AkiNik

American Journal of Essential Oils and Natural Products

Available online at www.essencejournal.com

AJEONP

American Journal of Essential Oils and Natural Products

ISSN: 2321 9114
AJEONP 2016; 4(4): 01-03

Received: 01-08-2016
Accepted: 02-09-2016

Olateju A Dawodu
Natural Product Research Unit, Department of Chemistry, Lagos State University, Badagry Expressway, PMB 0001 LASU Post Office, Ojo, Lagos, Nigeria

Oladipupo A Lawal
Natural Product Research Unit, Department of Chemistry, Lagos State University, Badagry Expressway, PMB 0001 LASU Post Office, Ojo, Lagos, Nigeria

Isiaka A Ogunwande
Natural Product Research Unit, Department of Chemistry, Lagos State University, Badagry Expressway, PMB 0001 LASU Post Office, Ojo, Lagos, Nigeria

Abdulateef A Giwa
Natural Product Research Unit, Department of Chemistry, Lagos State University, Badagry Expressway, PMB 0001 LASU Post Office, Ojo, Lagos, Nigeria

Correspondence
Isiaka A Ogunwande
Natural Product Research Unit, Department of Chemistry, Lagos State University, Badagry Expressway, PMB 0001 LASU Post Office, Ojo, Lagos, Nigeria

Volatile constituents of *Crescentia cujete* L

Olateju A Dawodu, Oladipupo A Lawal, Isiaka A Ogunwande and Abdulateef A Giwa

Abstract
The result of chemical investigation on essential oil obtained by hydrodistillation of the leaf of *Crescentia cujete* L. (family Bignoniaceae) is being reported. The essential oil was analysed by using gas chromatography (GC) and gas chromatography coupled with mass spectrometry (GC-MS). A total of 12 compounds accounting for 98.2% of the total oil contents were identified in the oil sample. The major constituents of the essential oil were kaur-16-ene (33.6%) and phytol (29.9%), along with significant quantity of *trans*-pinane (8.3%) and hexadecanal (4.6%). Other compounds such as (*Z*)-9, 17-octadecadienal (3.4%), neophytadiene (2.3%), selina-4(15), 6-diene (1.2%) and *allo*-aromadendrene (1.0%) were also identified in amount ≥ 1%.

Keywords: *Crescentia cujete*, GC, GC-MS, kaur-16-ene, phytol

1. Introduction
Calabash tree (*Crescentia cujete* L), is a small tree of the family Bignoniaceae, 6 to 12 m tall that grows as an ornamental plant. However, *C. cujete* is also used in traditional systems of medicine. The spoon-shaped leaves (5 to 18 cm long and 2 to 5 cm broad) are arranged in clusters along the stout twigs. It produces light green bell-shaped flowers (5 to 6.5 cm long). The very large and oval green or brown fruits which are 10 to 30 cm in diameter resemble gourds [1]. Extracts from the various parts of *C. cujete* possessed anti-inflammatory [2], antibacterial [2-4], DPPH radical scavenging [5], antioxidant [5, 6, 7], cytotoxic [5], anti-venom [8], CNS depressant [9], wound healing [10] activities. Research shows that the higher concentrations of the ethanolic fruit extract of *C. cujete* can alter the growth and development of the fetus and alters the blood profile of the maternal rats. Therefore, with utmost caution, should be advised in the use of this extracts during pregnancy in human [11].
The phytochemical compounds isolated from *C. cujete* includes acanthoside D, β-D-glucopyransoyl benzoate, (2*R*,4*S*)-2,4-pentanediol, (*R*)-4-hydroxy-2-pentanone and (*R*)-1,3-octanediol [12], 3-hydroxymethylfuro[3,2-b]naphtho[2,3-d]furan-5,10-dione and 9-hydroxy-3-hydroxymethylfuro[3,2-b]naphtho[2,3-d]furan-5,10-dione[13], 3-hydroxyoctanol glycosides, 2,4-pentanediol glycosides, 4-hydoroxy-2-pentanone glycosides, ajugol, 6-Op-hydroxybenzoylajugol, aucubin 6 - Op - hydroxybenzoyl - 6 - epi - aucubin, 1 - dehydroxy - 3, 4-dihydroaucubigenin, acanthoside D, benzoic acid glucosyl ester and 5 - hydroxymethylfurfural [14]. Other compounds such as 6-*O*-*p*-hydroxybenzoyl-10-deoxyeucommiol, 6-*O*-benzoyl-10-deoxyeucommiol, 6-*O*-benzoyl-dihydrocatalpolgeninningpogenin, and 6-*O*-*p*-hydroxybenzoylaucubin were isolated from *C. cujete* [15]. Crescentins I–V, crescentosides A, B and C, ajugol, 6-*O*-*p*-hydroxybenzoylajugol, aucubin, 6-*O*-*p*-hydroxybenzoyl-6-epiaucubin, agnuside, ningpogenin and 5, 7-bisdeoxycynanchoside were also characterized from the plant [16]. Included in the compounds were ningpogenin, 6-O-p-hydroxylbenzoylaucubin,3,3′-bisdemethylpinoresinol, (22E,24R)-ergosta-7,22-dien-3β-ol, ergosta-4,6,8(14),22-dien-3-one, cerevisterol, 5α,8α-epidiory-(22E,24R)-ergosta-6,22-dien-3β-ol, β-sitosterol, daucosterol, 3β,5α,9α-trihydroxyergosta-7,22-dien-6-one, ergosta-7,22-dien-3-one, sesquiterpene, 4-hydroxybenzonicacid, benzoicacid, p-hydroxybenzylethanol, p-hydroxybenzylalcohol, D-allitol and 5-hydroxymethyl-2-furancarboxaldehyde [17].
A previous study on the essential oil of *C. cujete* revealed that polysulfides such as dimethyl disulphide (10.8%), dimethyl trisulfide (28.5%) and dimethyltetrasulphide (36.7%) were the main constituents of the studied oil sample [18]. Limonene (16.7%) was the only terpene identified in significant quantity in the oil sample [18].

