Full Length Research Paper

Chemical constituents of the essential oil from Syzygium cordatum (Myrtaceae)

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Essential oil was extracted from dried leaves of *Syzygium cordatum* by hydrodistillation. The components were identified by gas chromatography (GC/FID) and gas chromatography coupled with mass spectrometry (GC/MS). Sixty compounds representing 79% of the *S. cordatum* oil were identified. The main constituents were 6,10,14-trimethylpentadecane-2-one (14.4%), 2,3-butanediol diacetate (13.3%), n-hexadeconic acid (7.2%), ethane, 2-chloro-1,-bis(2-chloroethoxy) (6.2%), Isopentyloxyethyl acetate (5.03%), methane and bis (2-chloroethoxy) (3.8%).

Key words: *Syzygium cordatum*, essential oil composition, 6,10,14-trimethylpentadecane-2-one, 2, 3-butanediol diacetate.

INTRODUCTION

Mankind has relied from ancient times on the usage of plant derived products for food and also for treating diseases. As much as 80% of the human population, especially from third world countries harvest plants for health care (Werka et al., 2007). From early Greece, Egypt and Rome, essential oils have been used in perfumery, as food flavorants and pharmaceuticals (Baris et al., 2006). The antimicrobial activities of essential oils are well known and with increasing interest of the applications in the food and pharmaceutical industries, there is increased interest in screening of plant extracts for active compounds (Amvam Zollo et al., 1998). Research into plant essential oils have also gained momentum due to their fumigant and contact insecticidal activities and the less stringent regulatory approval mechanisms for their exploration due to long history of use (Isman, 2006). Of late, the essential oils are being tried as potential candidates for weed control (Singh et al., 2003; Batish et al., 2004, 2007), and pest and disease management (Isman,

2000; Pawar and Thaker, 2006; Abad et al., 2007). Furthermore, essential oils are easily extractable, ecofriendly in that they are biodegradable and get easily catabolized in the environment (Zygadlo and Grosso, 1995), do not persist in soil and water (Misra and Pavlostathis, 1997; Isman, 2000, 2006), possess low or no toxicity against vertebrates, fishes, birds and mammals (Enan et al., 1998, Isman, 2000; Isman and Machial, 2006; Bakkali et al., 2008), and these enables the oils to have applications even in sensitive areas such as schools, restaurants, hospitals and homes.

Looking at the importance of ecosystem services to mankind, it is worthwhile to explore environmental benefits of the natural products to mankind. Plant essential oils are obtained from non-woody parts of the plant, particularly foliage, through steam or hydrodistillation. They are complex mixtures of mainly terpenoids and oxygenated compounds that determine the characteristic aroma and odour of the donor plant. Presence of volatile monoterpenes or essential oils in the plants, particularly against herbivorous insect pests and pathogenic fungi (Langenheim, 1994). These volatile terpenoids also play a vital role in plant-plant interactions and serve as attract-

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tants for pollinators (Tholl, 2006). They act as signaling molecules and depict evolutionary relationship with their functional roles (Theis and Lerdau, 2003).

Among various aromatic plants, plants from the Myrtaceae family are represented by over 700 species that are distributed throughout the world (Brooker and Kleinig, 2006) Eucalyptus belongs to this family and is an example of one of the most-extensively planted pulpwood species (Zobel, 1988). It consists of tall, magnificent and evergreen trees with fragrant foliage rich in oil glands and is an excellent source of commercially important eucalyptus oil that finds extensive use in pharmaceutical, perfumery and industry (Brooker and Kleinig, 2006). The essential oil from eucalyptus species are among the world's top traded oils and also, the oil extracted from *Eucalyptus citriodora* is one of the world's major oil in terms of trade volume (Green, 2002).

