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Pelagia Research Library

Der Chemica Sinica, 2011, 2 (2): 255-260

Pelagia Research Library
ISSN: 0976-8505
CODEN (USA) CSHIA5

Chemical compositions of leaf and stem essential oils of *Calotropis procera* Ait R.Br [Asclepiadaceae]

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ABSTRACT

Volatile oils from leaf and stem of Calotropis procera Ait, an Asclepiadeae were analyzed for their constituents by means of gas chromatography and gas chromatography coupled with mass spectrometry. Nine compounds were identified in leaf, and ten in stem, which are respectively responsible for 93.9% and 86.4% of leaf and stem oils. Leaf oil is dominated by tyranton (54.4%), 1- pentadecene (9.5%) and 1-heptadecene (8.2%). Most abundant compounds in stem oil are Z-13-docosenamide (31.8%), isobutyl nonane (13.7%) and 2,7,10-trimethyldodecane (12.3%). Both leaf and stem volatile oils contain octadecenamide and its saturated form in appreciable amounts. Also characteristic of these oils are the presence of long chain fatty acids and amides, sulfurate, halogen compounds and carbonyls like ketones. Chemical composition of Calotropis procera essential oil is reported for the first time in literature.

Keywords: *Calotropis procera*, Asclepiadeae, essential oil, hydro distillation, GC and GC-MS.

INTRODUCTION

Calotropis procera Ait, an Asclepiadeae is a drought-resistant, salt-tolerant weed found along degraded roadsides, lagoon edges and overgrazed pastures. It is native to tropical Africa including Nigeria, Asia and Latin America where the plant is of high socio-economic value. It is commonly referred to as swallow wart, milk weed, sodom apple and rooster tree. Yoruba (in Nigeria) call it 'bomubomu'[1,2]. *Calotropis procera* have antioxidant, antimicrobial and cytostatic properties [3]. The leaf, stem and root are utilized in traditional medicine for treatment of wounds, sores and skin diseases, diarrhea, sinus fistula and jaundice. It relieves stomach pain;

its sap is used for treating eye infections. Bark of plant is traditionally used for treating coughs, elephantiasis, leprosy and ulcers [4-6]. Stem is utilized in native roofing of huts and also serve as source of charcoal [7]. Occasionally goats and sheep eat the leaves, but cattle and other livestock do not because they are slightly toxic [8]. Ajagbonna *et al* 1994 [9] reported on hematological and biochemical changes in rats fed with extracts of *C.procera*. Oladimeji *et al* [10] proposed that plant contain potentially useful ethno medicinal compounds. Their in-vitro tests indicate *C.procera* as panacea for infectious diseases and also reveal a novel potential in the fight against tumors in man. Phytochemical investigation of *C.procera* root yielded two new compounds identified as urs-18α-H-12,20(30)diene-3β-yl acetate (procerursenyl acetate) and n-triacontan-10β-ol (proceranol) [11], along with earlier reported triterpenes, triterpenoids, phytosterols, saponins, alkaloids and cardinolides [12,13,14]. This study reports for the first time chemical composition of the volatile oil of leaf and stem of *Calotropis procera*. It was collected from *C. procera* growing in mini campus, Faculty of Science, Olabisi Onabanjo University, Ago-Iwoye.

MATERIALS AND METHODS

Plant material

Leaf and stem samples of *Calotropis procera* growing in Faculty of Science, mini campus of Olabisi Onabanjo University, Ago-Iwoye, Ogun-State, Nigeria were collected in April, 2009. The plant was authenticated by Soladoye M.O. & Oyesiku O.O. (Plant taxonomists) as well as staff of the herbarium, Department of Botany and Microbiology, University of Ibadan, Ibadan.

Isolation of essential oils

The plant was separated into leaf and stem parts, and air dried. Each part was crushed and hydro distilled for 2.5hours in an all glass Clevenger-type apparatus designed to British Pharmacoepia specifications, with very small quantity of distilled *n*-hexane (0.5 ml), which was removed afterwards. The leaf and stem essential oils were procured in 0.133% and 0.09% yields respectively. Each of the oils had distinct characteristic pleasant smell.

