Identification of Nonpolar Chemical Composition Spartium junceum flower Growing in Iran by GC-MS

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Abstract: The *Spartium junceum* is a medicinal plant that belongs to the fabacea family. The nonpolar chemical composition of it was investigated and fifty nine compounds were identified by GC-MS. The main constituents were the n-Hexadecanoic acid (14.27%), 9, 12, 15-Octadecatrien-1-ol(13.07%), Tetradecanoic acid (6.59%) Octadecanoic acid (3.68%) and γ -Sitosterol(3.67%).

Key words: Chemical composition, N-Hexane extraction, N-Hexadecanoic Acid, GC-MS, Spartium junceum

INTRODUCTION

Medicinal plants are believed to be an important source of new chemical substances with potential therapeutic effects. Thus study of plant species that traditionally have been used as pain killer should still be seen as a strategy in research for new analgesic drugs. The Spartium junceum belongs to the Fabaceae family,is asmall shrub indigenous in the Mediterranean countries and cultivated as an ornamental plant.Its flowers are large, vellow and by greeable scent [1]. S. junceum Contains flavenoids and saponins. Two new flavenoids and saponin were isolated from its flowers in astudy by Bilia et al. [2]. Another study showed that S.junceum seeds contain a large amount of lectin [3]. An herbal tea (known as Zahraa) widely consumed in Syria contains 6-14 species components including *S. junceum* [4].

Flowers of *S. junceum* are used for the treatment of gastric ulcers in Turkish flok medicine [5]. It has been showed in a study by Yesilada *et al.*, [6] that *S. junceum* did not have anti-helicobacter pylori activity. Flovenoids-rich fractions from the flowers *S. junceum* showed potent antioxidant activity reported by Yesilada *et al.* [7].

Its know to simulate uterine contractions and GI tract, help body to dispose excess fluid by increasing amount of urine and cause vomiting [8]. It has been studied for its antifertility activity in mammalian male [9].

Another study showed injection of *S. junceum* extract in adult male rats reduce the rate of fertility and acrosin enzyme activity [10]. Only one study reported the antinociceptive And anti-inflammatory effects of this plant [11]. Keeping this in view, the present study has been done to identification nonpolar compounds of n-hexane extract of this plant.

MATERIALS AND METHODS

The experimental protocol used in this study was approved by the research committee of North khorasan payame Noor University, Bojnourd, Iran.

Plant Material: Flower of *Spartium junceum* were collected at the flowering stage from the Bojnourd, Iran, In Jun 2011 and identified at the Research Center for Plant Sciences, Ferdowsi University of Mashhad, Iran. A voucher specimen has been deposited in the Enveriomental Department of Bojnourd Herbarium (EDBH:00107).

Isolation of Chemical Compounds: Flowers of *Spartium junceum* were air-dried for 3 days before isolation of chemical composition. The plant material (100g) was cut into small pieces. The plant powdered was macerated in 95% pure n-hexane as a solvent for 48 hours, filtered through a Wattman paper, then evaporated off the solvent in vacuum by rotary Evaporator to yield light

yellow oil and dried over by adding anhydrous NaSO₄. In absolute oil recovery, concentrate oil was dissolved in minimum volume of absolute alcohol to remove the natural waxes present in the essential oil. It was kept at in-14°C for 48 hrs and then it was filtered through a filter paper. Alcohol was removed by distillation and traces of alcohol were removed by passing nitrogen gas through it [12].

