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Chemical constituents in volatile oil of rhizome and flower of *Curcuma angustifolia* Roxb. growing in North-East India

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Abstract

Curcuma angustifolia Roxb. (Family: Zingiberaceae) is an important rhizomatous medicinal and aromatic plant. The rhizome and flower essential oil of *C. angustifolia* were extracted using methanol and analyzed by Gas chromatography/mass spectrophotometry (GC-MS). The oil yield (% v/w) of the rhizome and flower were 3.25 and 2.40, respectively. A total of 24 and 42 compounds were identified, representing 77-91% and 46-61% of the total rhizome and flower oil, respectively. The major constituents in the rhizome oil were germacrone (27.52%), β-pinene (16.46%) and β-elemene (15.32%) followed by β-elemenone (8.42%) and germacrene (5.68%). The major constituents in the flower oil were β-germacrene (22.44%), 2,2,7,7-tetramethyl tricycle [6.2.1.0](1,6)] undeca-4-en-3-one (15.63%), caryophyllene oxide (7.12%), followed by β-caryophyllene (6.84%) and methyl isomyristate (5.23%). Because of the presence of remarkable phytoconstituents, the rhizome and flower essential oil of *C. angustifolia* would have enough significance in the preparation of pharmaceutical drugs.

1. Introduction

Curcuma angustifolia Roxb. belonging to the family Zingiberaceae, is an important aromatic and medicinal herb (Srivastava *et al*., 2006). Though, distribution of Zingiberaceae is worldwide with over 100-150 species, they are predominantly concentrated in Thailand, China and the Indian Sub-continent (Wu and Larsen, 2000; Triboun, 2006). Many members of Zingiberaceae are of outstanding importance economically since their volatile oils form indispensable ingrediens of perfumery, flavour, fragrance and pharmaceutical industries. The family is of great ethnobotanical value being employed in many indigenous medical systems. Many members of Zingiberaceae are employed in Ayurvedic, Unani, and Homoeopathic systems of medicine.

C. angustifolia is distributed throughout central, southern and eastern India, but most commonly reported from the Northeast and Western Coastal Plains and hills of India (Srivastava *et al*., 2006; Sharma, 2012). People called *C. angustifolia* in different names, *viz.*, East Indian Arrowroot in English, tikhur in Hindi, *yaipan* in Manipuri and Koova in Malayalam. Rhizomes of *C. angustifolia* have been used in ethnomedicine in India as an antiasthmatic, antidysentry, antifungal and anmtipyretic (Tushar *et al*., 2010; Ray *et al*., 2011; Padal and Sandhyasri, 2013). Rhizomes are used for the treatment of bone fracture, inflammation and intestinal disorders (Jain, 1995). *C. angustifolia* produces beautiful flowers and foliages that have commercial value in floriculture as ornamental crops (Maciel and Criley, 2003). Various parts of this plant species have been used either raw or cooked as vegetables in many Asian countries (Devi *et al*., 2014). This plant grows wild in various locations of North East India and the localities of Manipur, Nagaland, Meghalaya and Mizoram consume the inflorescence as food. Local community of Manipur use the flower as a delicacy in various dishes because of its distinct intriguing taste and is popularly used in a chutney known as *Eromba*. Plants used in traditional medicine are still a large source of natural antioxidant, antimicrobial, anticancer agents that might lead to the development of novel drugs (Lee *et al*., 2004) and many naturally occurring substances present in the human diet have been identified as potential chemo-preventive agents against cancer (Atlas *et al*., 1995). Many plants are being utilized as medicines by humans to fight against various diseases since their inception on this Earth (Alam *et al*., 2019). Natural antioxidant effects of compounds derived from medicinal plants are most popular nowadays (Manoharachary and Nagaraju, 2016).

Rhizomes of *C. angustifolia* have been reported by some investigators as rich sources of volatile oils which give them pleasant aroma and medicinal value (Srivastava *et al*., 2006; Sudipta *et at*., 2017). However, the volatile oil composition of flower of *C. angustifolia* has not been reported so far. This prompted us to carry out detailed GC and GC-MS examination of rhizome and flower oils of *C. angustifolia* from North East India.

2. Materials and Methods

2.1 Plant materials

Rhizomes and flowers of *C. angustifolia* were collected (Figures 1; a, b) from Bishnupur district, Manipur, North East India (25º50'-24º43N; 92º58'-94º45'E) in November 2020. The plants were identified by taxonomist and voucher specimens (Accession No. PDDUIAS-CA101) were deposited at the Herbarium of Department of Agricultural Biotechnology, Pandit Deen Dayal Upadhyay Institute of Agricultural Sciences, Manipur.