Syzygium cordatum Hochst. ex C.Krauss commonly known as waterberry also belong to the family Myrtaceae. It is widely distributed and occurs in the eastern and north eastern parts of South Africa, from KwaZulu-Natal northwards to Mozambique (Van Wyk et al., 1997). The tree is commonly found near streams, forest margins or in swampy spots. It s a medium-sized tree of up to 15 m in height, with a rough dark brown bark. The leaves are broad, circular with a bluish-green colour. In traditional medicinal practices, it is used by the Zulu people for respiratory ailments, including tuberculosis, and for stomach complaints and as emetics (Cunningham, 1993, Watt and Brever-Brandwijk, 1962; Pooley, 1993) and in parts of central Africa, it is used for stomach ache, wounds and diarrhoea. Scientific studies show that the leaf extracts of S. cordatum contain compounds that could be effective in treating mild diabetes mellitus or glucose tolerance impairment (Musabayane et al., 2005) and the methanolic and water extracts of S. cordatum have been found to have antifungal activity against Candida albicans (Steenkamp et al., 2007). From the literature little is published on the chemical constituents of S. cordatum wood and bark by Candy et al., (1968) and composition of essential oil by Chisowa et al., (1998). This paper describes the composition of essential oils of leaves of S. cordatum.

MATERIALS AND METHODS

Plant material

Material from plants of *S. cordatum* was collected in October 2009 in the KwaZulu-Natal province of South Africa. The species was identified and a voucher specimen was deposited in the Ward Herbarium at University of KwaZulu-Natal, Westville Campus, Durban, South Africa. KwaZulu-Natal (Durban) lies in an altitude of ~40 m on latitude (29° 48'S) and longitude (30° 56'E).

Extraction of the essential oil

The leaves of S. cordatum were dried in a convection oven at 25 °C

and milled (waring blender). The essential oil was extracted by slight modification of the procedure outlined by Denny (1989). 100 g of milled leaves were triturated in Metsabo hammer mill and hydrodistilled in a Clevenger apparatus. After 6 h of distillation, the essential oil was removed from the water surface. The oil was dried over anhydrous sodium sulphate. After filtration, the solvent was removed by distillation under reduced pressure in a rotary evaporator at 42°C and the pure oil samples were sealed and kept in a dark bottle at 4°C in the refrigerator for further analysis. The resulting pale yellow oil (40 µl) was solubilised in 1 ml of methyl ethyl ketone before the injection. 1 µl of this solution was directly used for analysis.

Gas chromatography analysis (GC/FID)

Capillary gas chromatography was performed using a Agilent system consisting of a model 6820 gas chromatograph (Agilent, USA) and using a fused silica capillary column DB-5, 30 m x 0.35 mm, 0.1 μ m film thickness (J & W Scientific, USA). The temperature program was set from 80 °C (1 min) to 280 °C (20 min) at 15 °C/min. The injection temperature was 250 °C and the injection volume was 1.0 μ l. The inlet pressure was 100 kPa. Nitrogen was used as a carrier gas. Sampling rate was 2 Hz (0.01 min) and flow ionization detector temperature was set at 280 °C.

Gas chromatography analysis (GC/MS)

The GC/MS analysis of the essential oil was performed on an Agilent GC 6890 model gas chromatograph-5973N model mass spectrometer equipped with a 7683 series auto-injector (Agilent, USA). A DB-5MS column (30 m x 0.25 mm x 0.25 µm film thickness) was used. Temperature program was set from 80 °C (1 min) to 280 °C (20 min). Injection volume was 1.0 µl and inlet pressure was 38.5 kPa. Helium was used as carrier gas. Linear velocity (*u*) was 31 cm/sec. Injection mode was split (75:5). MS interface temperature was 230 °C, MS mode was EI, detector voltage was 1.66 Kv, mass range was 10 to 700 u, scan speed was 2.86 scans/s and interval was 0.01 min (20 Hz).

Identification of components

Compound identification was based on the comparison of retention indices (determined relatively to the retention times of n n-alkanes series) using a MS library. The NIST spectrometer data bank was used to determine the percentage composition of the compounds (Adams, 2007).