Chromatography and Mass Spectrometry: Gas chromatographic analysis was performed on an Hewlett-Packard(HP)6890A instrument equipped with a flame ionization detector and Rtx-5MS $(15 \text{ m} \times 0.25 \text{ mm} \times 0.25 \text{ } \mu\text{m})$ capillary column, while the essential oil components were identified on an Agilent Technologies 5973N mass spectrometer. The GC settings were as follows: the initial oven temperature was held at 35 °C for 6 min and ramped at 5 °C min⁻¹ to 150 °C for 0 min and then ramped at 10 °C min⁻¹ to 280 °C for 3 min. The injector temperature was maintained at 250 °C. The samples (1 μ L) were injected, with a split ratio of 1:10. The carrier gas was helium at flow rate of 1.0 mL min⁻¹. Spectra were scanned from 20 to 550 m/z at 2 scans s⁻¹. Most constituents were identified by gas chromatography by comparison of their retention indices with those of the literature or with those of authentic compounds available in our laboratories. The retention indices were determined in relation to a homologous series of *n*-alkanes under the same operating conditions. Further identification was made by comparison of their mass spectra on both columns with those stored in NIST 05 and Wiley 275 libraries or with mass spectra from literature [13-18]. Component relative percentages were calculated based on GC peak areas without using correction factors.

RESULTS AND DISCUSSION

The average yield of chemical composition of the leaves of *Spartium junceum* was about 0.58%. Table 1 reports the chemical composition of the phytochemical components under study. Fifty nine components were identified, accounting for %76.27 of the total oil.

The various compounds were identified by comparison of their Kováts retention indexes, determined utilizing a non-logarithmic scale on non-polar (Rtx-5MS) columns and by comparison of the mass spectra of each GC component with those of standards and with reported data [19].

Table 1: Percentage composition of the essential oil isolated from aerial parts of spartium junceum

NO	compound	Experimentally determined Ki ^a	HP GC-MS Peak area [%]	Method of identification
1	E thyl benzene	850	0.28	GC-MS,Ms
2	1,3-dimethyl-benzene	866	0.29	GC-MS,Ms
3	Mesitylene	995	0.14	GC-MS,Ms
4	Decane	1000	0.13	GC-MS,Ms
5	Phenylethyl Alcohol	1107	0.25	GC-MS,Ms
6	Benzothiazole	1216	0.17	GC-MS,Ms
7	2-Propenoic acid, 3-phenyl-, methyl ester	1380	0.11	GC-MS,Ms
8	Pentadecane	1499	0.14	GC-MS,Ms
9	2,3,6-trimethyl-Naphthalene	1550	0.11	GC-MS,Ms
10	Dodecanoic acid	1564	1.64	GC-MS,Ms
11	Dodecanoic acid, ethyl ester	1592	0.47	GC-MS,Ms
12	Hexadecane	1598	0.44	GC-MS,Ms
13	Pentadecane, 2,6,10,14-tetramethyl-	1649	0.21	GC-MS,Ms
14	Heptadecane	1663	0.08	GC-MS,Ms
15	2,6,10,14-tetramethyl-Pentadecane	1705	0.35	GC-MS,Ms
16	Phenol, 2-(1-phenylethyl)-	1721	0.41	GC-MS,Ms
17	Tetradecanoic acid	1763	6.59	GC-MS,Ms
18	Phenanthrene	1784	0.4	GC-MS,Ms
19	Tetradecanoic acid, ethyl ester	1792	1.47	GC-MS,Ms
20	Octadecane	1798	0.75	GC-MS,Ms
21	Hexadecane, 2,6,10,14-tetramethyl-	1808	0.65	GC-MS,Ms
22	Tetradecanoic acid,trimethylsilyl ester	1850	0.16	GC-MS,Ms
23	Pentadecanoic acid	1857	0.21	GC-MS,Ms
24	Dibenzothiophene, 3-methyl-	1862	0.19	GC-MS,Ms
25	1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester	1869	0.45	GC-MS,Ms
26	Nonadecane	1897.4	0.54	GC-MS,Ms
27	1,2-Benzenedicarboxylic acid, butyl 2-ethylhexyl ester	1915	0.11	GC-MS,Ms
28	Hexadecanoic acid, methyl ester	1924	0.3	GC-MS,Ms
29	n-Hexadecanoic acid	1964.15	14.27	GC-MS,Ms

