Research Article

Comparative Phytochemical Analysis of *Curcuma aromatica, C. aeruginosa and C. angustifolia*

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Submission Date: 12-02-2024; Revision Date: 12-03-2024; Accepted Date: 15-04-2024.

ABSTRACT

Curcuma, species of Family Zingiberaceae valued economically due to medicinal and commercial properties ; three species of which viz., *Curcuma aromatica*, *C. aeruginosa* and *C. angustifolia* was analysed for phytochemical components. The specimens were collected from Wayanad, Kerala selecting one of the most fertile regions of the state. The rhizome was used for analysis using GC-MS method. Various components were identified that are of much significance in regard to medicinal properties. The study showed variations in the components within the species as compared with earlier works that point to geographical or related stress variations or can be accounted for development of chemotypes. The common phytoconstituents of the three species identified were camphor, germacrone and curzerene. A survey of the biological properties of these compounds specifically shows the three species can be synergistically utilised as ecofriendly weedicide or more particularly, *Curcuma angustifolia* that has high content of camphor, germacrone and curzerene with potential inhibitory properties.

Keywords: *Curcuma aromatica, C. aeruginosa, C. angustifolia,* GC-MS, Phytoconstituents.

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INTRODUCTION

The enhanced efficacy of plant derived drugs has increasingly manifolded the elucidation of phytochemical studies. The emerging researches in medicinal plants and their bioactive component identification has essentially become the thread of plant science. The family Zingiberaceae or the ginger family includes representatives from different parts of the world. The plant body has remarkable subterranean rhizomes.[1] The genus *Curcuma* L. belonging to the Zingiberaceae family possess several biotypes. The commonly used parts, the rhizome appear in colours varying from light to dark shades. Studies show the presence of specific components make the genus in India as the best in the world.[2] Diseases like Alzheimer's, cancer and heart problems is reported to be treated by the curcuminoid components.[3] *Curcuma* is commonly used for the rhizome which is source of curcumin and it serves as possessing preservative, medicinal and flavouring properties.[4] In this study, three different species of *Curcuma* were taken to consideration. *Curcuma aromatica* Salisb. has aromatic yellow rhizome and is native to south India and parts of eastern India. Wild turmeric botanically known as *Curcuma aromatica* Salisb. is recognized as medicinal herb with strong antibiotic properties; used as stomachic, carminative and emmenagogue remedies for skin diseases and for snake bites.[5] *Curcuma aeruginosa* Roxb. belonging to Zingiberaceae family with vernacular name 'pink and blue ginger'. Rhizome of *C. aeruginosa* Roxb. is reported to be used in tumor, bronchitis, asthma treatments and for intestinal disorders,[6] internally for infections and externally for wounds.[7] *Curcuma angustifolia* Roxb. can be identified by the elongated linear greenish shining leaves. The plant possesses relevant properties like antioxidant, anti-inflammatory, hepatoprotective, anticancerous activities.[8] As per the literature studies, more studies are conducted on the phytocomponent

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DOI: 10.5530/ajbls.2024.13.30

analysis of *C. aromatica* and *C. aeruginosa* when compared to *C. angustifolia*. Also, the analysis showed variations in the presence of varied components in same species based on geographical locations as well as within the genus. Therefore, a comparative analysis of phytocompounds in the three species of *Curcuma* was done by means of GC-MS analysis, which is one of the most widely used techniques to detect volatile, non-polar bioactive compounds.[9] The three plants, much grown in the district of Wayanad in Kerala is said to be one of the most fertile and climatically suited area for plant cultivation and this was also taken as a factor for collecting specimens from this area for study.

