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Chemical Polymorphism in Asteraceae

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The qualitative and quantitative composition of the essential oils of seven plants of asteraceae family were investigated. The essential oils were obtained by hydro-distillation of plants and analysed by Gas Chromatograph. The major essential oil components identified, broadly belong to monoterpenoids, sesquiterpeniods and caryophyllene were found to be common in all the investigated species.

***Key words*: Chemical polymorphism, Asteraceae, Essential oil, Hydro-distillation, Monoterpinoids, Sesquiterpenoids, Caryophyllene.

Introduction

Asteraceae are chemically characterised by synthesis and accumulation of many classes of natural products. The essential oils are comprise of the volatile steam - distillable fraction, responsible for the characteristic scent, odour or smell found in many plants. They are commercially important as the basis of natural perfumes, and also of spices and flavouring in the food industry. A considerable number of quite different functions have been ascribed to plant terpenoids. Their growth regulating properties are very well documented, two of the major classes of growth regulators are the sesquiterpenoid abscisins and the diterpenoid based gibberellins. Less is generally known of the role of terpenoids in the more subtle interactions between plants and animals as agents of communication and defence among insects, but this is now an area of active research. Many studies have demonstrated that terpenoids display a wide range of pharmacological activities. Some of these compounds have been identified as the active principles of crude drugs used in folk medicines. The important class of sesquiterpenes, sesquiterpene lactones, which have a wide distribution in the family Asteraceae (Herout and Sorn 1969).

Even though a considerable amount of work has been done on the various members of Asteraceae, on the chemistry of essential oils, however, they have focussed only on a few economically or commercially valuable species. The screening of a large number of lesser oil yielding plants of great medicinal importance, has not been attempted so far. It appears from the previous literature that only few species of the family Asteraceae has so far been phytochemically screened (Riaz *et al* 1995; Chandra *et al* 1996; Wandji *et al* 1996; Chuihua 1998). The data on the chemistry of essential oils of Asteraceae too are very meagre. Considering the richness of this family in the vegetation of India, it has been thought that a systematic work with regard to its essential oil constituents and their significance may lead to a better understanding of these plants. As most of the plants of Asteraceae are used in folk medicines or grand mother's remedy for common diseases.

The present study is therefore, undertaken to find out the quantity of essential oil in medicinal plants of Asteraceae and also to identify the major components in these oils. A chemo taxonomical evaluation based on volatile oils in various taxa and on the range of various oil constituents suggested making use of both studies for understanding the variation and formation of taxa in nature as well as in the natural classification of plants.

Materials and Methods

Plant materials. For the extraction of essential oils, the raw materials were collected from seven aromatic members of Asteraceae. The collected plant materials were cleaned and shade dried at room temperature, as shade drying reduces the weight of herb to 1/3 of the fresh weight and maximizes the oil yield with out affecting the quality of the essential oil.

Isolation of essential oil. Separation of the volatile oils from the dried, flaked and powdered plant tissue was conducted by hydro-distillation in a Clevanger apparatus (Clevanger 1928) for 4 to 5 h, as prolonged extraction normally increases the yield (Gildemeister and Hoffman 1961). Extraction was carried out at 80 to 230°C temperature. The percentage of essential oils was calculated on a dry weight basis to avoid faulty estimation that may arise due to the different water content of the tissue analysed each time (Von Rudloff

