

CHEMICAL COMPOSITION AND ANTIMICROBIAL ACTIVITY OF THE ESSENTIAL OIL OF *KLEINIA ODORA*

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تم تحليل الزيت الطيار لنبات كلينيا أودورا فورسك دي سي، باستعمال تقنية كروماتوجرافيا الغاز المتصلة بطياف الكتلة، وقد تم التعرف على 32 مركباً وقد تبين من الدراسة أن (+) إبي بايسيكلوسيسكوفلاندرين وكاريوفيلين وألفابينين تمثل المركبات الأساسية في هذا الزيت. وقد وجد أن العينة لها تأثير مثبط معتدل مضاد لكل من الميكروبات والفطريات.

The essential oil of the herb *Kleinia odora* (Forssk) DC was analyzed by gas-chromatography-mass spectroscopy (GC/MS) technique. Thirty two components were identified. Among these (+)-epi-bicyclosesquiphellandrene, caryophyllene, α -caryophyllene and α -pinene were the major constituents. The oil showed a moderate antimicrobial and antifungal activities.

Key words: *Kleinia odora*; Asteraceae; essential oil composition; (+)-epi-bicyclosesquiphellandrene, caryophyllene, α -caryophyllene, α -pinene; antimicrobial activity.

Introduction

Genus *Kleinia* (Family Asteraceae) is represented in Saudi Arabia by three species namely *K. odora* (Forssk)DC, *K. deflersii* (O.Schwartz) and *K. pendula* (Forssk) Sch. Bip. These are commonly distributed in the Southern regions of the Kingdom (1-3).

Literature reviews showed that this genus is a rich source of oxygenated sesquiterpenes and triterpenoids (4, 5). Abrotanifolone derivatives were reported in *K. tomentosa* (6) and two new acylpyrrole derivatives were identified in *K. kleinioides* (7). Also, three triterpenoids namely lupeol, lupenone and lupeol acetate were isolated from *K. odora* (8).

As a continuation to our previous chemical investigation of the bioactive constituents of *K. odora* grown in the Kingdom of Saudi Arabia (4, 9), we have decided to examine the composition of the essential oil distilled from this species.

Literature survey revealed that only one report on the composition of an essential oil obtained from *K. pendula* grown in Somalia, with five major mono- and sesquiterpenoids (myrcene, α -humulene, β -elemene, T-cadinol and 4 α H –eudesm-5 α -ol) were identified in that oil (4).

As no report has been found concerning *K. odora* volatile oil constituents, therefore, analysis of this oil was performed to identify its constituents and to compare them with those of *K. pendula*. In addition the antimicrobial activity of the oil was determined.

Experimental

Plant Material:

Kleinia odora (Forssk)DC (Asteraceae) has been collected in the Southern region of the Kingdom of Saudi Arabia (from Fayfa Mountain (Jabal Fayfa) about 100 km North East of Gizan (in June 1997) and from Asir, between Al-Khamees and Abha (very close to Abha Airport) in September 1998). The collected plant materials were kindly identified by Dr. Sultan Abedin, plant taxonomist, College of Pharmacy, King Saud University, Riyadh Saudi Arabia.

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Preparation and Analysis:

The oil was prepared by hydrodistillation (10) of the fresh herb. The GC/MS analysis was carried out on Gas Chromatography Mass Spectrometer GC-17A Shimadzu, Auto Injector- AOC -201, equipped with Shimadzu Electron Multiplier Detector, Shimadzu, Japan. Capillary column of fused silica (30 m x 0.25 mm i.d.) coated with 5% phenyl methyl polysiloxane (DB-5). Helium ultra pure was used as carrier gas at flow rate 25 ml/ min. Oven temperature was programmed at 60-200 °C at 10° C/min. The total run time for GC was 60 minutes, the injection mode split was 50, the MS detection routine was from 9-60 min. The scan of m/z was from 80-350 and the Start-Stop masses were from 40-600. Identification of the components was performed by aid of the computer library search (CAS No. 5989-27-5, Entry 8747, LIB #1), comparison of mass spectra with literature data and by comparison of their retention indices with those reported (11).

Antimicrobial Screening:

The antimicrobial screening of the oil was performed according to the general qualitative assay described by Clark *et al* (12) using the following microorganisms: *Escherichia coli*, *Staph. aureus* and *Candida albicans*, all of them are pure isolated strains obtained from stock cultures of the Department of Microbiology, College of Pharmacy, KSU. Tetracyclin was used as a positive control for bacteria and Amphotericin for fungi.

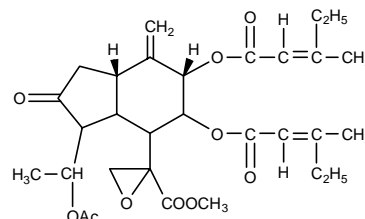
Results and Discussions

The volatile oil of *K. odora* was prepared by hydrodistillation (11) of the fresh herb. The percentage of the essential oil was 0.17 % w/v. It is a pale yellow liquid with a characteristic aromatic odour. It showed RI 1.50223 and specific gravity 0.744. The oil is soluble in ethanol and ether, insoluble in water. GC/MS analysis resulted in the identification of thirty two components (Table 1). Among these (+)-Epi- bicyclosesquiphellandrene (29.58 %), caryophyllene (14.25 %) α -caryophyllene (13.96) and α -pinene (10.15 %) were the main components.

