Bull. Chem. Soc. Ethiop. **2015**, 29(2), 327-330.
Printed in Ethiopia
DOI: http://dx.doi.org/10.4314/bcse.v29i2.15
ISSN 1011-3924

SHORT COMMUNICATION

VOLATILE CONSTITUENTS OF *HAPLOPHYLLUM BUHSEI* BOISS. FLOWERING AERIAL PARTS

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(Received March 5, 2014; revised May 11, 2015)

ABSTRACT. The essential oil composition of flowering aerial parts of *Haplophyllum buhsei* Boiss. (Rutaceae) from Iran was analyzed for the first time using gas chromatography (GC) and gas chromatography–mass spectrometry (GC–MS). Thirty six compounds comprising 92.2% of the total oil were characterized. The main components were β-caryophyllene (12.9%), limonene (9.7%), β-pinene (7.9%), linalool (7.4%), α-pinene (6.4%) and 1,8-cineole (5.5%). Volatile oil of *Haplophyllum buhsei* Boiss. flowering aerial parts was dominated by the high content of monoterpene hydrocarbons (39.2%) and oxygenated monoterpenes (23.2%).

KEY WORDS: *Haplophyllum buhsei* Boiss., Rutaceae, Essential oil, β-Caryophyllene, Limonene

INTRODUCTION

The genus *Haplophyllum* A. Juss. belongs to Rutaceae family and encompasses approximately 70 species which mainly distributed around the Mediterranean region of Europe and through western Asia up to Siberia [1, 2]. Eighteen species of *Haplophyllum* A. Juss. genus are found in Iran which among them nine species are endemic and *Haplophyllum buhsei* Boiss. is one of them [3].

Plants of *Haplophyllum* A. Juss. genus have long been used in folk medicine for toothache, stomach and skin diseases, and in the treatment of some types of cancer. The extracts of some *Haplophyllum* species exhibit cytotoxic activity [4]. This genus has been shown to possess alkaloids, lignans, flavonoids, coumarins and essential oils with important biological properties [5].

A thorough literature review revealed that volatile constituents of many species of the genus *Haplophyllum* A. Juss. have been widely investigated in various regions from all over the world [6-17] but to the best of our knowledge, there was no report on *Haplophyllum buhsei* Boiss. and in this article essential oil composition of this endemic species is informed for the first time.

EXPERIMENTAL

Plant material. Flowering aerial parts of *Haplophyllum buhsei* Boiss. were collected in May 2013 from Dalahu mountain (Dalahu County, Kermanshah Province, Iran).

Essential Oil Isolation. The air-dried crushed flowering aerial parts of *Haplophyllum buhsei* Boiss. were subjected to hydrodistillation using a Clevenger-type apparatus for 4 hour. The obtained essential oil was dried over anhydrous sodium sulphate and stored at 4-6 ^{o}C.

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GC and GC–MS analyses. Chemical composition of the volatile oil was investigated by gas chromatography (GC) and gas chromatography–mass spectrometry (GC–MS). *Haplophyllum buhsei* Boiss. oil was analyzed by GC–MS using a Hewlett-Packard 6890 gas chromatograph with DB-5 capillary column (30 m x 0.25 mm; film thickness 0.25 μm). The carrier gas was helium with a flow rate of l mL/min. The column temperature was programmed from 60 ^{o}C to 220 ^{o}C at 6 ^{o}C/min. The gas chromatograph was coupled to a Hewlett-Packard 5973 mass selective detector. The MS was operated at 70 eV ionization energy. The retention indices were calculated by using retention times of *n*-alkanes that were injected after the essential oil at the same conditions. The components were identified by comparison of retention indices with those reported in the literatures and also by comparison of their mass spectra with the published mass spectra or Wiley library [18, 19]. Gas chromatography using flame ionization detection (GC–FID) analysis was carried out under the same experimental conditions with the same column as described for the GC–MS. The relative percentage of the identified compounds was computed from the GC peak area without applying correction factors.

RESULTS AND DISCUSSION

The dried flowering aerial parts of *Haplophyllum buhsei* Boiss. yielded 0.35% V/W of a pale yellow volatile oil. Thirty six compounds comprising 92.2% of the total oil were detected which are presented in Table 1.

Essential oil of *Haplophyllum buhsei* Boiss. flowering aerial parts was characterized by the high amount of monoterpene hydrocarbons (39.2%) with limonene (9.7%), β-pinene (7.9%), *α*-pinene (6.4%), sabinene (4.6%) and myrcene (3.8%) as main components. Oxygenated monoterpenes represented 23.2% of the volatile oil which dominated by linalool (7.4%) and 1,8-cineole (5.5%). The sesquiterpene hydrocarbons fraction comprised 22.8% of the oil with β-caryophyllene (12.9%) as the abundant constituent. Oxygenated sesquiterpenes came up to 6.5% and caryophyllene oxide (3.5%) constituted the major ingredient of this fraction.

Essential oil compositions of numerous species of *Haplophyllum* A. Juss. genus were previously reported which depend on many factors such as genetic, geographic distribution, phenological cycle, seasonal variation, plant organ and analytical method used. Volatile compounds that have been most commonly reported from *Haplophyllum* species contain monoterpenoids (e.g. linalool, *α*-pinene, β-pinene, limonene, *α*-phellandrene, β-phellandrene, *α*-terpineol, sabinene, 1,8-cineole and myrcene) and sesquiterpenoids (e.g. β-caryophyllene, *α*-humulene, elemol, caryophyllene oxide and β-eudesmol) as the principle constituents in the most of them [6-17].
Regarding to our literature survey, there was no research on essential oil composition of *Haplophyllum buhsei* Boiss. and this study describes volatile oil isolation and identification of this endemic species for the first time.

Table 1. The essential oil composition of *Haplophyllum buhsei* Boiss. aerial parts.

No.	Compound	RI[a]	Content (%)
1	*α*-Pinene	937	6.4
2	*α*-Fenchene	951	0.9
3	Benzaldehyde	963	Trace[b]
4	Sabinene	974	4.6
5	*β*-Pinene	979	7.9

6	Myrcene	990	3.8
7	*n*-Decane	1000	0.3
8	*α*-Phellandrene	1005	2.9
9	*p*-Cymene	1025	0.4
10	Limonene	1028	9.7
11	1,8-Cineole	1030	5.5
12	(*Z*)-*β*-Ocimene	1034	1.8
13	*γ*-Terpinene	1061	0.8
14	Linalool	1100	7.4
15	*α*-Thujone	1105	1.5
16	Camphor	1145	2.8
17	Borneol	1165	0.3
18	*α*-Terpineol	1189	2.5
19	Carvone	1242	0.3
20	Linalyl acetate	1255	2.9
21	*α*-Ylangene	1372	0.9
22	*α*-Copaene	1376	0.8
23	*β*-Elemene	1391	2.7
24	*β*-Caryophyllene	1419	12.9
25	*α*-Humulene	1453	2.6
26	*γ*-Muurolene	1476	Trace[b]
27	Bicyclogermacrene	1496	0.2
28	*γ*-Cadinene	1511	0.1
29	*α*-Cadinene	1536	2.6
30	Spathulenol	1576	1.3
31	Caryophyllene oxide	1582	3.5
32	*β*-Eudesmol	1649	1.7
33	Tetradecanol	1670	0.1
34	Hexadecanoic acid	1692	Trace[b]
35	Pentadecanal	1711	0.1
6	*n*-Pentadecanol	1777	Trace[b]

[a]Retention indices; relative to *n*-alkane series on DB-5 capillary column. [b]Trace (<0.05%).

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