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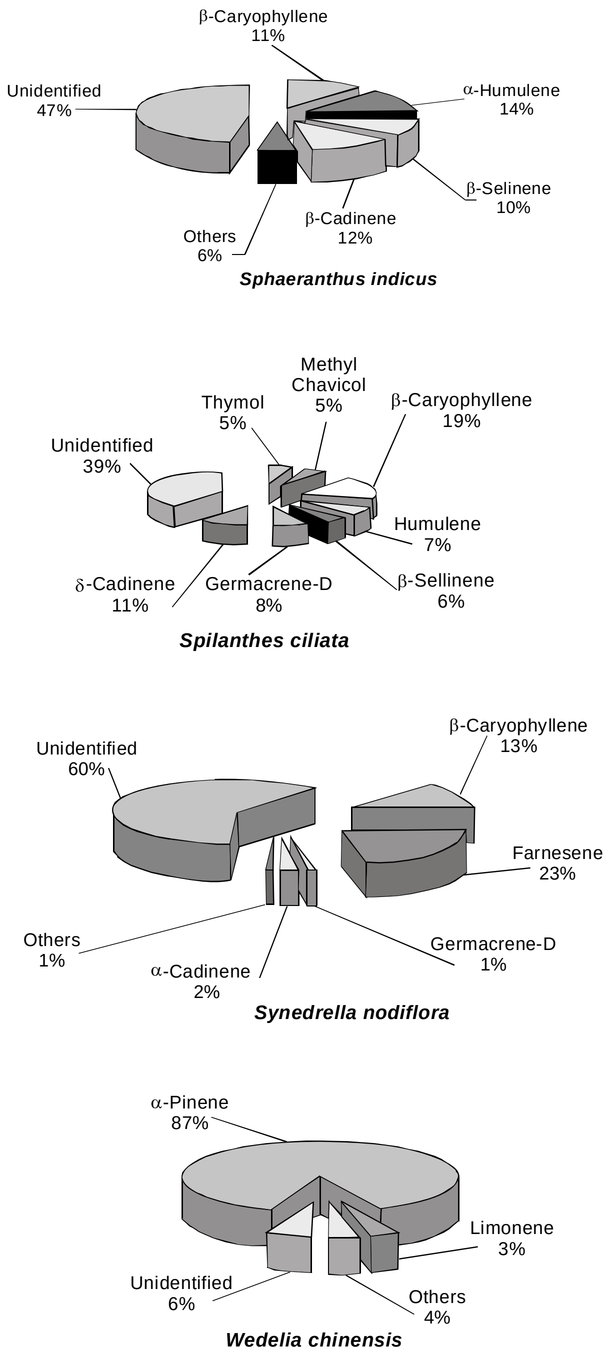


Fig 1. Composition of major essential oil components of *S. indicus,S.ciliata, S. nodiflora and W. chinensis.*

1972). The isolated oil was then dried over anhydrous sodium sulphate and stored at 4 to 6°C.

Qualitative analysis. Qualitative estimation of the essential oils is done by capillary Gas Chromatography (GC) (Bahl and Bahl 1983). For each plant, GC analysis was performed by using a Chemito Gas Chromatograph (model CHEMITO 8510) equipped with a flame ionisation detector (FID). The capillary

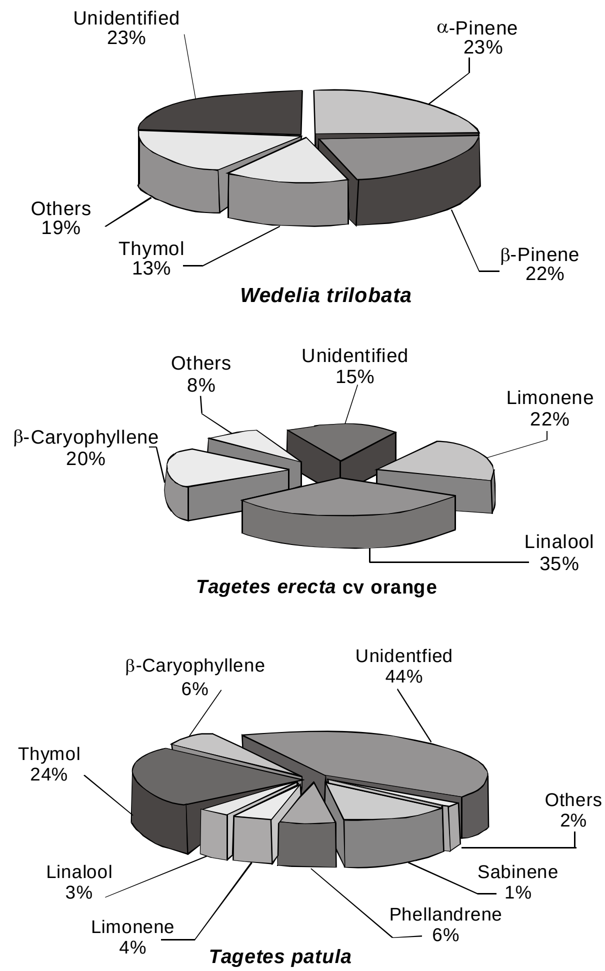


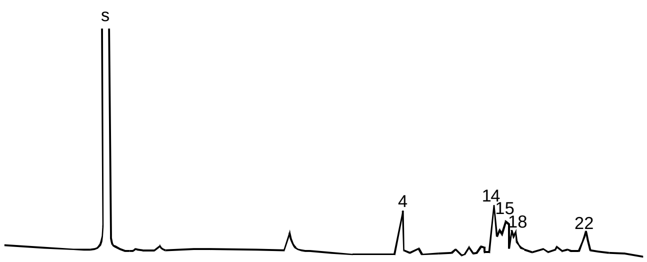
Fig 2. Composition of major essential oil components of *W. trilobata, T. erecta* cv orange and *T. patula.*

GC was carried out on 5% SE – 30 packed column with the following dimensions mesh size – 80/100; length - 8 feet, diameter - 1/8 inch. Nitrogen was used as a carrier gas at 10 psi (inlet pressure) with a flow rate of 30 ml/min. Temperature programming was performed from 80 to 230°C at the rate of 6°C/ min. Major components were identified by retention time (RT) analysis (Finar 1975) and peak enrichment by co-injection with authentic standards (Jefferys *et al* 1989). Relative retention times of these reference standards were obtained by taking gas chromotograms of their mixtures in similar conditions.

