected medicinal aromatic plants were found to possess phenols, flavonoids, terpenoids and essential oils. On the other hand -Tanins, Saponins, Alkoloids, Steroids, Gums, Glycosoid were not detected in most of the solvent extracts of all the five plants. Terpenoids can be used as protective substances in storing agriculture

products as they are known to have insecticidal properties as well- surprisingly, in the present report (Table), all the plants exhibited terpenoids in most of the solvent extracts indicates that these plants have a insect repellent property.

CONCLUSION

The preliminary phytochemical analysis of these selected plants showed that aromatic significance is due to the occurrence of many phenolic compounds, essential oils of secondary metabolites. The preliminary phytochemical screening will help to understand variety of chemical compounds produced from different parts of the plants. As a defense mechanism, many plants contain biomolecules which will be released by the plants to protect against insects and pathogens. Therefore these bioactive compounds isolated from different plants may contribute in future to develop novel plant based drugs and also biocides for safe organic food production.

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REFERENCES

1. Abdel-Aal EI, Haroon AM, Mofeed J. Successive solvent extraction and GC-MS analysis for the evaluation of the phytochemical constituents of the filamentous green alga Spirogyra longata. Egypt J Aquat Res., 2015; 41(3): 233-246.
2. Bakkali F, Averbeck S, Averbeck D, Idaomar M. Biological effects of essential oils—A review. Food and Chemical Toxicology, 2008; 46: 446-475.
3. Bharathidasan, R., V.Sathya, V S.S. Tamil, N.R. Sophia, R. Lakkiya and Prabakaran, M. Quantitative, qualitative phytochemical analysis and in vitro antibacterial activity of *Bauhinia tomentosa L. J. Nat. Prod. Plant Resour*., 2013; 3(2): 31-36.
4. Bimakr M, Rahman RA, Taip FS, Ganjloo A, Salleh LM, Selamat J. Comparison of different extraction methods for the extraction of major bioactive flavonoid compounds from spearmint (Mentha spicata L.) leaves. Food Bioprod Process, 2011; 89(1): 67-72.
5. Das K, Tiwari RKS, Shrivastava DK. Techniques for evaluation of medicinal plant products as antimicrobial agent: Current methods and future trends. J Medicinal Plants Research, 2010; 4(2): 104-111.
6. Devmurari, V. P. Phytochemical screening study and antibacterial evaluation of *Symplocos racemosa* Roxb. *Archives of Applied Science Research*, 2010; 2(1): 354-359.
7. Essential_Oils_Introduction-Web page: http://www.theherbsplace.com/index.html.
8. Gershenzon J The cost of plant chemical defense against herbivory: a biochemical perspective. In Insect-Plant Interactions, 1993; 121-190.
9. Harborne JB Phytochemical methods: A guide to modern techniques of plant analysis, Springer International 3rd edition, 1973.
10. Lapornik B, Prošek M, Wondra AG. Comparison of extracts prepared from plant by-products using different solvents and extraction time. J Food Eng, 2005; 71(2): 214-222.
11. Making Essential Oils - Methods of Essential Oil Extraction" from the Webpage of http://www.anandaapothecary.com/essential-oils.html.
12. Nieto G. Biological activities of three essential oils of the Lamiaceae family. Medicine, 2017.
13. Parmar, P., S. Bhatt, S. Dhyani and Jain, A. Phytochemical studies of the secondary metabolites of *Ziziphusm auritiana*. *International Jou of current Pharmace. Res.*, 2012; 4(3): 153-155.
14. Rassem HHA, Nour AH, Yunus RM. Techniques for extraction of essential oils in plants: A review. Australian Journal of Basic and Applied Sciences, 2016; 10(16):117-127.
15. Sabri, F.Z., M. Belarbi, S. Sabri and Alsayadi, M.M.S. Phytochemical screening and identification of some compounds from Mallow. *J. Nat. Plant Resour.*, 2012; 2(4): 512-516.
16. Sharif-Rad J, Sureda A, Tenore GC, Daglia M, Sharif-Rad M, Valussi M, Tundis R, Sharif-Rad M, Loizzo MR, Ademiluyi AO, Sharif-Rad R, Ayatollahi SA, Iriti M. Biological activities of essential oils: From plant chemoecology to traditional healing systems. Molecules, 2017; 22(70): 1-55.
17. Singh, N.P. and Karthikeyan S. Flora of Maharashtra State Dicotyledons, 2000; 1(BSI).
18. Singh, N.P., Lakshmiarasimhan P., Karthikeyan S. and Prasanna P.V. Flora of Maharashtra State Dicotyledons, 2000; 2(BSI).
19. Solihah, M. A., W.I. Rosli and Nurhanan, A.R. Phytochemical screening and total phenolic content of Malaysian *Zea mays*. *Int. food Res. J.*., 2010; 19(4): 1533-1538.
20. Ugochuhwu, S.C., A. Uche and Ifeanyi, O. Preliminary phytochemical screening of different solvent extracts of stem, bark, and roots of *Dennetia tripetala*. *Asia J of Plant Sci and Res.*, 2013; 3(3): 10-13.