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Research Article

ISOLATION AND CHEMICAL CHARACTERIZATION OF VOLATILE OILS OF CENTRATHERUM ANTHELMINTICUM AND CITRUS AURANTIFOLIA

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ABSTRACT

Introduction: *Centratherum anthelminticum* is an ethnomedicinal plant in India and a common ingredient in Ayurvedic formulations. The pharmacological effects of *C. anthelminticum* ranges from anti-oxidant, anti-diabetic, anti-microbial to recently found anti-cancer activity. *Citrus aurantifolia* (Rutaceae) is mainly used in daily consumption, in many cultural cuisines, and in juice production. It is widely used because of its antibacterial, anticancer, antidiabetic, antifungal, anti-hypertensive, anti-inflammation, anti-lipidemia, and antioxidant properties. **Objective:** To isolate and identify the compounds in the essential oils from the seeds of *C. anthelminticum* Kuntz and peels of *C. aurantifolia* (Christm.) swingle collected from the Delhi NCR region. **Materials and Methods:** The essential oils from seeds of *C. anthelminticum* Kuntz and peels of *C. aurantifolia* were extracted by steam distillation and analyzed by gas chromatograph and gas chromatography–mass spectrometer. **Results:** Chemical characterization of the volatile oil and seeds of *C. anthelminticum* Kuntz showed that the oil is greenish and semi viscous. The major component of the oil was found to be beta-caryophyllene (64.43%), beta-pinene (9.81%), alpha-humulene (4.64%), caryophyllene oxide (2.24%), gamma-elemene (1.73%), limonene (1.48%) and myrecene (0.11%). Whereas the major constituents of volatile oil from peels of *C. aurantifolia* comprises of twenty nine components viz. sesquiterpne alcohols (20.68%) including Globulol, Hurmularel, 6 - dien - 3 - ol. (-) -Spathulenol, ledol,alpha-bisabolol, - (-) Spathulenol and Sesquíterpene esters (20.68%), caryophyllene, alpha- Farnesene, alpha-Caryophylline, gamma - elemene, gamma-.neoclovene, monoterpene alcohol (13.79%). **Conclusion:** The present study revealed the chemical composition of essential oils of both the drugs under study which can be used as references in the food, pharmaceutical and cosmetic industries.

Keywords: Aromatic, Essential oil**,** Centratherum anthelminticum, Citrus aurantifolia, GC-MS, Terpenes.

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INTRODUCTION

In the recent years, there has been an increased interest in the use of aromatic plants, which are used in different areas of life due to their various properties. According to the literature, essential oils have been known to possess antioxidant properties, expectorant, diuretic, antimicrobial and antifungal activities as well. Besides their strict medicinal applications, essential oils are extensively used in many fields such as food industry, perfume products, cosmetics and chemical industry. Since antiquity, aromatic plants have been used for their medicinal properties.[1]

Essential oils, which are complex mixtures of volatile compounds particularly abundant in aromatic plants, are mainly composed of terpenes biogenerated by the mevalonate pathway. These volatile molecules include monoterpenes (hydrocarbon and oxygenated monoterpens), and also sesquiterpenes (hydrocarbon and oxygenated sesquiterpenes). Furthermore, they contain phenolic compounds, which are derived via the shikimate pathway. Thanks to their chemical composition, essential oils possess numerous biological activities (antioxidant, anti-inflammatory, antimicrobial, etc.) of great interest in food and cosmetic industries, as well as in the human health field. *Centratherum anthelminticum* (L.) Kuntze is an ethnomedicinal plant commonly grown in India and Southeast Asia. It is a member of Asteraceae family of the flowering plants. *Vernonia anthelmintica* and *Conyza anthelmintica* are scientific synonyms of this plant. Locally, the plant is known as black/bitter cumin, or kalijiri in India. The plant is an erect, pubescent, annual herb which can grow up to 85 cm in height. The leaves of the plant are elliptic-lanceolate, 3 to 8 cm long and 2 to 3 cm wide. The apex of the leaves is acute, base tapering into the petiole, margins coarsely serrate and pubescent on both surfaces. It has homogamous purple florets, which can be found as solitary, axillary or terminal heads. The seeds are brownish in color, with a hot sharp taste and astringent properties. It is widely used as folk medicine for diabetes in Rayalaseema, India and a popular ingredient in Ayurvedic medicine. In other places, *C anthelminticum* has been traditionally applied as anthelmintic, stomachic, digestive, diuretic, tonic, alterative, anti-phlegmatic, anti-asthmatic, anti-phlegmatic

treatment, as well as a therapeutic agent for cough, diarrhea, helmint, skin diseases, ulcers, leucoderma and fevers. [2] [3] [4] [5]

Lime fruit (*Citrus aurantifolia* (Christm.) Swingle) Lime, Key lime (Mexican), Kagji nimboo (Hindi) is a member of Rutaceae family. It is an evergreen, spiny shrub or small tree 3 to 5 m in height. The plant has single or multiple stems and irregular branches covered with smooth brown to gray bark.