Quantitative analysis. In GC, the quantification of the peak areas were done by the Shimadzu integrator having a built in computer (model CR3A - CHROMATOPAC). The quantitative data, obtained thereby, were based on computer integrated peak area calculations. Since the essential oils in most cases were diluted with a solvent (toluene), corrections have been made accordingly and accurate quantitative values were determined for each component separately. The corrected values are shown in the pie chart, Fig 1 & 2.

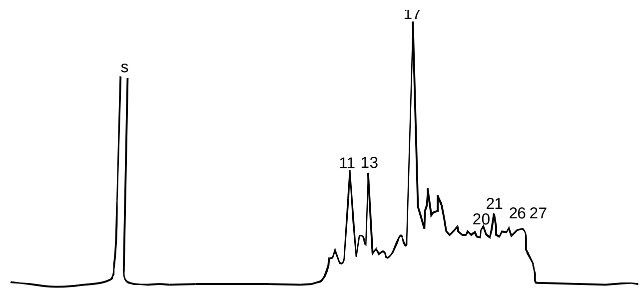
Table 1
List of various taxa investigated for their major essential oil components and chemotypes

S.No.	Name of taxa	Essential oil components	(%)	Chemotype
1.	*Sphaeranthus indicus*	α-Humulene	14.10%	
		δ-Cadinene	12.50%	Mixed
		β-Caryophyllene	10.80%	
		β-Selinene	9.85%	
		β-Ionone	5.50%	
2.	*Spilanthes ciliata*	β-Caryophyllene	19.36%	
		δ-Cadinene	10.97%	Mixed
		Germacrene-D	8.00%	
		α-Humulene	6.60%	
		β-Selinene	5.60%	
		Methyl chavicol	5.30%	
		Thymol	5.00%	
3.	*Synedrella nodiflora*	Farnesene	22.75%	
		β-Caryophyllene	12.70%	Mixed
		δ-Cadinene	2.35%	
		Germacrene-D	1.27%	
		Limonene	0.27%	
		β-Pinene	0.20%	
		α-Pinene	0.03%	
4.	*Wedelia chinensis*	α-Pinene	87.60%	α–pinene
		Limonene	3.33%	
		β-Pinene	2.70%	
		β-Caryophyllene	0.38%	
		Borneol	0.23%	
5.	*Wedelia trilobata*	α-Pinene	23.70%	
		β-Pinene	21.99%	Mixed
		Thymol	12.70%	
		β-Caryophyllene	5.26%	
		Limonene	5.21%	
		β-Phellandrene	3.65%	
6.	*Tagetes erecta* cv orange	Linalool	34.50%	
		Limonene	21.95%	
		β-Caryophyllene	20.36%	Mixed
		Piperitone	3.13%	
		β-Pinene	2.96%	
		Sabinene	1.29%	
		α-Pinene	0.36%	
7.	*Tagetes patula*	Thymol	23.65%	
		Sabinene	11.29%	Mixed
		β-Caryophyllene	6.19%	
		β-Phellandrene	5.96%	
		Limonene	4.18%	
		Linalool	3.38%	
		β-Pinene	0.97%	
		Piperitone	0.64%	
		α-Pinene	0.37%	



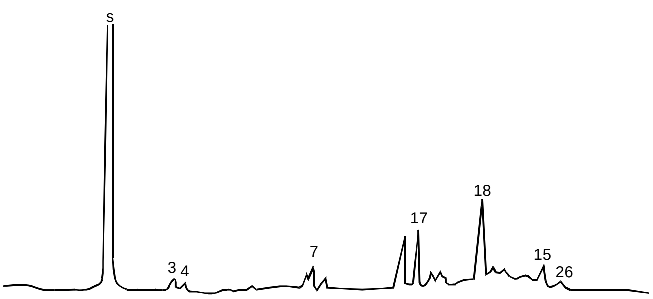
(a) Sphaeranthus indicus

S-solvent, 4-β-caryophyllene, 14-humulene, 15-selinene,18-ionone, 22-δ-cadinene.