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2.2 Extraction of essential oil

The fresh rhizomes (750 g) and flowers (500 g) were harvested and washed under running tap water separately and then subjected to distillation for 5 h in a Clevenger type apparatus (Guether, 1972) in methanol. The essential oils were dried over anhydrous sodium sulphate and stored in air tight eppendorf tube at 4ºC for further investigation.

2.3 Gas chromatography/mass spectrometry analysis

GC-MS analysis was performed on a Clarus 680 GC with Clarus 600 C mass spectrometer system, Perkin Elmer, USA with Liquid Auto Sampler, Column 60.0 m long, 250 µm coating thickness. The GC was operated under the following conditions: Initial temperature 60ºC for 2 min, ramp 4ºC/ min to 200ºC, hold 5 min, ramp 8ºC/min to 260ºC, sample volume 2 µl, split 0:1; Solvent delay = 10.00 min, Transfer temp = 200ºC, Scan : 40 to 400 Da. 2 µl volume of ethyl acetate extract was injected into the GC. Carrier gas was helium (99.99% purity) at a constant flow rate of 1.0 ml/min. Mass spectrometer was operated in full scan with 70 eV.

2.4 Identification of compounds

Identification of compounds was done by means of their retention time and final confirmations of compounds were done by matching their mass spectra (MS) with NIST 2005 Library data and literature. Quantitative analysis was done on the basis of its peak area measurement from the TIC without response factors.

3. Results

3.1 Rhizome volatile oils

The composition of the rhizome volatile oil has been investigated by combination of GC and GC-MS analysis. The volatile oils were obtained by conventional hydrodistillation technique. The oil yield was 3.25% (v/w). GC-MS identified a total of 24 compounds from the rhizome representing 77-91% of the total rhizome oils (Table 1; Figure 2, a). The major constituents in the rhizome oils of *C. angustifolia* were germacrone (27.52%), α-pinene (16.46%), α-elemene (15.32%), α-elemenone (8.42%), germacrene (5.68%) and caryophyllene (2.49%). The percentage composition of other trace constituents in the rhizome oils did not show much difference, as evident from Table 1.

Table 1: Volatile oil composition of rhizome and flower of *C. angustifolia* from North East India

Peak	Name of compound[a]	RT	Rhizome oil (%)	Flower oil (%)
1.	β-pinene	12.925	16.460	tr
2.	β-phellandrene	12.870	tr	tr
3.	Eucalyptol	15.191	-	0.121
4.	Camphore	20.323	tr	15.635
5.	α-bergamotene	25.080	tr	tr
6.	β-germacrene	25.135	5.681	22.442
7.	Acoradien	25.165	-	tr
8.	Cis-chrysanthemol	25.200	tr	tr
9.	Trimethyl- 1,5,9- cyclododecatriene	25.595	0.267	-
10.	β-elemene	25.780	1.301	-
11.	β-caryophyllene	26.336	2.486	6.844
12.	Ò-elemene	26.426	15.321	0.257
13.	β-farnesene	26.571	-	tr
14.	α-caryophyllene	27.406	2.486	0.456
15.	Germacrene D	28.116	tr	0.674
16.	α-farnesene	28.172	tr	tr
17.	α-funebrene	28.446	-	0.367
18.	Hexadeca- 1,11 diyne	28.827	0.023	-
19.	Isocericenin	29.077	-	0.782
20.	Curzerene	29.102	0.680	tr
21.	Petasitene	29.972	-	tr
22.	Caryophyllene oxide	31.753	tr	7.120
23.	β-elemenone	32.373	8.423	0.403
24.	Germacrone	32.393	27.519	4.034
25.	Methenolone	33.394	tr	tr

