



Newsletter • 2012 • vol.3 • 24 - 29

ESSENTIAL OIL CHEMICAL COMPOSITION OF CROTON OVALIFOLIUS VAHL FROM PARAGUANÁ - VENEZUELA

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Abstract

The essential oil from fresh leaves of *Croton ovalifolius* Vahl was obtained by hydrodistillation and analysed by GC/MS. Constituents were identified by their mass spectra and Kovats' indexes. Forty five compounds, which represent 88.24% of the total constituents of the oil, were identified of the oil obtained in a yield of 0.14% ($^{w}/_{m}$). The most abundant constituents were β -caryophyllene (19.49 %), germacrene D (17.62 %), β -myrcene (11.71%), germacrene B (11.68%) and α -pinene (6.33%).

Keywords: Euphorbiaceae, Croton ovalifolius, Essential oil, GC-MS, β -Caryophyllene, Germacrene D.

This paper was presented by the author at the XXI Congress of Italo-Latin American Society of Ethnomedicine (SILAE), Paestum, Italy, 25-29 September 2012

Introduction

Within families of plants that have long had an important role in traditional medicine in Africa, Asia and South America is the Euphorbiaceae, which is well known around the world as toxic as well as medicinal, it also presents a wide range of effects. The genus Croton, belonging to this family, consists of herbs, shrubs, vines and trees, it comprises about 1300 species widespread in warm regions of the planet, especially in South America and Africa; it is the largest genus of Crotonoideae subfamily. In Venezuela there are 80 species [1]. Several genus species are aromatic, indicating volatile constituents' presence.

One of the Croton species found in Venezuela is C. ovalifolius, a small aromatic shrub known as "amorgosito" or "matejea" [2], a native of the West Indies, southern Mexico, and northern Venezuela and Colombia. In Venezuela, the C. ovalifolius is found Lara, Falcón, Mérida and around Caracas [3], being widely used in traditional medicine, the leaves and roots are used in tea to treat respiratory problems as well as are used by producers of goats when the animals have retained placenta. To date studies of this species are few, there are no reported phytochemical or pharmacological studies. The composition of the essential oil has been reported for the species collected in the town of Chiguará, in Mérida - Venezuela [4].

This paper presents a study of essential oil chemical composition extracted from leaves of *C. ovalifolius* from Paraguaná, Falcón state.

Materials and methods

Plant material

Leaves of plants were recollected when *C. ovalifolius* bloomed in Charaima town, located at east of the Paraguana Peninsula, during mid November 2011. Taxonomic identification information and voucher specimen (voucher CR-07) is sheltered in MERF Herbarium (Faculty of Pharmacy, University of the Andes).

Essential oil Obtaining

1300 g of freshly harvested leaves from under study plant was minced with 4 litters of water using a mechanical mixer and subjected to a ball hydrodistillation of 12 litters for three hours. Oil was collected in a Clevenger trap, and then it was drying over anhydrous sodium sulphate which was stored under refrigeration for further analysis.

Oil Analysis

The essential oil chemical composition was analysed by gas chromatography-mass spectrometry (GC-MS). The equipment used was a HP6890 chromatograph coupled to a HP 5973 mass detector equipped with an automatic injector and a HP-ALS silica capillary column HP-5MS of phenyl (5%)-methyl (95%)-polysiloxane of 30 m long with an internal diameter of 0.25 mm and a film thickness of 0.25 µm. The ionization energy of 70 eV was used. The oven temperature was programmed as follows: initial temperature 60 °C, final temperature 260 °C, and heating rate of 4 °C/minute. The total analysis time was 50 minutes. The injector temperature was 200 °C. Helium was used as carrier gas at a rate of o.8 mL/min, at constant volume. 1.0 µL was injected from a 2% solution of oil dissolved in diethyl ether. We applied a cast of 100:1.

Quantification of essential oil components studied was established based on the relative areas of each peak, according the chromatographic criterion. Components identification was done by computed mass spectra comparison with Wiley library (6th Ed.). Components identification was confirmed by Kovats's indexes, which are interpreted by comparing the calculated retention values with the values published in literature [5,6].