The aim of the present paper was to report to the volatile compounds identified in the essential oil of *C. cujete* growing in Nigeria.

2. Materials and methods

2.1 Plant material

Fresh leaves of *C. cujete* were collected from plants growing at National Museum and Monuments, Ile-Ife, Osun State, Nigeria, in June 2016. The identification of the plant material was confirmed by Curators at the Herbarium of the Department of Botany, University of Lagos, Nigeria, where a voucher specimen (LUH-7124) was deposited.

2.2 Hydro distillation of essential oil

Air-dried and pulverized leaves (100.0 g) were subjected to hydro distillation in a Clevenger-type apparatus for 4 h in accordance with the British Pharmacopoeia specification [19]. The distilled oil was preserved in a sealed sample tube and stored under refrigeration at 4ºC until analysis.

2.3 Analysis of essential oil

Gas chromatography (GC) analysis was carried out on a Hewlett Packard Gas Chromatography HP 6820 equipped with FID detector and HP-5MS column (60m x 0.25mm id), 0.25 µm film thickness and split ratio of 1:25. The oven temperature was programmed from 50 ºC (after 2 min) to 240 ºC at 5 ºC/ min and the final temperature was held for 10 min. Injection and detector temperatures were 200 ºC and 240 ºC respectively. Hydrogen was the carrier gas at flow rate of 1 mL/min. 0.5 µl of the diluted oil was injected into the GC. Peaks were measured by electronic integration. *n*-Alkanes were run at the same condition for retention indices determination.

Gas chromatography-mass spectrometry (GC-MS) was performed on a Hewlett Packard Gas Chromatography HP 6890 interfaced with Hewlett Packard 5973 mass spectrometer system equipped with a HP-5MS capillary column (30m x 0.25 mm id, film thickness 0.25 µm). The oven temperature was programmed from 70-240 ºC at the rate of 5 ºC/min. The ion source was set at 240 ºC and electron ionization at 70 eV. Helium was used as the carrier gas at a flow rate of 1 mL/min. Scanning range was 35 to 425 amu. 1.0 µL of diluted oil in hexane was injected into the GC/MS.

The identity of the oil components were assigned by comparison of their retention indices with the authentic samples and matching of their mass spectra with the NIST [20] library mass spectra database as well as with published data.

3. Results & Discussion

Hydro distillation of the dried leaves of *C. cujete* offered pale yellow essential oil in yield of 0.70% (w/w) calculated on a dry weight basis. The compositions of the oil were presented in Table 1, where all compounds are listed according to their elution from a HP-5MS column. The GC chromatogram shows the presence of fourteen volatile compounds of which twelve were identified from the GC-MS, accounting for 98.2% of the total compounds. Diterpenes (75.8%) represents the main class of compound present in the oil. The main constituents of the oil of *C. cujete* were kaur-16-ene (33.6%) and phytol (29.9%). Other significant components of the oil included *trans*-pinane (8.3%), hexadecanal (4.6%), (*Z*)-9,17-octadecadienal (3.4%) and neophytadiene (2.3%).