RESULTS

The essential oil from 100 g of dried leaf powder gave a yield of 1.15% essential oil. The oil was a yellowish liquid with a strong aromatic fragrance. The chemical compound and their percentages are shown in Table 1. Sixty compounds were identified by comparison of their retention indices and the mass spectra of each GC component with those of standards and with reported data. Terpenes and derivatives predominated with the most abundant compound (14.4%) identified as 6,10,14-trimethylpentadecane-2-one (Figure 1). This was followed by 2,3-butanediol diacetate (13.3%), n-hexadeconic acid (7.2%), ethane, 2-chloro-1,-bis (2-chloroethoxy) (6.2%), isopen-

Peak number	Compound	RI*	Area peak (%)
1	Ethene, chloro-	297	2.90
2	Toluene	311	0.84
3	Oxirane, 2,3-dimethyl-	347	0.31
4	1,2-Epoxy-3-propyl acetate	367	0.11
5	1,3-Dioxan-4-one	383	0.21
6	Ethylene maleic anhydride	401	1.41
7	2,3-Butanediol diacetate	414	13.13
8	1,1-Ethanediol, diacetate	425	0.19
9	Isopentyloxyethyl acetate	430	5.03
10	Glycine, N-acetyl-	433	0.12
11	Hydrazine, 2-propenyl	437	0.12
12	4-Methylthiazole	446	0.26
13	2,5-Hexanedione	455	0.30
14	3-Heptanol	464	0.70
15	Silane	470	0.19
16	2-Heptanone	484	0.09
17	3-Hexanol	494	0.17
18	2-Butanone, 4-(acetyloxy)-	504	0.14
19	3-Hepten-2-one, 5-methyl	511	0.36
20	Triacetin	519	1.07
20	2,4-Pentanedione	535	0.53
22	Propane, 1,1,2-trichloro-	541	0.33
22	•	548	1.96
23 24	N,N,N',N'-Tetraacetylethylenediamine	540 552	1.96
	2,4-Dimethyl-3-pentanol acetate		
25	2,4-Pentanedione	555 550	0.22
26	3-Heptanol, 3,6-dimethyl-	559	1.10
27	6,10,14-trimethylpentadecane-2-one	574	14.41
28	5-Undecanone	604	0.34
29	3-Decanone	613	0.88
30	(Trimethylsilyl)diazomethane	645	0.14
31	Methane, bis(2-chloroethoxy)	654	3.88
32	1,3,4-Oxadiazole	687	0.32
33	Oxazole, trimethyl	730	0.84
34	Ethane, 1,1-dichloro	780	0.33
35	Ethane, 2-chloro-1,1-bis(2-chloroethoxy)	838	6.25
36	Ethanesulfonyl chloride, 2-chloro	848	0.27
37	Ledol	866	0.09
38	Ethane, 1,2-bis(2-chloroethoxy)-	881	0.08
39	Naphthalene, 1,2,3,4,4a,5,6,8a-octahydro(1.alpha.,4a.beta.,8a.alpha.)-	887	0.68
40	Eudesma-4(14),11-diene	903	0.29
41	Naphthalene, 1,2,3,4-tetrahydro-1,6-dimethyl-4-(1-methylethyl)-, (1S-cis)	921	2.07
42	2(4H)-Benzofuranone	944	0.10
43	3-Penten-2-one, 4-phenyl-	951	0.13
44	trans-ZalphaBisabolene epoxide	977	0.20
45	Isoaromadendrene epoxide	994	0.29
46	Nonanoic acid	1000	0.10
47	Diepialphacedrene epoxide	1011	0.11
48	6-Isopropenyl-4,8a-dimethyl-1,2,3,5,6,7,8,8a-octahydro-naphthalen-2-ol	1010	0.20
49	Azulene	1022	0.09

Table 1. Chemical composition of essential oil of S. cordatum.