Table 1: Continue

NO	compound E	xperimentally determined Kia	HP GC-MS Peak area [%]	Method of identification
30	Hexadecanoic acid, ethyl ester	1992.4	2.34	GC-MS,Ms
31	Eicosane	1999	0.48	GC-MS,Ms
32	Cyclic octaatomic sulfur	2055	0.61	GC-MS,Ms
33	1-Octadecanol	2082	0.18	GC-MS,Ms
34	14-beta –H-PREGNA	2091	0.14	GC-MS,Ms
35	Heneicosane	2097	0.51	GC-MS,Ms
36	E-15-Heptadecenal	2104	0.08	GC-MS,Ms
37	Methyl stearate	2024.7	0.05	GC-MS,Ms
38	9,12,15-Octadecatrien-1-ol	2042	13.07	GC-MS,Ms
39	Octadecanoic acid	2161	3.68	GC-MS,Ms
40	9,12,15-Octadecatrienoic acid, ethyl ester, (Z,Z,Z)-	2169	2.05	GC-MS,Ms
41	Octadecanoic acid, ethyl ester	2192.5	0.58	GC-MS,Ms
42	Docosane	2197.8	0.74	GC-MS,Ms
43	1-chloro-7-Heptadecene	2206.7	0.18	GC-MS,Ms
44	Benzene, 1,1'-sulfonylbis[4-chloro-	2248	0.07	GC-MS,Ms
45	Methyl dehydroabietate	2361	0.39	GC-MS,Ms
46	Tetracosane	2398.8	1.16	GC-MS,Ms
47	p-Menth-8(10)-en-9-ol	2418	0.05	GC-MS,Ms
48	2,4-bis(1-phenylethyl)phenol	2425.6	1.23	GC-MS,Ms
49	Dehydroabietic acid	2457	1.6	GC-MS,Ms
50	Pentacosane	2497.5	3.18	GC-MS,Ms
51	Bis(2-ethylhexyl) phthalate	2550.6	0.57	GC-MS,Ms
52	Hexacosane	2597	0.33	GC-MS,Ms
53	Heptacosane	2698.6	0.97	GC-MS,Ms
54	Squalene	2831.8	0.74	GC-MS,Ms
55	4-(benzylamino)-1,3-diphenyl-5,6,7,8-tetrahydroquinolin-2(1H)	-one 3045	1.98	GC-MS,Ms
56	Vitamin E	3142	0.69	GC-MS,Ms
57	Campesterol	3243.7	0.98	GC-MS,Ms
58	Stigmasterol	3274.7	2.66	GC-MS,Ms
59	γ-Sitosterol	3790	3.67	GC-MS,Ms
Total:	76.27			•

a: Retention Indices on RTX-5MS

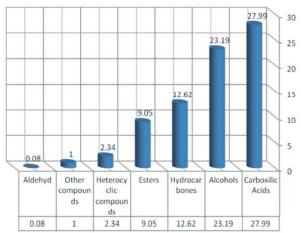


Chart 1: Percentage of compounds find in essential oil of Spartium junceum

High resolution gas chromatography-mass spectrometric (HP GC-MS) analysis and Kováts Index values showed that its principal components are the n-Hexadecanoic acid (14.27%), 9,12,15-Octadecatrien-1-ol (13.07%),

Tetradecanoic acid (6.59%) Octadecanoic acid (3.68%) and γ -Sitosterol (3.67%).

chart 1 shows that number of carboxilic acid compounds as the largest group of compound in the n-hexane extract.

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REFERENCES

- 1. Yesilida, E. and Y. Takaishi, 1999. A saponin with anti-ulcerogenic effect from the flowers os *Spartium junceum*, Phytochemistry, 51: 903-908.
- Bilia, A.R., F. Flammini, G. Flammini, I. Morelli and A. Marsili, 1993. Flavonoids and saponin from *Spartium junceum*, Phytochemistry, 34: 847-852.