Gas Chromatography

Each of the two essential oils was subjected to GC analyses on GC-2010[AOC-20i] gas chromatograph. Column oven temperature is 60^0C, injection temperature of 250^0C, split injection mode, at 100.2kPa; column flow of 1.61ml/min and total flow of 6.2ml/min; 1.0 split ratio; oven temperature programming is 60^0C (for 5mins), and at the rate of 5^0/min till 140^0C, 15^0/min till 280^0C.

Gas Chromatography-Mass Spectrometry

The GC-MS analyses were performed on GC-MS QP2010 Plus. Ion source temperature 200^0C; interface temperature 250^0C; solvent cut time 2.5min; with relative detector gain mode and threshold 3000; scan MS ACQ mode; detector FTD; mass range of m/z 40-400.

Identification of components

Identification of the essential oil components were based on their retention indices (determined with a reference to a homologous series of n-alkanes), along with comparison of their mass spectral fragmentation patterns in computer matching against in-built data and commercials such

as Joulain and Koenig (1998), Adams (1995), and Massada (1976) [15-17] Libraries, as well as in-house "Başer Library of Essential Oil Constituents" built up by genuine compounds and components of known oils.

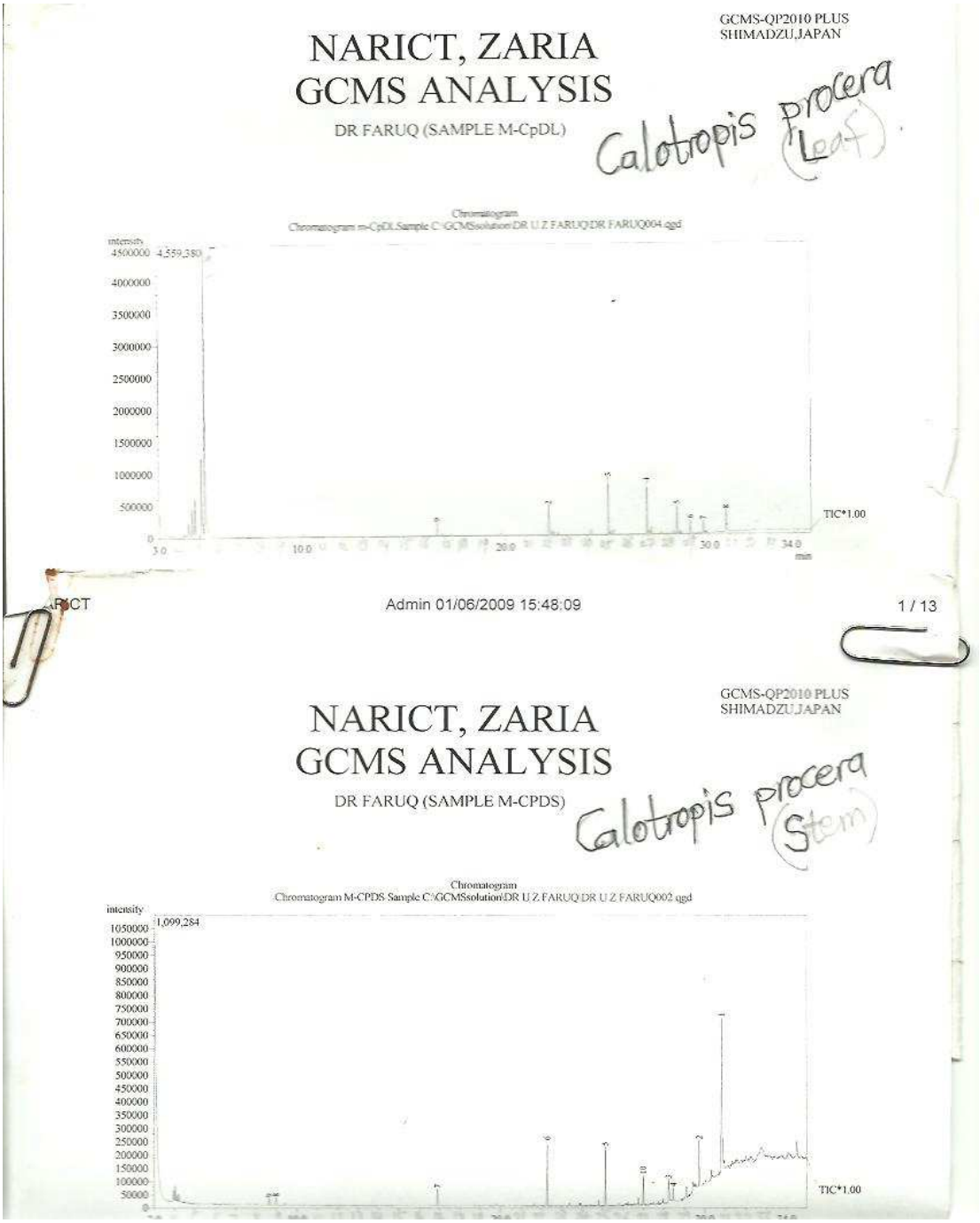

Fig. 1: Gas chromatograms of leaf and stem essential oils of *Calotropis procera* on GC-2010[AOC-20i] gas chromatograph

Oven temperature [60^{0}C]; injection temperature [250^{0}C]; column flow [1.61ml/min] and total flow [6.2ml/min]; 1.0 split ratio; oven temperature programming [60^{0}C (for 5mins) at rate of 5^0/min till 140^{0}C, 15^0/min till 280^{0}C].

RESULTS AND DISCUSSION

Leaf and stem essential oils of *Calotropis procera* Ait, an Asclepiadeae were procured in 0.133% and 0.09% yields (table 1).

Table 1: Yields of essential oils procured from leaf and stem parts of *Calotropis procera*

Plant	Parts	Weight of sample (g)	Weight of volatile oil procured (g)	% Yield of essential oil procured	Physical examination