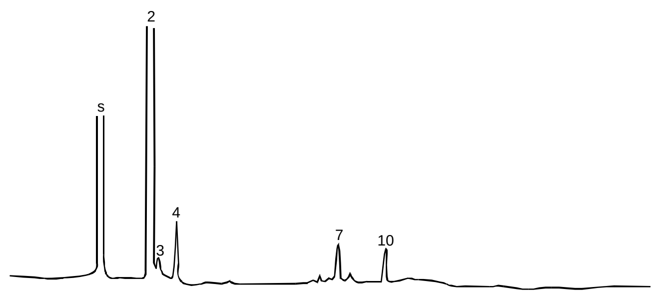
(b) Spilanthes ciliata

S-solvent, 11-thymol, 13-methyl chavicol, 17-β-caryophyllene, 20-humulene, 21-β-selinene, 26-germacrene-D, 27-δ-cadinene.



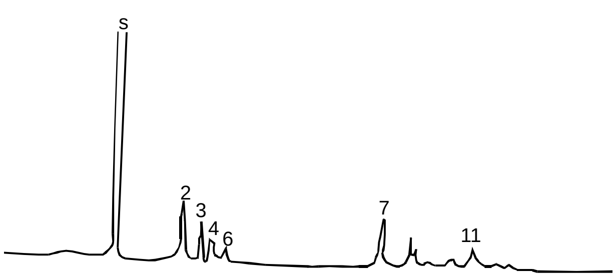
(c) Synedrella nodiflora

S-solvent, 3-α-pinene, 4-β-pinene, 7-limonene, 13-β-caryophyllene, 18-farnesene, 25-germacrene-D, 26-δ-cadinene.



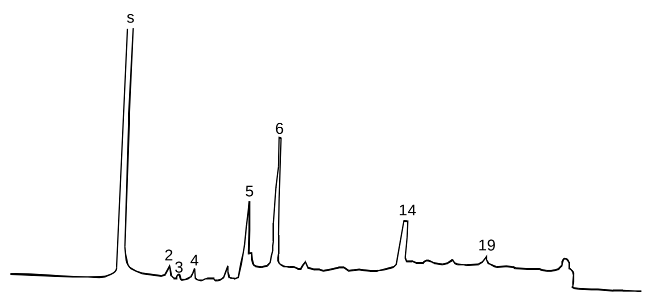
(d) Wedelia chinensis

S-solvent, 2-α-pinene, 3-β-pinene, 4-limonene, 7-borneol, 10-β-caryophyllene.



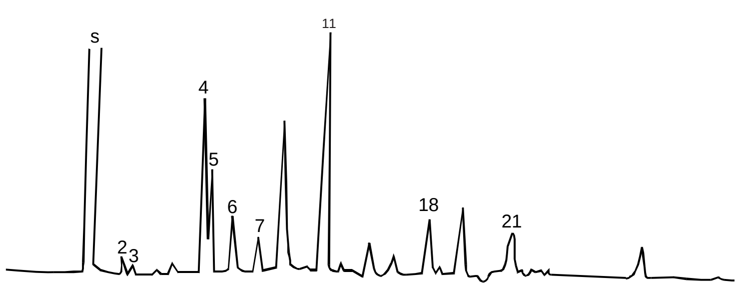
(e) Wedelia trilobata

S-solvent, 2-α-pinene, 3-β-pinene, 4-β-phellandrene, 6-limonene, 7-thymol, 11-β-caryophyllene.



***(f) Tagetes erecta* cv orange**

S-solvent, 2-α-pinene, 3-β-pinene, 4-sabinene, 5-limonene, 6-linalool, 14-β-caryophyllene, 19-piperitone.



(g) Tagetes patula

S-solvent, 2-α-pinene, 3-β-pinene, 4-sabinene, 5-β - phellandrene, 6-limonene, 7-linalool, 11-thymol, 18-β - caryophyllene, 21-piperitone.

Fig 3. Gas chromatograms of various essential oil yielding plants of (a) *Sphaeranthus indicus* (b) *Spilanthes ciliata* (c) *Synedrella nodiflora* (d) *Wedelia chinensis* (e) *Wedelia trilobata* (f) *Tagetes erecta* cv orange (g) *Tagetes patula*.

Evaluation of ecological role of terpenoids. The role of terpenoids in the ecology of different plant is evaluated, based on the field survey and observations noted during the collection of plants from original localities and cultivation of different plants in the botanical garden. The available literature data is also being used to correlate these information to discuss the role of these chemical components in the well being of these plants.

