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Chemical Composition of Essential Oil from *Equisetum ramosissimum*

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Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

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ABSTRACT

Objective: To determine the chemical compositions of essential oil from the aerial parts of *Equisetum ramosissimum* (*E. ramosissimum*).
Methods: The essential oil from the aerial parts of *E. ramosissimum* was detected using gas chromatography -mass spectrometry (GC-MS).
Results: Chemical analyses detected thirty seven compounds.
Conclusions: α-bisabolol Oxide A (12.3%) and cuminaldehyde (9.8%) were identified as major constituents of the oil. Moreover, the presence of carvacrol was unusual as compared to other *Equisetum spp*. Given the ovicidal effect of carvacrol on neonate larva, further investigations are urged to question the toxicological effect of the essential oil of *E. ramosissimum* on reproductive health in animals.

Keywords: Equisetum ramosissimum; essential oil; hydrodistillation; GC-MS.

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1. INTRODUCTION

In recent years, the use of volatile oil in both the food and pharmaceutical industries has increased. Essential oils, which are mixtures of many natural components, are important sources of aromatic and flavoring chemicals in food and in pharmaceutical products [1]. Components of essential oils are mainly terpenes and aromatic polypropanoid which are products of acetate-mevalonic acid and the shikimic acid pathways, respectively [1].

Jordan is rich in medicinal and aromatic plants, which are distributed all over the country [2]. The importance of these plants as a source of preventive or curative health value has been recognized by local people since ancient times [2]. One of these medicinal plants, *Equisetum ramosissimum (E. ramosissimum),* is found in Jordan [3]. In particular, in the south of Jordan, Wadi [4] and Wadi Faynan [5]. Also, it is found in damp places localized near the borders between Jordan and Palestine [6]. *Equisetum* (horsetail) is one of approximately 15–25 surviving hollow stemmed taxa [7]. It is the only link to a group of extinct, diverse and dominant pteridophytes [7]. In fact, *Equisetum* might be regarded as the oldest free-sporing surviving vascular plant [8].

Studies of the scavenger activities of *Equisetum* showed that the genus has high scavenger abilities against free radicals and high total antioxidant activity [9,10]. However, it is thought that the high thiaminase activity found in *Equisetum* may cause neurodevelopmental toxicity through thiamine depletion [11-13], since *Equisetum* is frequently used in pregnancy for preventing edema [14].

To determine the chemical compositions of essential oil from the aerial parts of *E. ramosissimum*, the present study investigated these compositions using GC-MS.

This study reports, for the first time to our knowledge, the chemical composition of the essential oil of *E. ramosissimum.*

2. MATERIALS AND METHODS

2.1 Plant Material

Equisetum ramosissimum was obtained from a local market and authenticated by Dr. Dawud Al-Eisawi, Professor of Botany, Biology Department /The University of Jordan.

A voucher specimen (Eq/214A) has been deposited in the Herbarium of the Department of Biological Science, Faculty of Science, The University of Jordan. The aerial parts of *E. ramosissimum* plant were isolated and ground to 0.5 mm particle size (30-35 mesh).

2.2 Oil Distillation

To obtain the essential oil, ground plant material (500 g) was subjected to hydrodistillation using Clevenger-type apparatus (JSGW, India) for 4 hours. Isolated oil were dried over anhydrous sodium sulfate, Na_2SO_4 (Analar, England), and stored in dry dark glass bottles at 4℃ until analysis. The yield (0.125%) was calculated as percentage volume per weight (% v/w) of the dried plant materials.

2.3 GC/MS Analysis

For each oil sample prepared as above, 5 µL was diluted to 1 mL using GC-grade *n*-hexane (Scharlaue, Barcelona-Spain). Then, 1 µL samples of diluted oil were injected into the GC-FID and GC-MS systems for analysis. A varian chrompack CP-3800 GC/MS/MS-200 equipped with split-splitless injector and DB-5 GC column (5% diphenyl 95% dimethyl polysiloxane), (30 m × 0.25 mm ID, 0.25 µm film thickness) was used. The injector temperature was set at 250℃ with a split ratio of 1:10. Detector and transfer-line temperatures were 160℃ and 230℃, respectively. A linear temperature program was used to separate the different oil components. Temperature programming was applied at 3℃/min heating rate starting from 60℃ (initial temperature) to 250℃ (final temperature) and held at 250℃ for 5 min with a total run time of about 68 min. Each sample was analyzed twice. The mass detector was set to scan ions between 40-400 *m/z* using full scan mode and electron impact (EI, 70 eV). A hydrocarbon mixture of *n*-alkanes (C8-C20) was analyzed separately by GC-MS using the same column (DB-5) and under the same chromatographic conditions. Linear retention index (Arithmetic-Kovats index) was calculated for each component (each peak) separated by GC-MS using the values of its retention time and the retention times of the reference *n*-alkanes applying the Van Den Dool equation [15]. Compounds identification of volatiles was based on computer matching with the WILEY, NIST, and ADAMS libraries, and also by comparing their calculated arithmetic indices with reported values in literature [16]. Identification of certain compounds (e.g. α-Bisabolol Oxide A) was further confirmed by co-chromatography of their authentic standards (Sigma-Aldrich) under the same chromatographic conditions mentioned above.