Results

The yield of essential oil obtained from the fresh leaves of C. *ovalifolius* from Paraguaná was 0.14% ($^{v}/_{m}$). The chemical constituents identified of the oil obtained are shown in Table 1. In total it was identi-

fied 88.24% of constituents (45 of 51) what represents 96.88% of essential oil mass. Sesquiterpenes accounted for 58.82% of the oil extracted from leaves, while monoterpenes accounted for 23.53%. The main components of essential oil are: θ caryophyllene (19.49%), germacrene D (17.62%), θ myrcene (11.71%), germacrene B (11.68%) and α pinene (6.33%).

see Table 1.

Discussion

The oil composition studied, significantly differs from the oil of the same species collected in Chiguará, Mérida [4], since in this 28 components accounted for 90% of oil and are absent some of the major components of the oil collected in Paraguaná as germacrene-D, α -pinene and β -myrcene. These results indicate that C. ovalifolius from Paraguaná requires biosynthesize a higher proportion of volatile secondary metabolites compared with the species from Chiguará, Mérida, what suggests that the plant from Paraguaná is subject to an increased environmental stress and needs this type of compounds as a defence mechanism against potential predators or other ecological functions [7,8]. Also, the results of the analysis of essential oils leaves which contains a greater number of Croton spp. reported in the literature show significant qualitative and quantitative differences in oils composition, however the β -caryophyllene, germacrene D and *á*-pinene, often appear as main constituents of the essential oil of Croton several species (see table 2).

Note that some of the major substances constituting the oil studied exhibit important biological activities, such as θ -caryophyllene with spasmolytic [9] activity, local anesthetic [10] and antiinflammatory [11] and θ -myrcene, which is an analgesic substance [12,13] with antioxidant activity [14], therefore *C. ovalifolius* essential oil could present similar biological activities.

Acknowledgements

Authors would like to thank forest engineer Juan Carmona (MERF Herbarium, Facultad de Farmacia y Bioanálisis, Unidersidad de Los Andes) for botanical material identification.

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see Table 2.

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| Compound | RT | % | KI cal |
|----------------------------------|--------|-----------|---------|
| 3-hexen-1-ol | 3,675 | 0,7 | 848,27 |
| a-pinene | 5,089 | 6,33 | 935,39 |
| camphene | 5,41 | 0,54 | 950,50 |
| β-pinene | 6,039 | 1,25 | 977,68 |
| β-myrcene | 6,039 | 11,71 | 977,68 |
| 2-methylbutyl-2-methylpropionate | 6,942 | 0,1 | 1013,92 |
| <i>p</i> -cymene | 7,212 | 0,13 | 1013,92 |
| limonene | 7,326 | 0,91 | 1029,21 |
| trans-β-ocimene | 7,824 | 0,12 | 1047,88 |
| | 8,153 | 0,12 | 1059,58 |
| γ-terpinene linalool | 9,331 | 0,19 | 1097,45 |
| 2-methylbutyl 2-methylbutanoate | 9,449 | 0,42 | 1101,80 |
| bomeol | 11,412 | 0,42 | 1167,11 |
| myrtenol | 12,396 | 0,08 | 1195,73 |
| Endobomil acetate | | | |
| a-cubebene | 15,253 | 0,12 0,09 | 1267,49 |
| | 17,288 | | 1354,18 |
| a-copaene | 18,132 | 1.41 | 1377,97 |
| β-bourbonene | 18,415 | 0,88 | 1385,70 |
| β-cubebene | 18,58 | 0.56 | 1390,16 |
| β-elemene | 18,647 | 4,04 | 1391,95 |
| β-caryophyllene | 19,58 | 19,49 | 1421,98 |
| γ-elemene | 19,909 | 0,42 | 1433,18 |
| trans-a-Bergamoteme | 19,972 | 0,35 | 1435,30 |
| a-patchoulene | 20,39 | 0,13 | 1449,23 |
| a-humulene | 20,563 | 5,23 | 1454,90 |
| alloaromadendrene | 20,758 | 0,72 | 1461,25 |
| γ- <u>muurolene</u> | 21,281 | 0,72 | 1477,97 |
| germacrene D | 21,458 | 17,62 | 1483,54 |
| β -selinene | 21,564 | 0,99 | 1486,85 |
| Bicliclogermacrene | 21,851 | 2,46 | 1495,74 |
| a-muurolene | 21,956 | 0,23 | 1498,96 |
| germacrene A | 22,125 | 2,52 | 1504,91 |
| γ-cadinene | 22,357 | 0,58 | 1513,28 |
| δ-cadinene | 22,628 | 0,81 | 1522,94 |
| selina-3,7(11)-diene | 23,172 | 0,29 | 1541,99 |
| elemol | 23,404 | 0,17 | 1549,98 |
| germacrene B | 23,687 | 11,68 | 1559,62 |
| germacrene D-4-ol | 24,156 | 0,52 | 1575,35 |
| spathulenol | 24,215 | 0,17 | 1577,30 |
| caryophyllene oxide | 24,379 | 1,21 | 1582,72 |
| 1,2-epoxy-humulene | 25,118 | 0,1 | 1607,89 |
| τ-cadinol | 25,992 | 0,66 | 1640,33 |
| δ-cadinol | 26,144 | 0,13 | 1645,86 |
| a-cadinol | 26,376 | 0,49 | 1654,23 |
| Juniper camphor | 27,52 | 0,12 | 1694,50 |
| monoterpene Hydrocarbons (%) | | 15,69 | 23,53 |
| Oxygenated monoterpenes (%) | | 7,84 | |
| Sesquiterpene Hydrocarbons (%) | | 43,14 | 58,82 |
| Oxygenated sesquiterpenes (%) | | 15,69 | |
| Others (%) | | 5,88 | |
| Total Identified (%) | | 88,24 | 45 |