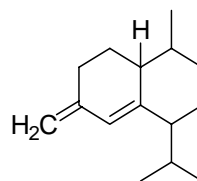
It is worthy to note that apart of cadinol none of the major constituents reported in *K. pendula* oil (4) occurs even in trace amounts in *K. odora* oil. β -cadinol is one of the major constituents of the first

oil, while α -cadinol occurs as minor constituent (0.18%) in *K. odora* oil.

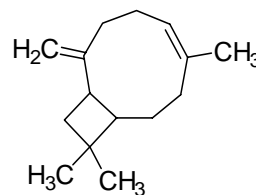
The oil showed a moderate antimicrobial activity against *Escherichia coli* and antifungal activity against *Candida albicans* (Table 2).



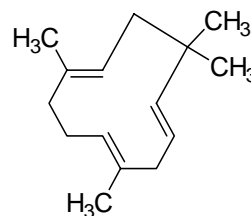
Abrotanifolone derivative
(isolated from *K. tomentosa* Bak)



(+)-Epi-bicyclosesquiphellandrene



Caryophyllene



Alpha-caryophellene

Major constituents of the oil

Table 1: Identified constituents of the essential oil of *K. odora*

Peak No	Compound name	RT	M+	Base peak	Main fragments*	Relative%
1	1-Nonanol	7.750	144	56	43,70,41,83,44	0.67
2	α -Pinene	8.800	136	93	77,121,105,136,39	10.15
3	4-Methylethyl-1-(1-methylethyl)bicyclo[3.1.0]hexane	9.492	136	93	77,41,92,136,27	0.82
4	β -Pinene	9.650	136	93	41,69,39,67,121,136	0.74
5	(+)-4-Carene	10.273	136	121	93,136,91,77	2.30
6	1-Methyl-4-(1-methylethyl)-1,3-cyclohexadiene	10.305	136	121	93,136,77,105	2.30
7	1-Methyl-4-(methylethyl)-benzene	10.367	134	119	134,91,77,65	1.23
8	3-Cyclohexen-1-ol	13.275	154	71	111,93,43,55,154	0.45
9	(-)- α -Terpineol	13.467	154	43	59,93,43,121,81,154	0.11
10	1-Phenyl-2-butanone	13.833	148	57	91,71,29,148	0.53
11	Copaene	16.617	204	161	119,105,43,93,81,204	2.43
12	(+)-Epi-bicyclosesquiphellandrene	16.767	204	161	105,119,91,81,204	29.58
13	Aristolene	16.933	204	162	105,91,119,133,147	0.71
14	Caryophyllene	17.383	204	41	93,69,133,55,107,120	14.25
15	alpha-Caryophyllene	17.867	204	93	80,121,41,147,204	13.96
16	1,2,3,4,4a,5,6,8a-Octahydro-7-methyl-4-methylene-1-(1-naphthalene	18.0	204	161	204,43,105,119,134,204	0.70
17	Germaecrene D	18.183	204	161	105,91,119,41,77,133	1.97
18	1,3-Cyclopropa(1,2)benzene, octahydro-7-methyl-3-meth-1H-cyclopental	18.2	204	161	105,91,120,41,55,147	1.97
19	1H-Decahydro-1,1,7-trimethyl-4-methylene-, (1a cycloprop(e)azulene	18.242	204	41	93,67,161,105,204	1.90
20	Decahydro-4a-methyl-1-methylene-7-(1-methyl ethenyl)-naphthalene	18.308	204	105	41,161,204,93,81,67	2.06
21	1-Hydroxy-1,7-dimethyl-4-iso-propyl-2,7-cyclodeca-diene	18.408	222	43	201,161,105,81,95,222	2.34
22	1,2,3,4,4a,5,6,8a-Hexahydro-4,7-dimethyl-1-(1-methylethyl)-naphthalene	18.592	204	161	204,134,41,119,105,204	2.35
23	τ -Muurolol	18.692	222	43	207,161,205,105,81	3.14
24	3,7-Cyclodecadiene,1- methanol, alpha, alpha,4,8-tetramethyl	18.992	222	59	93,161,81,41,107,222	0.27
25	Caryophyllene oxide	19.200	220	43	79,93,95,109,55,220	0.17
26	2,5,9-Trimethylcoundeca-4,8-dienone	20.042	206	96	138,123,81,67,41,206	0.44
27	Cubenol	20.175	222	119	161,43,93,59,105,204	0.46
28	α -Bisabolol	20.392	222	109	43,69,109,93,95,204	0.81
29	Humulane-1,6-dien-3-ol	20.417	222	109	43,204,95,81,161,189	0.81
30	α -Cadinol	20.467	222	43	95,121,204,161,79, 105	0.18
31	Decahydro-alpha,4a-trimethyl-8-met-2-naphthalenemethanol	20.533	222	59	149,164,108,41,81,67,222	1.22
32	1,2,3,3a,4,5,6,7-Octahydro- α , 3,8-t-5-azulenemethanol	20.658	222	59	135,107,93,161,81,41,222	0.37

*Compounds were identified by aid of the computer library search , comparison of mass spectra with literature data (11) and by comparison of their retention indices with those reported.

Table 2: Antimicrobial screening of *K. odora* oil

Microorganism	Zone of Inhibition		
	<i>Kleinia</i> oil	Tetracycline	Amphotericin
<i>Staphylococcus aureus</i>	-	29	-
<i>Escherishia coli</i>	9	18	-
<i>Candida albicans</i>	9	-	20

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