Results and Discussion

The investigation results of seven following taxa for their major essential oil components and chemotypes have been summarised in Table 1. A mixed chemotype is noted in *Sphaeranthus indicus* and sesquiterpene predominate over monoterpenes in the oil sample. α-Humulene (14.10%), δ-cadinene (12.50%), β-caryophyllene (10.80%), β-selinene (9.85%) and β-ionone (5.50%) are the major components present. The medicinal properties and other value added properties reported on this plant are attributed to these chemical compounds (Fig 3a - g).

The medicinal plant *Spilanthes ciliata* belong to mixed chemotype. The main components are β-caryophyllene (19.36%), δ-cadinene (10.97%), germacrene-D (8.00%), α-humulene (6.60%), β-selinene (5.60%), methyl chavicol (5.30%) and thymol (5.00%). Previous reports of Lemos confirmed the presence of these components in *S.ciliata* (Lemos *et al* 1991). The presence of these components seems to be responsible for the medicinal properties reported for *S.ciliata*. In *Synedrella nodiflora* mixed chemotype is noticed, α-farnesene (22.75%) and beta-caryophyllene (12.70%) are the major components identified. Previous chemical reports of Aalbersberg and Sing (1990) also agreed with the presence of these components.

The two species studied under the genus *Wedelia* show clear interspecific variations with regard to the chemical constituents. The chemotype of *W. chinensis* is seemed to be an α-pinene chemotype. More than 50% of the oil contains monoterpenes. The major essential oil constituents, identified are α-pinene (87.60%), limonene (3.33%) and β-pinene (2.70%). While in the oil rich taxa *W.trilobata* is a mixed chemotype. Major components are α-pinene (23.70%), β-pinene (21.99%), thymol (12.70%), β-caryophyllene (5.26%), limonene (5.21%) and β-phellandrene (3.65%). Previous reports show the existence of these components in *Wedelia* (Craverio *et al* 1993).

The two species studied under the genus *Tagetes* show marked similarity. *Tagetes erecta* cv orange and *T. patula* are characterised with a mixed chemotype. The identified major components of the oil of *T. erecta* are linalool (34.50%), limonene (21.95%) and β-caryophyllene (20.36%). In the case of *T. patula*, the main components were found are thymol (23.65%), sabinene (11.29%), β-caryophyllene (6.19%), β-phellandrene (5.96%), limonene (4.18%), and linalool (3.38%) along with traces of β-pinene (0.97%), piperitone (0.64%) and α-pinene (0.37%). Thymol is a reported chemical compound well known for its medicinal, aromatic and flavouring properties (Sobti *et al* 1977). This might have been the major factor responsible for the value added properties reported on *Tagetes.* Tagetes oil is commercially produced from *T.patula.* It is mainly used for the compounding of high grade perfumes. The oil, with its floral note and strongly waxy character, could find some special applications in perfumery.

Sesquiterpenes have a common occurrence in the essential oil of Asteraceae. Sesquiterpene lactones have an important role in Asteraceae members as chemical defence agents. They are often present in high concentration and relatively complex mixtures of different structures are found in many species. Usually bitter tasting, they are obviously deterrent to many kinds of herbivore, including man himself (Harborne 1991). Their effects on insects are very damaging and they are also poisonous to live stocks. Other properties possessed by these sesquiterpene lactones include their pungent taste and their ability to act as allergens (Mitchell *et al* 1970).

The biological activities associated with sesquiterpenes, are many and varied, from plant growth regulation (abscissic acid) to interfere with insect metamorphosis. The sesquiterpene lactones likewise have a wide range of demonstrated biological activities, as cytotoxic compounds, vertebrate poisons insect feeding deterrents, schistosomicidal substances, and allergenic agents (Rodriguez *et al* 1976). Some sesquiterpenes protect plants from insect attack because of their antimicrobial properties. This appears to be true of caryophyllene and caryophyllene epoxide which are repellent to the leaf cutting ants *Attacephalotes* and *Acromyrmex octopinosus*. Caryophyllene is present in all investigated species as revealed in Fig 3.

Conclusion

The natural purpose of the synthesis and accumulation of terpenoids in the plant kingdom still possess a considerable question mark to phytochemists. As discussed above, they have some role in plant growth regulation. In pollination biology, in various interactions with other plants, animals or insects, in defence against diseases and in other attacks. However, the studies on the chemical defensive mechanism of terpenoids are still at an early stage. Hence, it is high time to collect sufficient data and to duly emphasis the ecological role of terpenoids as primarily defensive agents against over grazing. This topic therefore, offers ample scope for future research programmers.

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