The twigs are quadrangular (when young), green, and bare sharp axillary spines 2 to 15 mm long. The leaves are yellow-green to dark green, with 4 to 25 mm winged petioles and elliptic to oval leathery 4- to 13-cm long blades with edges that have minute rounded teeth. The crushed foliage has a strong, distinct, spicy (citrus) odor and taste. The four- to five-petaled white flowers occur in few-flowered axillary clusters. The fruits are ellipsoidal, 3 to 5 cm in diameter, have juicy, greenish yellow flesh, and are yellow at maturity. They contain a few white, pointed seeds about 1 cm long. [5] [6] [7]

The present work revealed the isolation and identification of major compounds of aromatic plant *Centratherum anthelminticum* kutz and *C. aurantifolia* (christm.) swingle.

MATERIALS AND METHODS

Plant material

The plant material was collected from the Delhi NCR region. The plant was authenticated and identified as *Centratherum anthelminticum* Kuntz. (Compositae) by the Taxonomist Dr. H.B. Singh, National Institute of Science Communication and Infomation Resources AIR), New Delhi. A voucher specimen (Specimen No: NISCAIR/RHMD/Consultid -11/1503/101) has been preserved in Herbarium section of Taxonomy Department of ol NISCAIR, New Delhi.

Fresh peels of *C. aurantifolia* (Christm.) Swingle were collected from the herbal garden of Ram-Eesh Institute of Vocational and Technical Education, authenticated and identified as C. aurantifolia (Christm) Swingle, Family Rutaceae by Dr. Anjula Pandey (Taxonomist), National Bureau of Plant Genetic Resources (NBPGRL Pusa Campus, New Delhi.) A voucher specimen (Specimen No: NHCP / NBPGR 2010-30 / 2675) is preserved in herbarium section of taxonomic deptt. of NBPGR New Delhi and also the Pharmacognosy laboratory, Dept of pharmacy, Ram-Eesh Institute of Vocational and Technical Education, Greater Noida, Uttar Pradesh

Extraction of essential oils

The dried seeds of *C. anthelminticum* and fresh peels of *C. aurantifolia* (500 gm) were hydro-distilled for 3 hours according to the recommended method (British pharmacopoeia, 1998) .The yellow greenish colored volatile oil of *C. anthelminticum* and yellow colored volatile oil of *C. aurantifolia* was collected in the graduated tube.

The collected volatile oil was dried over anhydrous sodium sulphate and stored at 4^0C in the dark. Volatile constituents were identified by comparing their area and retention times with those of authentic standards available in the author's laboratory and with those of literatures. [8] [9]

Chemical analysis of essential oils

Analytical GC was carried out on a Varion 3300 GC fitted with a silicone DB-1 capillary column 30 m X025 mm), film thickness 0.25 um, carrier gas Nitrogen, flow rate 15 mU min., split mode, temperature programmed 180- 250 °C at 4 ° C / min. Injector temperature and detector temperature were 250 °C and 300 ° C respectively. Detector used was FID.

Injector volume for all samples was 0.1 ul. 8.1.4 GC-MS Analysis GC-MS analysis was carried out on a Shimadzu QP-2010 instrument at 70 eV and 250 ° C. GC column Ublon HR-1 fused silica capillary 025 mm X 50 m with film thickness 0.25 um. The initial temperature was 100% C for 6 min, and then heated at a rate of 10 C / min. to 250 C. Carrier gas Helium, flow rate 2 ml / min, detector used was FID. [8]

Identification

The volatile were components were identified by comparing their retention indexes of GC with those of literature. Further identification was done by GC-MS. The fragmentation patterns of mass spectra were compared with those of the database using NBS S4 AL and Wiley L-built in libraries and also with those reported in the literature. Many constituents were identified by comparing their retention time and area % with those of authentic standards available in Software's library. [8]