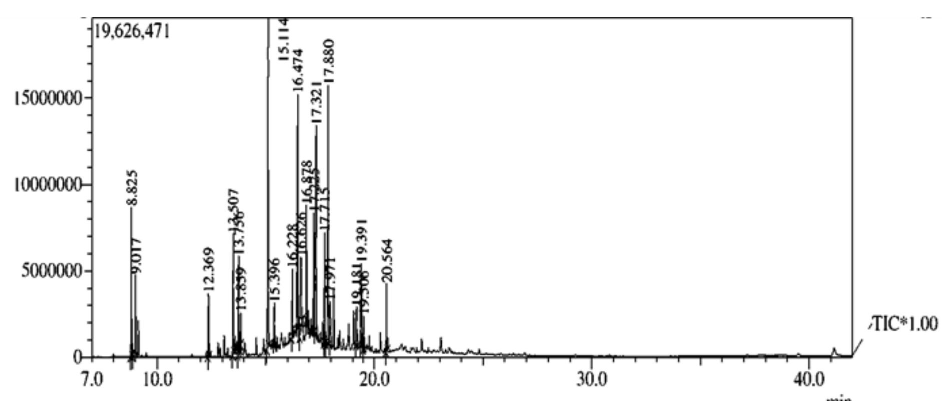
MATERIALS AND METHODS

Fresh rhizomes of *Curcuma aromatica, C.aeruginosa* and *C.angustifolia*, were collected from Wayanad. After the collection, each of the rhizomes were botanically identified properly on the basis of morphological features like leaf shape, flower color, rhizome color, etc. with the help of authentic literature and documentation with their characteristic features. The collected plant material was washed well to remove all the dirt and sliced into small pieces, kept for shade drying, later ground into fine powder used for study with hydro distillation and oil obtained was used with Shimadzu GC-MS of model Number: QP2010, Ultra operated in the Electron Impact (EI) mode (electron energy=70 eV), scan range=50-500 atomic mass units, scan speed=1000 and GC-MS solution software. The GC column was an ELITE-5MS column with a (5% phenyl)-polymethyl siloxane stationary phase and a film thickness of 0.25 μm, a length of 30 m and an internal diameter of 0.25 mm. The carrier gas was helium with a column head pressure of 57.5 k Pa and flow rate of 1.00 mL/min. The injector temperature was 260 and the ion source temperature was 200. The GC oven temperature was programmed for 60 initial temperatures and then temperature was increased to 260. The sample 0.1μL injected with a splitting mode into the GC inlet where it is vaporised and swept onto a chromatographic column by the carrier gas. The process used a mass analyser, which separates the positively charged ions according to various mass related properties. Major compounds were identified based on retention index, by reference to a homologous series of n-alkanes and the mass spectral fragmentation patterns were compared with reported literature and library. Rhizome analysis in three species of *Curcuma* was done in this manner and comparative investigations of the phytochemicals in these are done.

RESULTS

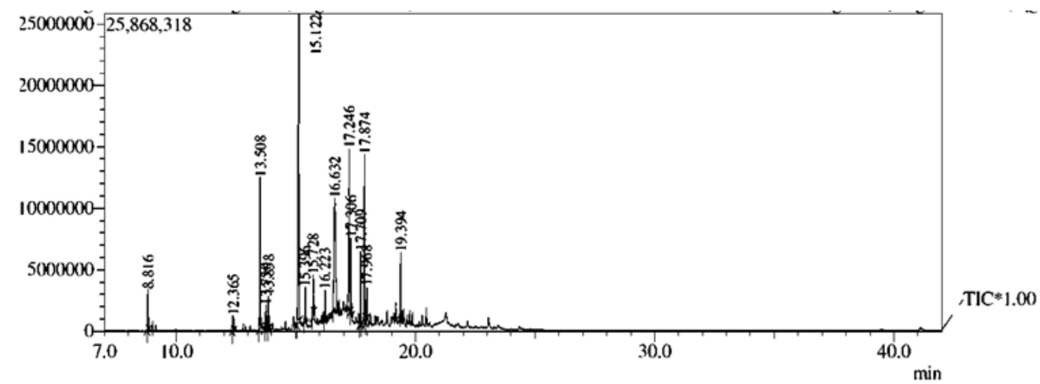
The GC-MS analysis of three species of *Curcuma* showed presence of different components, much similar as well as much varied. An analysis of the past studies along with the results obtained here, showed variations within the same species collected from different regions.

The phytochemicals screened from *C. aromatica* showed Camphor (5.09%), Isoborneol (2.63%), β-elemene (1.96%), ar-curcumene (3.89%), Curzerene (3.75%), β-curcumene (0.74%), 2,4-Di-spironorbornylcyclobuta-1,3-dione (ketene dimer) (14.17%), Spathulenol (1.44%), Germacrone (2.36%), Curdione (10.80%), Aristolene (5.73%), 3-Isopropyl-6,10-dimethyl-6-cyclodecene-1,4-dione (4.58%), Dodecatriene (5.13%), Isovelleral (11.25%), Elemene (3.75%), 1,4-Dimethyloctadiene (12.28%), 3-Methyl-5-(2,6,6-trimethyl-1-cyclohexen-1-yl)-1-pentyn-3-ol (2.22%) andrographolide (1.51%), Neoclovene (2.95%), 3-Ketomanoyl oxide (1.42%) and Diazoprogesterone (2.36%).