- 3. Hankins, C.N., E.M. Herman, J. Kindinger and L.M. Shannon, 1991. The purification, Properties and Localization of an Abundant Legume Seed Lectin Cross-Reactive Material from *Spartium junceum*, Plant Physiol., 96: 98-103.
- Carmona, M.D., R. Llorach, C. Obon and D. Rivera, 2005. Zahraa, a Unani multicomponent herbal tea widely consumed in Syria: Components of drug mixtures and alleged medicinal properties. J. Ethnopharmacology, 102: 344-350.
- Yes, ilada, E., Y. Takaishi, T. Fujita and E. Sezik, 2000. Anti-ulcerogenic effects of *Spartium junceum* flowers on *in vivo* test models in rats.
 J. Ethnopharmacology, 70: 219-226.
- Yes, ilada, E., I. Gu"rbu"z and H. Shibata, 1999.
 Screening of Turkish anti-ulcerogenic folk remedies for anti-Helicobacter pylori activity.
 J. Ethnopharmacology, 66: 289-293.
- Yes, ilada, E., K. Tsuchiya, Y. Takaishi and K. Kawazoe, 2000. Isolation and characterization of free radical scavenging flavonoid glycosides from the flowers of *Spartium junceum* by activity-guided fractionation. J. Ethnopharmacology, 73: 471-478.
- 8. Baytop, T., 1984. Phytotherapy in Turkey, Past and Present, Istanbul University Press, Istanbul.
- Baccetti, B., A.G. Burrini, J.S. Chen, G. Collodel, D. Giachetti, F. Matteucci, M.G. Menesini-Chen, E. Moretti, P. Piomboni and C. Sensini, 1993. Evalution of the antifertility of the broom Spartium junceum in the mammalian male, Zygote., 1: 71-78.
- Chen, J.S., M.G. Menesini-Chen, D. Giachetti, F. Matteucci, M. Barbetti, C. Sensini and B. Baccetti, 1993. Correlation between male fertility and acrosin-like protease activity in rats treated with Spartium junceum, Zygote., 1: 309-313.

- Menghini, L., P. Massarelli, Gi. Bruni and R. Pagiotti, 2006. Anti-Inflammatory and Analgesic Effects of Spartium junceum L. Flower Extracts: A Preliminary Study. J. Med. Food, 9: 386-390.
- Aslam, M. Khan and Shoaib-UR-Rehman, 2005.
 Extraction and Analysis of Essential Oil of *Rosa species*. Int. J. Agri. Biol., 7(6): 973-974.
- Adams, R.P., 2001. Identification of Essential Oil Components by Gas Chromatography/Quadrupole Mass Spectroscopy; Allured: Carol Stream, IL, USA.
- Pino, J.A., J. Mesa, Y. Munoz, M.P. Marti and R. Marbot, 2005. Volatile components from mango (Mangifera indica L.) cultivars. J. Agric. Food Chem., 53: 2213-2223.
- Bianchi, F., M. Careri, A. Mangia and M. Musci, 2007.
 Retention indices in the analysis of food aroma volatile compounds in temperature-programmed gas chromatography: database creation and evaluation of precision and robustness. J. Sep. Sci., 30: 563-572.
- 16. Kohl, E., B. Hölldobler and H.J. Bestmann, 2001. Trail and recruitment pheromones in Camponotus socius (Hymenoptera: Formicidae). Chemoecology, 11: 67-73.
- Dos Santos, P.R.D., D.L. Moreira, E.F. Guimaraes and M.A.C. Kaplan, 2001. Essential oil analysis of 10 Piperaceae species from the Brazilian atlantic forest. Phytochem, 58: 547-551.
- Davies, N.W., 1990. Gas chromatographic retention indices of monoterpenes and sesquiterpenes on methyl silicone and Carbowax 20M phases. J. Chromatogr. A., 503: 1-24.
- Davies, N.W., 1990. Gas chromatographic retention indices of monoterpenes and sesquiterpenes on methyl silicone and Carbowax 20M phases. J. Chromatogr. A., 503: 1-24.