Calotropis procera	Leaf	780	1.04	0.133%	With characteristic pleasant smell
	Stem	1100	0.99	0.09%	With characteristic pleasant smell

They were analyzed for their constituents by means of gas chromatography (GC) and gas chromatography coupled with mass spectrometry (GC-MS) (fig.1).

Nine compounds were identified in leaf, and ten in stem, which are respectively responsible for 93.9% and 86.4% of leaf and stem oils [tables 2 and 3].

Table 2: Composition of *Calotropis procera* Leaf Essential Oil

Peak No[a]	MS [Base peak+most abundant peaks] [b]	Identified compound [c]	%TIC [d]	Retention time [mins] [e]	Calculated RI[f]
1	43,59,101,58,41,83,56, 98,	$C_6H_{12}O_2$ Tyranton /(4-hydroxy,methyl,2pentanone)[116]	54.366	5.3	477.44
2	55,43,41,69,57,83,56,7 0,97,82,71,	$C_{13}H_{26}$ 1-tridecene [182]	5.766	22.1	1972.09
3	55,83,57,43,69,97,41,5 6,70,71,84,	$C_{15}H_{30}$ 1-pentadecene[210]	9.473	25.1	2373.58
4	55,57,83,97,69,56,70,	$C_{17}H_{34}$ 1-heptadecene [238]	8.237	26.9	2417.17
5	97,57,83,55,69,43,41,7 1,56,70,111,	$C_{19}H_{38}$ 1-nonadecene[266]	4.942	28.4	2767.31
6	83,55,97,57,69,43,41,5 6,70,82,71,111,	$C_{17}H_{36}O$ 1-heptadecanol [256]	2.471	29.1	2784.86
7	97,83,57,55,69,71,43,7 0,41,56,111,	$C_{20}H_{40}$ 3-eicosene [280]	2.059	29.7	2799.89
8	59,72,55,41,43,60,98,	$C_{18}H_{35}NO$ 9-octadecenamide [281]	4.119	30.8	2827.46
9	55,43,41,56,69,70,57,8 3,97,84,	$C_{11}H_{22}$ 1-undecene [154]	2.471	16.6	1514.19

[a]According to %TIC from relative percentage abundances of total ion concentration [TIC] from GC [fig.1]. Retention time from GC is also given for each; [b][m/e] values of fragment ions with base peak 1st stated, and other most prominent ions; [c]General formula, name and molecular weight of authenticated and identified compound are stated; where common name exist, this is also stated. Also see identification of components; [d]Total ion concentration in % from GC; [e]Retention time in minutes from GC; [f]Retention Index with reference to homologous series of n-alkanes.