3. RESULTS AND DISCUSSION

Aerial parts of *E. ramosissimum* were isolated and used to determine the chemical composition of its essential oil. Results show that chemical analyses, utilizing hydrodistillation and GC/MS, detected thirty –seven compounds (Table 1).

Table 1. The composition of essential oil obtained from aerial parts of *Equisetum ramosissimum*

RI_{exp}	RI_{lit}	Compound*	% Content
1075	NA	3,7-dimethyl-1,7-octanediol	0.2
1104	1095	Linalool	1.1
1107	NA	*E*-2-Decen-1-ol	0.6
1184	1161	Neomenthol	0.7
1186	1174	Terpinen-4-ol	0.8
1203	1186	α-terpineol	2.5
1243	1233	Pulegone	1.5
1251	1238	Cumin aldehyde	9.8
1256	1250	*cis*-piperitone epoxide	0.7
1260	1255	Carvenone	1.4
1293	1282	*trans*-Anethole	3.3
1295	1293	2-undecanone	3.6
1307	1289	Thymol	4.1
1315	1298	Carvacrol	7.5
1349	1346	α-Terpinyl- acetate	5.6
1376	1374	α-Copaene	1.2
1406	1403	Methyl eugenol	1.9
1421	1417	β-Caryophyllene	4.7
1425	1428	*E*-α-Ionone	1.0
1450	1453	Geranyl acetone	0.6
1452	1454	*cis*-β-Farnesene	0.9
1457	1452	α-Humulene	0.8
1482	1481	γ-Curcumene	3.7
1491	1489	β-Selinene	0.6
1508	1505	β-Bisabolene	0.9
1524	1423	2*E*,4*E*-Dodecadienol	1.9
1583	1477	Spathulenol	1.2
1586	1582	Caryophyllene oxide	2.7
1625	1620	Dill Apiole	1.2
1659	1656	α-Bisabolol Oxide B	2.1
1669	1668	ar-Tumerone	1.2
1685	1684	α-Bisabolone Oxide A	1.6
1756	1748	α-Bisabolol Oxide A	12.3
1842	-----	Unk	8.0
1889	1890	*trans*-Spiroether	1.5
1980	1959	Hexadecanoic acid	4.6
1993	1992	Ethyl Hexadecanoate	0.3
		Monoterpene Hydrocarbons	5.6
		Oxygenated Monoterpenes	36.1
		Sesquiterpene Hydrocarbons	12.8
		Oxygenated Sesquiterpenes	23.6
		Hydrocarbons and miscellaneous	12.2
		Unk	8.0
		Traces	1.7
		Total identified	90.3

*RIexp: linear (arithmetic) retention index calculated on DB-5 equivalent column; RIlit: reference retention index value from literature. *: Compounds are listed based on their elution order on DB-5 column; trace: <0.01%. NA: data not available in literature; Unk: unknown compound*

Two compounds, α-bisabolol oxide A (12.3%) and cumin aldehyde (9.8%), were found to be major constituents of the essential oil of *E. ramosissimum* (Table 1).

Oil from aerial parts of *E. ramosissimum* is composed mainly of terpenoids with monoterpenoids comprising 41.7% and sesquiterpenoids 36.4%. More than 86% of the monoterpenoids were oxygenated; cuminaldehyde is the dominate oxygenated species. This is also applicable to the sesquiterpenoids in that the oxygenated part was predominant (65%) compared to the non-oxygenated fraction. The principle oxygenated compound in sesquiterpenoids is α-bisabolol oxide A.

Comparing the chemical compositions of essential oil for other *Equisetum* spp. revealed that *Equisetum arvense* have 23.18% monoterpenoids and 14.04% sesquiterpenoids [17]. For *Equisetum palustre, Equisetum telmateia* and *Equisetum fluviatile,* terpenoids total 19.6%, 6.3% and 19.1%, respectively [18,19].

The remarkably high content of monoterpenoids and sesquiterpenoids in the oil of *E. ramosissimum* as compared to other *Equisetum* spp. promotes our interest in further investigating its potential for reproductive toxicity in mammals [20]. Others have observed that monoterpenoids, especially carvacrol, cause reproductive inhibition in insects [21]. Specifically carvacrol was shown to have ovicidal effect on neonate larva [21]. Interestingly, carvacrol is not detected in the essential oil of four other *Equisetum spp* [17-19], but is the fourth highest monoterpenoid at 7.5% in *E. ramosissimum*. Finally, the presence of an unknown compound at 8% motivates research to further characterize this unidentified chemical.

4. CONCLUSION

Of the thirty -seven compounds detected in the oil of *E. ramosissimum*, the high content of monoterpenoids and sesquiterpenoids as well as the presence of carvacrol is unusual as compared to other *Equisetum spp*. Given that carvacrol has been shown to have ovicidal effect on neonate larva, this finding motivates further studies that question the toxicological effect of the essential oil on reproductive health in animals.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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