RESULT AND DISCUSSION

Physical properties and yield

Chemical characterization of the volatile oil of seeds of C. *anthelminticum* Kuntz showed that the oil is greenish and semi viscous. The major components of the oil were identified as beta-caryophyllene (64.43%), β-pinene (9.81%), α-humulene (4.64%), caryophyllene oxide (2.24%), γ-elemene (1.73%), limonene (1.48%) and myrecene (0.11%). Chemical characterization of the volatile oil of lemon peel revealed the presence of twenty nine components. The volatile oil showed the presence of high amount of sesquiterpme alcohols constituents (20.68%); Globulol, Hurmularel, 6 - dien - 3 - ol. (-) -Spathulenol, ledol,alpha-bisabolol, - (-) Spathulenol and Sesquíterpene esters (20.68%), caryophyllene, alpha- Farnesene, alpha-Caryophylline, gamma - elemene, gamma-.neoclovene, monoterpene alcohol (13.79%); Eugenol, p-menth-1-en-8-ol, Dihydronopol, p-menth-1-en-8-ol, monoterpene hydrocarbon (10.34); alpha- Pinene, 3 -Carene, Limonene, sesquiterpene ketone (3.44%); corymbolone, sesquiterpene ether (3.44%); Eudesma - 4- (14), 11 - diene, Ditemene ketone (3.44%); Shyobonone, Others components (10.34%); Neric acid, Trans - Z - a - bisabolene epoxide etc .

The relative percentage area, RT (retention time) and identity of components of volatile oils of *C. anthelminticum* Kuntz seeds and *C. aurantifolia* peels were presented in Table 1 and Table 2 respectively.

The author declares that he has no competing interests.

Table 1: Area% & RT of Volatile Constituents of *C. anthelminticum* Kuntz seeds.

S. No.	Volatile Components	Area%	RT
1.	Alpha-Pinene	1.3674	6.124
2.	Beta- Pinene	9.8153	7.224
3.	Sabinenc	0.1673	7.375
4.	Myrcene	0.1109	7.973
5.	Limonene	1.4883	8.767
6.	Eucalyptol	0.9726	9.024
7.	Gamma-terpinene	0.1351	9.718
8.	p-cymene	0.2078	10.329
9.	Alpha-copaene	0.3194	16.187
10.	Camphor	0.7831	17.234
11.	Beta-linalool	0.1259	17.784
12	Beta-caryophyllene	65.4358	19.534
13.	Carvone	0.5510	19.927
14.	Dihydrocarvone	0.2816	20.434
15.	Alpha-humulene	4.6228	21.369
16.	Borneol	0.8568	22.376
17.	Azulene	0.7544	23.681
18.	Beta-sesquiphelland	0.2961	24.001
19.	Ar-curcumene	0.4080	24.120
20.	Methyl chavicol	0.1054	24.774
21.	Caryophylene oxide	2.2480	30.055
22.	Carotol	0.2505	30.862
23.	Humulene epoxide	0.2048	31.643
24.	Caryophyllenyle alcohol	0.1541	31.963
25.	Gamma-elemene	1.7312	36.221
26.	Alpha-farnesene	1.7025	37.797
27.	GermacreneB	1.8393	39.733
28.	Apiole	0.9443	40.706
29.	Juniperol	1.0141	40.706
30.	Emersol	1.1061	55.874

Table 2: Area% & RT of Volatile Constituents of *C. aurantifolia* peels

S. No.	Volatile Components	Area%	RT
1.	Alpha-Pinene	3.977	7.217
2.	Limonene	36.096	11.184
3.	3-carene	0.073	11.227
4.	Eugenol	0.050	18.583
5.	Caryophyllene	0.966	19.561
6.	Eudesma-4-(14),11-diene	0.060	20.926
7.	Alpha-farnesene	1.575	21.381
8.	Alpha-caryophylline	0.064	21.930
9.	Gamma-elemene	0.224	22.342
10.	Caryophylline oxide	0.457	22.883
11.	Globuol	0.079	23.123
12	Humulane-1,6-diene-3-ol	0.175	23.535
13.	Gamma neoclovene	0.056	23.716
14.	(-)-spathulenol	0.203	23.861
15.	Diepi-alpha-cedrene epoxide	0.076	24.024
16.	Iedol	0.167	24.308
17.	Alpha-bisabolol	0.388	24.840
18.	Isoaromadendrene epoxide	0.092	25.594
19.	Aristolene epoxide	0.137	26.393
20.	Corymbolone	0.101	27.587
21.	p-menth-1-en-8-ol	0.109	14.83
22.	Dihydronopol	0.050	32.891
23.	p-menth-1-en-8-ol	2.503	15.149
24.	(-)-spathulenol	0.188	25.286
25.	Isoaromadendrene epoxide	0.059	27.990
26.	Neric acid	0.144	32.960
27.	Shyobonone	0.088	34.316
28.	Trans-z-a-bisabolene epoxide	0.370	53.432
29.	hexadecanal	0.061	26.840

CONCLUSION

The present study showed the chemical profile of volatile oils from the seeds of C. *anthelminticum* and *C. aurantifolia* along with the percentage content of various components present. This profile can be used as standard for chemical standardization of the above volatile oils and can be successfully employed in food, pharmaceutical, cosmetics and perfumery industries.

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CONFLICT OF INTEREST

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