Table 3: Composition of *Calotropis procera* Stem Essential Oil

Peak No[a]	MS [Base peak+most abundant peaks] [b]	Identified compound [c]	%TIC [d]	Retention time [mins] [e]	Calculated RI [f]
1	59,72,55,41,43,	$C_{22}H_{43}NO$ Z-13-docosenamide [337]	31.847	30.8	2827.46
2	59,72,43,57,71,41,85,55, 99,	$C_{18}H_{37}NO$ octadecanamide [283]	9.099	29.7	2799.89
3	73,60,43,57,41,55,97,87, 71,	$C_{11}H_{22}O_2$ undecanoic acid [186]	4.095	28.2	2762.30
4	71,57,85,43,83,99,41,	$C_{14}H_{30}O_3S$ 2-ethylhexyl-isohexylSulfurate [278]	2.730	28.5	2769.82
5	57,71,43,85,41,55,99,56, 70,69,42,113,	$C_{15}H_{32}$ 2,7,10/ 2,6,11-trimethyldodecane [212]	12.284	25.1	2373.58
6	57,43,71,85,41,55,56,99,	$C_{13}H_{28}$ 5-isobutylnonane [184]	13.649	22.3	1976.74
7	57,43,71,41,85,56,55,40,	$C_{12}H_{26}$ 3-methylundecane [170]	3.185	16.9	1521.16
8	43,45,41,57,85,44,	$C_7H_{16}O_2$ 1-methylhexylhydroperoxide [132]	1.820	9.0	761.67
9	43,41,57,55,85,44,	$C_7H_{12}O_2$ 2,3-heptanedione [128]	1.820	8.7	753.18
10	57,71,43,85,41,55,99,56, 69,70,83,	$C_{10}H_{21}I$ 1-iododecane [268]	5.915	27.0	2419.59

[a]According to %TIC from relative percentage abundances of total ion concentration [TIC] from GC [fig.1]. Retention time from GC is also given for each; [b][m/e] values of fragment ions with base peak 1st stated, and other most prominent ions; [c]General formula, name and molecular weight of authenticated and identified compound are stated; where common name exist, this is also stated. Also see identification of components; [d]Total ion concentration in % from GC; [e]Retention time in minutes from GC; [f]Retention Index with reference to homologous series of n-alkanes.

Leaf oil is dominated by tyranton (54.4%) i.e. (4-hydroxy,4methyl,2pentanone), 1- pentadecene (9.5%) and 1-heptadecene (8.2%) [table2]. Most abundant compounds in stem oil are Z-13-docosenamide (31.8%), isobutyl nonane (13.7%) and 2,7,10-trimethyldodecane (12.3%) [table3]. Both leaf and stem essential oils contain octadecenamide and its saturated form in appreciable amount. Also characteristic of these oils are the presence of long chain fatty acids and amides, sulfurate, halogen compounds and carbonyls like ketones. Chemical composition of *Calotropis procera* essential oil which has not been reported earlier in literature is presented in tables2 and 3.

CONCLUSION

Chemical composition of the volatile oil of traditionally useful *Calotropis procera* which is reported for the first time in literature consist of nine identified compounds in leaf, and ten in stem, which are respectively responsible for 93.9% and 86.4% of leaf and stem oils. Leaf oil is dominated by tyranton (54.4%),1- pentadecene (9.5%) and 1-heptadecene (8.2%). Most abundant compounds in stem oil are Z-13-docosenamide (31.8%), isobutyl nonane (13.7%) and 2,7,10-trimethyldodecane (12.3%). These and other compounds in *Calotropis procera* are presented in tables 2 and 3.

Acknowledgement
We acknowledge the assistances of Chemical Science students, supervised by D.O.Moronkola, who assisted in the isolation of the essential oils. Thanks to Dr Faruq who linked us to NARICT, Zaria making possible the GC and GC-MS analyses which we fully paid for.

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