Table 1. Relative abundance of the compounds present in the essential oil of C. ovalifolius from Paraguaná.

| Species | Main Compounds | % | Ref. | Species | Main Compounds | % | Ref. |
|-------------------|-------------------------------|-------|------|-----------------|-------------------------|------|------|
| C. adenocalyx | α-pipene | 32.62 | 15 | C. heterocalix | germacrene-D | 12.5 | 22 |
| | bicyclogermacrene | 13.96 | | | bicyclogermacrene | 11.2 | |
| | β -caryophyllene | 10.23 | | | δ -elemene | 9.2 | |
| | germacrene-D | 10.14 | | | β -elemene | 8.2 | |
| C. bogotanus | limonene | 55.2 | 16 | C. jimenezil | methyleugenol | 29.5 | 23 |
| | safrole | 11.3 | | | germacrene-D | 15.6 | |
| | | | | | β -caryophyllene | 12.9 | |
| C. cajucara | linalool | 41.2 | 17 | | | | |
| | (E)-nerolidol | 12.6 | | C. lanjouwensis | α -pinene | 26.6 | 24 |
| | $\beta\text{-}$ caryophyllene | 6.9 | | | α -phellandrene | 8.5 | |
| C. campestris | caryophyllene oxide | 29.9 | 18 | C. palanostigma | linalool | 25.4 | 25 |
| | humulene oxide II | 8.0 | | | β -caryophyllene | 21.0 | |
| | | | | | metilleugenol | 17.2 | |
| C. flavens | viridiflorene | 12.22 | 19 | | β -elemeno | 6.0 | |
| | germacrene | 5.27 | | | | | |
| | (E) - γ -bisabolene | 5.25 | | C. sacaquinha | β -elemene | 12.0 | 26 |
| | | | | | germacrene D | 10.1 | |
| C. gossypiifolius | α-cedrene oxide | 18.6 | 20 | | linalool | 5.8 | |
| | spathulenol | 16.3 | | | β -caryophyllene | 5.7 | |
| | valencene | 5.8 | | | | | |
| | geranyl-pentanoate | 5.3 | | C. sonderianus | β -phelandreno | 20.4 | 27 |
| | | | | | bicyclogermacrene | 29.1 | 21 |
| C. grewioides | (E)-anethole | 65.5 | 21 | | β -elemene | 17.8 | |
| | methyleugenol | 10.6 | | | | | |
| | (Z)-anethole | 4.6 | | C. zambesicus | caryophyllene oxide | 19.5 | 28 |
| | (E)-methyl isoeugenol | 4.7 | | | β - caryophyllene | 10.8 | |
| | 9993 9999 9999 | | | | a-copaene | 6.3 | |

Table 2. Main compounds of essential oils of leaves of Croton spp. in previous research.