26.	Bicyclo (3.3.0) oct-1-en-3-one	33.598	-	0.391
27.	Phenol 4-((dimethylamino) sulfony) methylamino	34.409	0.076	-
28.	2- norbornanone	34.489	1.233	-
29.	Methyl isotridecanoate	38.035	-	0.724
30.	Methyl isomyristate	38.070	tr	5.230
31.	Methyl-11-methyl-dodecaonte	38.101	tr	tr
32.	Phthalic acid, isobutyl octadecylsilane	39.916	tr	tr
33.	1,3-benzodioxole,5-[1-[2-(2-butoxyethoxy) ethoxy]butyl]	39.946	-	tr
34.	Neocurdione	40.536	-	tr
35.	Butyl lauryl phthalate	42.082	-	tr
36.	Methyl 11,14-octadecadienoate	42.282	-	tr
37.	Methyl-9-cis-11-trans-octadecadienoate	42.302	-	tr
38.	Ethyl lenolenate	42.727	-	tr
39.	Isoquinoline, 1,2,3,4-tetrahydro-1-methyl	42.888	tr	1.279
40.	1- bromotricontane	43.588	-	tr
41.	α- guariene	44.748	0.594	-
42.	Fumaric acid, 2-dimethyl amino ethyl octadecyl ester	45.063	tr	0.254
43.	Trans- isolongifolene	45.183	0.112	tr
44.	10-chlorotricyclo[4.2.1.1(2,5)] Deca-3,7-Dion-9-ol	45.303	3.21	-
45.	Dimethyl 1,2,5-cycloheptatriene	45.353	0.092	-
46.	2-(3-isopropyl-4-methyl-pent-3-en-1-ynyl)-2-methyl-cyclobutanone	45.503	0.073	-
47.	9-acetyl-s-octahydrophenanthrene	45.618	0.995	-
48.	Stearyl iodide	46.644	tr	0.091
49.	Isoheneicosane	46.654	tr	0.082
50.	Di-isooctyladipate	47.184	-	0.091
51.	Bis (2-ethylhexyl) adipate	47.189	-	0.070
52.	Retinene	49.150	2.293	-
53.	Isoaromadendrene	50.750	0.795	-
54.	Methyl acetoacetate	50.955	0.466	tr
55.	Trans- longipinocarveol	51.059	0.754	tr
56.	Trans- retinal	52.896	0.710	-

[a] Compounds are listed in the order of elution on BP-1 column. t, trace (<0.05%).

Figure 1: ***C. angustifolia*** **: (a) Rhizome and (b) Flower.**

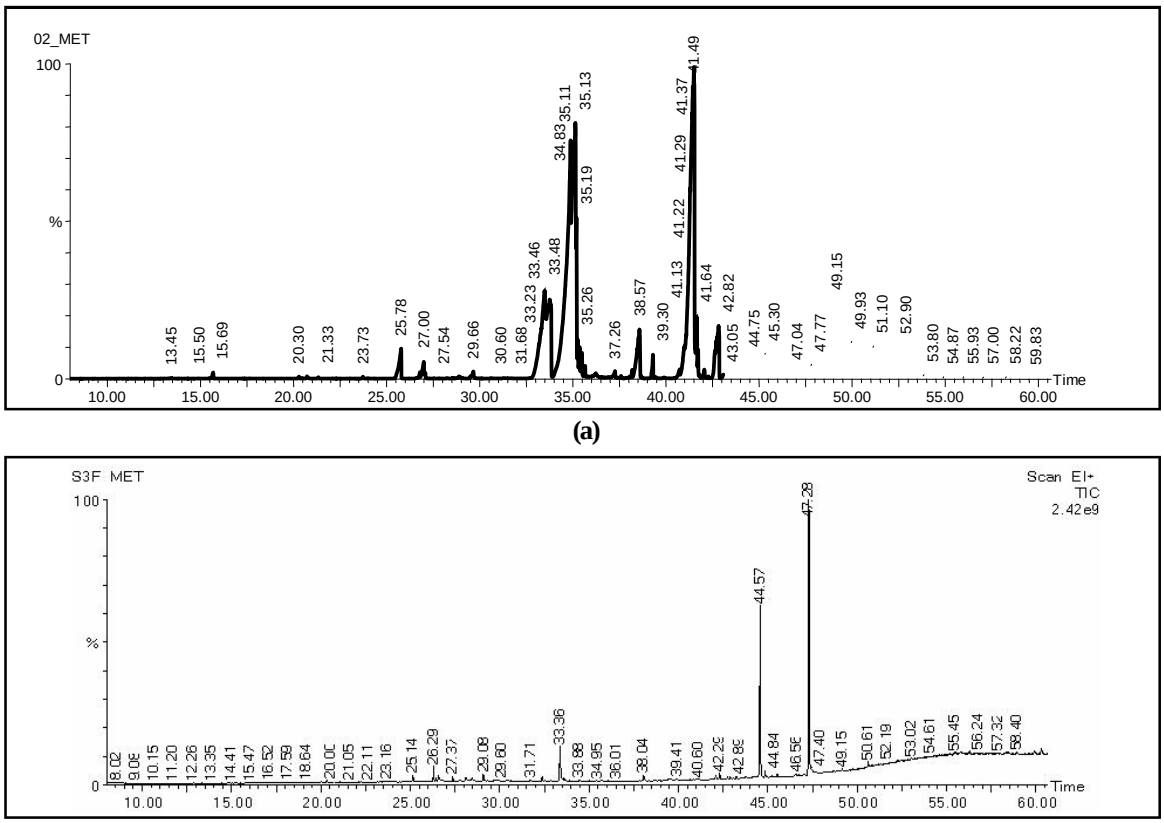

Figure 2: GC-MS chromatogram: (a) Rhizome and (b) Flower extracts of *C. angustifolia*.