A comparison of the present oil composition with previous study [18] indicated dimethyl disulphide, dimethyl trisulfide, dimethyltetrasulphide and limonene that were present in previous study were conspicuously absent in the present investigated oil sample. The previous study was a headspace analysis of floral volatiles, so it is not surprising that the composition should be different from hydrodistilled leaf oil. Moreover, the main compounds present in the present study namely kaur-16-ene, phytol, *trans*-pinane, hexadecanal, (*Z*)-9, 17-octadecadienal and neophytadiene were not reported previously to be constituents of *C. cujete* oil.

The biological activity of an essential oil may depend on potency of the major compounds or a synergy between the major and some minor constituents. For example, phytol, one of the major constituents of *C. cujete* has various biological effects, such as peroxisome proliferation in skin sebaceous glands [21], anti-inflammatory [22], anti-acetyl cholinesterase [23], antischistosomal [24], antimicrobial [25], antinociceptive [26], antioxidant [26] and cytotoxicity [27]. Further research aimed at the determination of the biological activity and active compounds of *C. cujete* is in progress.

Table 1: Chemical constituents of essential oil of *Crescentia cujete*

Compounds [a]	RI [b]	RI [c]	Percent composition
Hexadecane	550	550	0.5
1,1-Dimethyl -3-hexyl- Cyclopentane	685	690	0.7
4-Methyl-2-heptanone	950	949	0.3
trans-Pinane	980	972	8.3
Selina-4(15),6-diene	1447	1449	1.2
allo-Aromadendrene	1470	1467	1.0
Globulol	1580	1578	0.4
Neophytadiene	1845	1840	2.3
Hexadecanal	1830	1822	4.6
Kaur-16-ene	2044	2041	33.6
Phytol	2119	2129	29.9
(*Z*)-9,17-Octadecadienal	2300	2297	3.4
Total			98.2
Monoterpene hydrocarbons			8.3
Sesquiterpene hydrocarbons			2.2
Oxygenated sesquiterpenes			0.4
Diterpenes			75.8
Hydrocarbons			1.5
Fatty acids			10.0

[a] Elution order on HP-5MS column; [b] Retention indices on HP-5MS column; [c] Literature retention indices

4. Conclusions

The chemical constituents of essential oil of *C. cujete* are being reported. The oil contained large amount of diterpene compounds. Due to the non-availability of literature citation, the results could not compare with previous study on the essential oil of the plant.

5. Acknowledgments

Dawodu AO is grateful to Ismail Raji for the assistance in the hydro distillation of oil sample.