Table	1.	Continue
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50	Cedrol	1040	0.14
51	Naphthalene, 1,6-dimethyl-4-(1-methylethyl)-	1057	1.12
52	Isoaromadendrene epoxide	1141	0.24
53	2-Furanone	1243	1.25
54	Hexadecanoic acid, methyl ester	1392	2.72
55	Isophytol	1439	0.11
56	n-Hexadecanoic acid	1481	7.25
57	9,12,15-Octadecatrienoic acid, methyl ester, (Z,Z,Z)-	1890	0.71
58	Phytol	1931	0.45
59	Octadecanoic acid, methyl ester	1999	0.20
60	1-Eicosene	2057	0.31

Total (%), 79.16; ^aRI, Retention indices relative to *n*-alkanes on DB-5 capillary column.

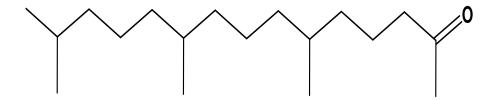


Figure 1. 6,10,14-trimethylpentadecane-2-one.

tyloxyethyl acetate (5.03%), methane, bis (2-chloroethoxy) (3.8%), ethene, chloro- (2.9%), hexadecanoic acid, methyl ester (2.7%), naphthalene, 1,2,3,4tetrahydro-1,6-dimethyl-4-(1-methylethyl)-, (1S-cis) (2%), N,N,N',N'-tetraacetylethylenediamine (1.9%), ethylene maleic anhydride (1.4%), 2-furanone (1.2%), naphthalene, 1,6-dimethyl-4-(1-methylethyl) (1.2%), 3-heptanol, 3,6-dimethyl- (1.1%),2,4-dimethyl-3-pentanol acetate (1%) and triacetin (1%).

DISCUSSION

The yield of essential oil from *S. cordatum* is relatively higher than that reported for other industrially exploited plants such as lavender (0.8 to 1.8%), mint (0.5 to 1%), neroli (0.5 to 1%) and laurel (0.1 to 0.35%) (Edward et al., 1987). In another study (Leitão et al., 2008), *Lippia rotundifolia* illustrated a low percentage (0.01%) of essential oil.

The major compound in the oil was 6,10,14-trimethylpentadecane-2-one, a C15 aliphatic methyl ketone. Similar long chain aliphatic methyl ketones have been reported and shown to be repellent to arthropods including blood sucking insects (Blum et al., 1996; Ndungu et al., 1995; Torr et al., 1996; Gikonyo et al., 2002; Roe, 2004). In a study by Innocent et al. (2008) on the efficacy of such compounds against *Anopheles gambiae*, a malaria vector, C11-C15 compounds were more effective than C7-C10 compounds and among the C11-C15 compounds, odd-carbon compounds were more effective than even-carbon compounds. In these studies, the C15 compound was found to be as effective as N, N-diethylm-toluamide (DEET) which is used to repel mosquitoes.

With further research, the essential oil from *S. cordatum* could prove to be an alternative to organisms that are resistant to DEET because it has 6,10,14-trimethylpentadecane-2-one. It could also find other applications that are linked to the diversity of the other chemicals, that is, 2,3-butanedioldiacetate (13.1%), n-hexadecanoic acid (7.2%), ethane, 2-chloro-1, 1-bis (2-chloroethoxy) (6.2%), isopentyloxyethyl acetate (5%), methane, bis (2-chloroethoxy (3.8%), ethane, chloro-(2.9%), hexadecanoic acid (2.7%) and naphthalene (2.0%) which comprised more than 79% of the essential oil.

The GC-FID and GC-MS study of the essential oil from *S. cordatum* from KwaZulu-Natal province of South Africa led to the report of the analysis and identification of 60 compounds, representing 79.16% of the total mass. The major compound, 6,10,14-trimethylpentadecane-2-one (14.4%) was relatively higher than that in *Minuartia meyeri* (5.1%) from the family Caryophyllaceae (Nurettin et al., 2006) and close to that in *Calotropis procera* (15.3%) from the family Apocynaceae (Okiei et al., 2009). As a continuation of this research, it would be necessary

to evaluate the use of the essential oil as a repellant for insects including its role in combating malaria.

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