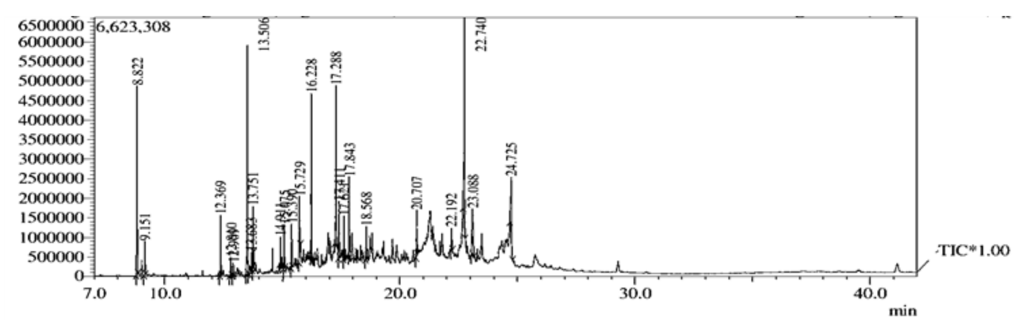
GC-MS chromatogram of *C. aromatica*

The phytochemicals screened from *C. aeruginosa* showed Camphor (2.09%), Elemene (0.75%), α-curcumene (7.87%), curzerene (1.24%), β-curcumene (1.48%), 2,4-Di-spironorbornylcyclobuta-1,3-dione (ketene dimers) (23.108%), 7-Oxabicyclo[4.1.0]heptane, 2,2,6-trimethyl-1-(3-methyl-1,3-butadienyl)-5-methylene (2.16%), ar-turmerone (1.94%), Germacrone (1.31%), Isocurcumenol (17.92%), Dihydrocarvone (14.34%), Valencene (3.97%), Elemene (4.14%), 1,4-dimethylocta-diene (11.65%), Propyl 7,10,13,16,19-docosapentaenoate (1.84%) and Megastigma-4,6(E),8(E)-triene (4.22%).



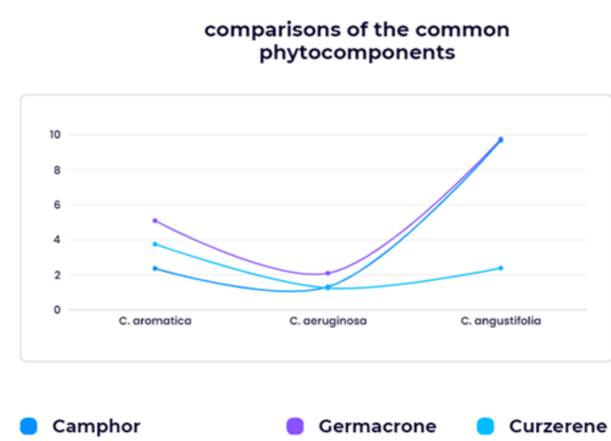
GC-MS chromatogram of *C. aeruginosa*

The phytochemicals identified in *C. angustifolia* showed Camphor (9.75%), Borneol (1.69%), β-elemene (2.82%), Caryophyllene (0.95%), Germacrene B (0.60%), Benzene, 1-(1,5-dimethyl-4-hexenyl)-4-methyl (11.61%), β-selinene (1.85%), Curzerene (2.38%), Caryophyllene oxide (1.47%), β-elemonene (2.24%), Spathulenol (2.16%), ar-turmerone (3.07%), Germacrone (9.67%), Isovelleral (10.05%), Trimethylbicyclo[4.1.0]hept-1-yl) ethyl vinyl acetate (3.07%), 3,7-Cyclodecadien-1-one (2.52%), 1,4-Dimethyloctadiene (4.03%), 1,5-cycloundecadiene, 8,8-dimethyl-9-methylene (2.08%), β-caryophyllene oxide (2.25%), Caryophyllene oxide (15.25%), 1-Heptatriacotanol (4.51%) and Bisabolene epoxide (5.41%).



GC-MS chromatogram of *C. angustifolia*

By comparing the phytocomponents from all the three rhizomes of the *Curcuma* species i.e., *Curcuma aromatica*, *C. aeruginosa* and *C. angustifolia*, three compounds were identified to be present in all three species under study. The common compounds present are the camphor, curzerene and germacrone. In *Curcuma aromatica* 5.09% camphor, 3.75% curzerene and Germacrone 2.36% are present. In *Curcuma aeruginosa* 2.09% camphor, 1.24% curzerene and Germacrone 1.31% are present. In *Curcuma angustifolia* 9.75% camphor, 2.38% curzerene and Germacrone 9.67% are present.