3.2 Flower volatile oils

The composition of the flower volatile oil has been carried out by GC and GC-MS. The oil yield was 2.40% (v/w). Forty-two components were identified (Table 1; Figure 2, b) representing 46-61% of the total flower oils. β-germacrene (22.44%), 2,2,7,7-tetramethyl tricycle[6.2.1.0](1,6)]undeca-4-en-3-one (15.63%), caryophyllene oxide (7.12%), β-caryophyllene (6.84%), methyl-isomyristate (5.23%) and germacrone (4.03%) were the major constituents in the flower oils. The percentage composition of other trace constituents in the flower oils did not show much difference, as evident from Table 1.

4. Discussion

4.1 Qualitative and quantitative report

The essential oil of rhizome of *C. angustifolia* produced from North East India was compared to earlier reports. Rhizome oil of *C. angustifolia* showed to contain ar-curcumin, α-pinene, α-terpineol, camphor, zinbiberol, borneol (Banerjee *et al*., 1980), curzerenone (Nguyen *et al.*, 2001), furanodienone, isofuranodienone (Nguyen *et al.*, 2001), xanthorrhizol isomer, methyl eugenol, palmitic acid, germacron, isoborneol, curdione, 1,8-cineole (Srivastava *et al*., 2006; Thongkhwan *et al*., 2017). Young rhizome exhibited the presence of major compounds such as α-amorphene, camphor, 2,7-napthalenediol, trans-nerolidol, octadecanoic acid, butyl ester, humulen-6,7-epoxide (Nayak *et al*., 2014) which is not fully in support to our results. Presence of camphor, germacrene, β-elemene and β-pinene has also been found in some *Curcuma* species (Zwaving and Bos, 1992; Devi *et al*., 2012; Chane-Ming *et al*., 2003). Rhizomes of *C. angustifolia* obtained from central and southern India was also reported to be rich in camphor, germacrene, α-elemene and α-pinene (Srivastava *et al*., 2006). The presenr report was performed in methanol extracts of *C. angustifolia* rhizome and flower which revealed the presence of major phytochemicals. This result supported with previous reported studies on methanolic extracts of *Aegle marmelos* (Tiwari *et al*., 2016). The volatile oil of flower of many important medicinal and aromatic herbs had not been exploited. The flower essential oils of *Zingiber kerrii* Craib were reported to contain α-caryophyllene, α-pinene, α-pinene as dominant compounds (Aknerin *et al.*, 2020) which is in partial corroborate with our report for *C. angustifolia* flower. Variation in qualitative and quantitative data of essential oil in medicinal and aromatic herbs is affected by various environmental factors such as temperature, rainfall, humidity, plant nutrition, genetic variation, stress during maturity and geographical location (Raut and Karuppayil, 2014; Sangwan *et al*., 2001; Rahimmalek *et al*., 2013). Genetic pattern of the plant also affect the production of essential oil (Hassiotis *et al*., 2014).

4.2 Bioactiviy of methanolic extract of *C. angustifolia*

Germacrone which is major constituent of rhizome oil showed anti-inflammatory (Hossain *et. al*., 2015), antiandrogenic (Sephrom *et. al*., 2012) and antimicrobial (Kamazeri *et al*., 2012) activities. β-caryophyllene which is a major constituent in flower oil exhibited antitumor (Tisserand and Young, 2014), antileishmanial (Soares *et*

al., 2013) and antitrypanosomal (Izumi *et al*., 2012) properties. These phytocompounds present in rhizome and flower extracts of *C. angustifolia* has potent bioactivity and is useful for effective herbal formulations and preparation of drugs in pharmaceutical companies.

5. Conclusion

The chemical constituents in volatile oils distilled from rhizome and flower of *C. angustifolia* was investigated using GC-MS technique. The composition and quantitative data of the flower volatile oil of *C. angustifolia* are reported for the first time. These constituents could be used for the utilization of development of traditional medicines and investigation in the field of antioxidant and antimicrobial assay needs to be conducted. The present investigation has significantly contributed to the existing knowledge of volatile oil composition of methnolic extract of rhizome and flower of *C. angustifolia* found in North East India. The identified compounds can be utilized as pharmacognostical tools. The current study suggests that methanolic extract of this plant parts is a potent therapeutic agent. It paves the way for the development of several treatment regimens using the extracts and further research is necessary to identify and purify the active compounds responsible for therapeutic activities.

Author contributions

All the authors contributed to the concept and design of the experiments. N. Shilpia Devi executed the labwork while N. Sandhyarani Devi contributed in statistical analysis, interpretation of data and manuscript preparation. R.K. Imotomba Singh helped in conceptualization and support.

Conflict of interest

The authors declare no conflicts of interest relevant to this article.

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