6. References

1. Arango-Ulloa J, Bohorquez A, Duque MC, Maass BL. Diversity of the calabash tree (*Crescentia cujete* L.) in Colombia. Agroforestry Systems, 2009; 76(3):543-553.
2. Parvin MS, Das N, Jahan N, Akhter MA, Nahar L, Islam ME. Evaluation of in vitro anti-inflammatory and antibacterial potential of *Crescentia cujete* leaves and stem bark. BMC Res Notes, 2015; 8:412. doi: 10.1186/s13104-015-1384-5.
3. Mahbub KR, Hoq MM, Ahmed MM, Sarker A. *In vitro* antibacterial activity of *Crescentia cujete* and *Moringa oleifera*. Bangladesh Research Publication Journal. 2011; 5(3):337-343.
4. Agarwal M, Chauhan S. Anti-mycobaterial potential of *Crescentia cujete* (Bignoniaceae). International Journal of Advanced Research in Botany. 2015; 1(1):1-9
5. Juceni PD, Marilena M, Jorge MD, Hugo NB, Alexsandro B, Fátima MA *et al*. Radical scavenging, antioxidant and cytotoxic activity of Brazilian Caatinga plants. Fitoterapia. 2007; 78(2):215-218.
6. Das N, Islam ME, Jahan N, Islam MS, Khan A, Islam MR. Antioxidant activities of ethanol extracts and fractions of *Crescentia cujete* leaves and stem bark and the involvement of phenolic compounds. BMC Complementary Alternative Medicine, 2014; 14:45. doi: 10.1186/1472-6882-14-45.
7. Akinmoladun AC, Obuotor EM, Farombi EO. Evaluation of antioxidant and free radical scavenging capacities of some Nigerian indigenous medicinal plants. Journal of Medicinal Food. 2010; 13(2):444-451.
8. Shastry CS, Aswathanarayana BJ, Bhalodia Maulik M. Anti-venom activity of ethanolic extract of *Crescentia cujete* fruit. International Journal of Phytomedicine. 2012; 4(1):108-114
9. Aderibigbe AO, Olufunmilayo T, Agboola CNSO. Depressant properties of the crude extract of *Crescentia cujete* in mice. Planta Medica, 2013; 79(13):423-427.
10. Campos GS, de Oliveira Jr SA, Borges LBP, Ribeiro IP, Martinez SB, Ayer IM *et al*. Use of extract of coite (*Crescentia cujete*) as a phytotherapic in injuries of horses. Investigação, 2016; 15(4):95-97.
11. Almadin FJF, Jumawan JC. Preliminary study of the effects of calabash (*Crescentia cujete*) ethanolic fruit extract to gestating sprague dawley rats. International Journal of Technical Research and Applications. 2015; 19(1):1-4.
12. Kaneko T, Ohtani K, Kasai R, Yamasaki K, Nguyen MD. *n*-Alkyl glycosides and *p*-hydroxybenzoyloxy glucose from fruits of *Crescentia cujete.* Phytochemistry, 1998; 47(2):259-263.
13. Heltzel CE, Gunatilaka AAL, Glass TE, Kingston DGI, Hoffmann G, Johnson RK. Bioactive furanonaphthoquinones from *Crescentia cujete*. Journal of Natural Product. 1993; 56(9):1500-1505.
14. Kaneko K, Tetsuo K, Ohtani K, Kasai R, Yamasaki K, Thoi NN. Iridoids and other glycosides from Vietnamese *Crescentia cujete*. Natural Organic Compounds, 1996; 38(3):331-336.
15. Wang G, Yin W, Zhou ZY, Hsieh KL, Liu JK. New iridoids from the fruits of *Crescentia cujete.* Asian Natural Products Research, 2010; 12(9):770-775.
16. Kaneko T, Ohtani K, Kasai R, Yamasaki K, Nguyen MD. Iridoids and iridoid glucosides from fruits of *Crescentia cujete*. Phytochemistry, 1997; 46(5):907-911.
17. Yin W, Wu PY, Liang YM, Liu JS, Wang G. Chemical constituents of sarcocarp of *Crescentia cujete*. Chinese Traditional Patent Medicine, 2012; 8(3):203-206.
18. Bestmann HJ, Winkler L, von Helversen O. Headspace analysis of volatile flower scent constituents of bat-pollinated plants. Phytochemistry, 1997; 46(7):1169-1172.
19. British Pharmacopoeia Specification, H.M. Stationary Office. 1980, II.
20. National Institute of Science and Technology. Chemistry Web Book. Data from NIST Standard Reference Database, 2011, 69. (http://www.nist.gov/).
21. Kagoura M, Matsui C, Morohashi M. Phytol is a novel tumor promoter on ICR mouse skin. Japan Journal of Cancer Research. 1999; 90(4):377-384.
22. Silva RO, Sousa FB, Damasceno SR, Carvalho NS, Silva VG, Oliveira FR *et al*. Phytol, a diterpene alcohol, inhibits the inflammatory response by reducing cytokine production and oxidative stress. Fundamental of Clinical Pharmacology. 2014; 28(12):455-464.
23. Eldeen IMS, Hamid A, Wong KC, Abdullah MA, Tengku-Muhammad TS, Abdillahi HS *et al*. In vitro Repression of Cyclooxygenase, Acetyl cholinesterase Activities and Bacterial Growth by Trans-phytol and a Glycolipid from the Leaves of Homalomena sagittifolia. Research Journal of Medicinal Plants, 2016; 10(5):320-329.
24. de Moraes J, de Oliveira RN, Costa JP, Junior ALG, de Sousa DP, Freitas RM *et al*. Phytol, a diterpene alcohol from chlorophyll, as a drug against neglected tropical disease *Schistosomiasis Mansoni*. PLoS Neglected Tropical Disease, 2014; 8(1):e2617. doi: 10.1371/journal.pntd.0002617.
25. Ghaneian MT, Ehrampoush MH, Jebali A, Hekmatimoghaddam S, Mahmoudi M. Antimicrobial activity, toxicity and stability of phytol as a novel surface disinfectant. Environmental Health Engineering and Management Journal. 2015; 2(1):13-16.
26. de Menezes Santos CCP, Salvadori MS, Mota VG, Costa LM, de Almeida AAC, de Oliveira GAL *et al*. Antinociceptive and antioxidant activities of phytol *in vivo* and *in vitro* models. Neuroscience Journal. 2013, 1-9. doi.org/10.1155/2013/949452
27. Pejin B, Kojic V, Bogdanovic G. An insight into the cytotoxic activity of phytol at in vitro conditions. Natural Product Research, 2014; 28(22):2053-2056.