DISCUSSION

Curcuma species is grown in parts of world for diverse applications like culinary as well as medicinal utilities. The plant has shown intense biological and pharmacological properties that can be accounted for the presence of biocomponents. The objective of this work was to examine the variation in the phytochemical composition of *Curcuma* rhizome phytoconstituents. In the present study, the rhizomes of *C. aromatica, C. aeruginosa* and *C. angustifolia* were studied by GC-MS method.

The phytochemicals screened from *C. aromatica* showed Camphor, Isoborneol, β-elemene, ar-curcumene, Curzerene, β-curcumene, Spathulenol, Germacrone, Curdione, Aristolene, 3-Isopropyl-6,10-dimethyl-6-cyclodecene-1,4-dione, Dodecatriene, Isovelleral, Elemene, 1,4-Dimethyloctadiene, Methyl-5-(2,6,6-trimethyl-1-cyclohexen-1-yl)-1-pentyn-3-ol andrographolide, Neoclovene, 3-Ketomanoyl oxide, Diazoprogesterone and 2,4-Dispironorbornylcyclobuta-1,3-dione (ketene dimer) in the highest concentration. *C. aeruginosa* showed Camphor, Elemene, α-curcumene, curzerene, β-curcumene, 7-Oxabicyclo[4.1.0]heptane, 2,2,6-trimethyl-1-(3-methyl-1,3-butadienyl)-5-methylene, ar-turmerone, Germacrone, Isocurcumenol, Dihydrocarvone, Valencene, Elemene, 1,4-dimethyloctadiene, Propyl 7,10,13,16,19-docosapentaenoate, Megastigma-4,6(E),8(E)-triene and 2,4-Di-spironorbornylcyclobuta-1,3-dione (ketene dimers) in highest concentration. *C. angustifolia* showed Camphor, Borneol, β-elemene, Caryophyllene, Germacrene B, Benzene, 1-(1,5-dimethyl-4-hexenyl)-4-methyl, β-selinene, Curzerene, Caryophyllene oxide, β-elemonene, Spathulenol, ar-turmerone, Germacrone, Isovelleral, Trimethylbicyclo[4.1.0]hept-1-yl)ethyl vinyl acetate, 3,7-Cyclodecadien-1-one, 1,4-Dimethyloctadiene, 1,5-cycloundecadiene, 8,8-dimethyl-9-methylene, β-caryophyllene oxide, 1-Heptatriacotanol, Bisabolene epoxide and Caryophyllene oxide in highest concentration. By comparing the phytocomponents from all the three rhizomes of the *Curcuma* species i.e., *Curcuma aromatica*, *C. aeruginosa* and *C. angustifolia*, three compounds were identified to be present in all three species under study. The common compounds present are the camphor, curzerene and germacrone.

Rhizomes collected show variations in the components along with presence of similar compounds which can be accounted for their taxonomic position in the genus based on chemotaxonomy. But the data analysis of similar studies show variation in the component within the same species among different regions.

The Japanese *C. aromatica* oil was reported to have curdione (32.2-44.0%), 1,8-cineole (7.5-25.3%) and germacrone (4.6-9.6%),[10] while a sample from Thailand contained camphor (26.9%), *ar*-curcumene (23.2%) and xanthorrhizol (18.7%) as the main components.[11] Indian samples of *C. aromatica* had camphor (18.2-48.3%), β-curcumene (28.4-31.4%), *ar*-curcumene (22.1-24.1%), xanthorrhizol (4.8-16.2%), 1,8-cineole (5.5-15.9%), isoborneol (8.2-12.2%), curzerenone (5.5-11.0%), germacrone (4.9-10.6%), camphene (7.4-10.2%), curdione (4.8-8.0%), borneol (4.9-8.2%), β-elemene (7.5%), curzerene (4.6-6.0%), α-pinene (5.7-5.9%) and terpinolene (5.2%).[12] The composition of the camphor identified in the present study was 5.09%, β-curcumene (0.74%), ar-curcumene (3.89%), Germacrone (2.36%), Curdione (10.80%), Isoborneol (2.63%), β-elemene (1.96%), Curzerene (3.75%). These compounds were common but the composition showed detectable variations. Also, the sample *C. aromatica* in the present study showed the presence of new compounds which can be accounted for the change in geographical and climatic conditions leading to development of different chemotypes.

Likewise, *C. aeruginosa* also exhibited similar results. The samples of *C. aeruginosa* from Malaysia had curzerenone (24.6-30.4%), 1,8-cineole (11.2-25.2%), camphor (6.8-10.5%) and curcumenol (5.6%),[13] while from India the oil was dominated by curcumenol (38.7%) and β-pinene (27.5%).[12] *C. aeruginosa* oil sample from Thailand was dominated by curzerenone (41.6%) followed by 1,8-cineole (9.6%) and β-pinene (7.7%).[11] The oil sample analysed from the present study showed variations as compared to the Indian sample in having ketene dimer (23.10%) and 17% isocurcumenol as major with the composition of isocurcumenol varying in percentage from the reported Indian oil analysis.

A comparison of oil of *C. angustifolia* made from Central India and South India[14] showed major composition variation in the components with the former showing xanthorrhizol isomer (12.7%), methyl eugenol (10.5%), palmitic acid (5.2%) and camphor (4.2%) and the latter with germacrone (12.8%), camphor (12.3%), isoborneol (8.7%), curdinone (8.4%) and 1,8-cineole (4.8%). The present analysis of *C. angustifolia* showed benzene compound (13.30%), caryophyllene oxide (12.21%), camphor (10.97%), Germacrone (12.8%) collected from Kerala, South India. Even though some components are same, the composition is varying for some within India itself. Also, literature survey shows component variation in the studies reported earlier from Vietnam[15] all of which are in conformation with the present study.

The species show variations among themselves in different geographical regions and these can implied as the reason of varying biological activities of these species in different locations.[16]

Also, the three components commonly observed in the three different species under the present study viz., Camphor, Germacrene and Curzerene have marked significance in the medicinal aspect as Camphor is used as insecticidal, antimicrobial, antiviral, anticancer agent; Germacrone acts as CNS depressant, anti-inflammatory, antiulcer, antifeedant, antibacterial, antifungal, antitumor and Curzerene has anticancer properties along with phytotoxic properties as per the studies indicated by the effect of phytotoxins on wheat coleoptile assay.[17] The study revealed the capability of species of *Curcuma* in inhibiting the germination of wheat coleoptiles may be due to the presence of Camphor, Germacrene and Curzerene which was detected in the all the three species under study. This property can be utilised as potential bioherbicide.

In the present study, the graph shows the largest concentration of these components in *Curcuma angustifolia*. So, either singly or the synergistic action of the three species can be utilised for the ecofriendly weedicide applications along with the medicinal properties of the species.

The present study has utilised the mature rhizomes of three species cultivated in the same season so that the factors influencing the metabolite development are the same and so the content comparison is of significant value. The variation in these species can be identified more if the species are consecutively studied in different developmental stages and further on to the varying biological activity of each according to the components.

CONCLUSION

Three species of *Curcuma* rhizomes were collected from different parts of the Wayanad, Kerala. The three *Curcuma* species taken were *Curcuma aromatica, Curcuma aeruginosa* and *Curcuma angustifolia*. The rhizomes were shade dried and powdered and oil extraction used for phytochemical study. In *Curcuma aromatica,* 21 components with 2, 4-Di-spironorbornylcyclobuta-1, 3-dione (ketene dimers) in high concentration; in *Curcuma aeruginosa* 16 compounds identified with 2, 4-Di-spironorbornylcyclobuta-1,3-dione (ketene dimers) in high concentration; in *Curcuma angustifolia* 23 compounds with caryophyllene oxide in high concentration was analysed. The phytocomposition of the species under study with earlier literature analysis showed variations which can be accounted for geographical or as chemotypes that has to be clarified by more studies. The common occurrence of components in the three species also point to the synergistic utilization of these species as biopesticides.

ETHICAL APPROVAL

The submitted work is original and not placed for publication in any journal. It has not utilised any animal for study or pose threat to nature. Also, no external fund was utilised for the study and no additional contributors have been involved in the study.

DECLARATION OF CONFLICTING INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

GC-MS: Gas Chromatography-Mass Spectrometry.

SUMMARY

Curcuma genus well known for the medicinal properties in India has several cultivated species and three species of the genus viz., *C. aromatica*, *C. aeruginosa* and *C. angustifolia* was studied for the phytocomponents and the utilisation based on component analysis. The fresh rhizomes collected were used for GC-MS analysis. The three species showed three components in common, viz., camphor, germacrone and curzerene. The study in comparison with similar species showed variable results in different geographical locations based on reported literatures. Also, the analysis revealed the plausible utilisation of the three species together or *C. angustifolia* as potential bioherbicide which can be further refined for commercialization.

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Cite this article: Kundamveetil S. Comparative Phytochemical Analysis of *Curcuma aromatica*, *C. aeruginosa* and *C. angustifolia*. Asian J Biol Life Sci. 2024;13(1):235-40.