Arab Gulf J. Scient. Res., Agric. Biol. Sci., **B6** (3), pp. 389-398 (1988)

Aromatic Plants of Saudi Arabia Part IX GC/MS Analysis of Essential Oil of *Triumfetta flavescens*

Jaber S. Mosa, Ibrahim A. Al-Meshal, Mohammed A. Al-Yahya and Mohamed S. Hifnawy

Department of Pharmacognosy and Medicinal, Aromatic and Poisonous Plants Research Centre, College of Pharmacy, King Saud University, P.O. Box 2457, Riyadh 11451, Saudi Arabia

ABSTRACT. The steam distilled oil of the aerial parts of *Triumfetta flavescens* Hochst. ex. A. Rich was analyzed by the coupled technique GC/MS. By the aid of the electronic data system and the study of the mass spectra, up to 63 components could be identified and the chemical classes of another 28 constituents were deduced. These ingredients amounted for about 96% of the total oil content. The oil is rich in sesquiterpene hydrocarbons and their oxygenated derivatives, most of which belong to the azulene series. Terpene alcohols *viz.* linalool, 4-terpineol, *cis*-piperitol and cuminol together with other common volatile oil constituents were found to be present in the investigated sample.

Triumfetta flavescens Hochst. ex A. Rich is an aromatic shrub belonging to the Family Tiliaceae. The plant is of yellow-green aspect with stem dark red, hairy and covered with black dots. The leaves are rounded, irregularly toothed, densly canescent, with long petioles (Migahid 1978).

This plant is of folk use in Saudi Arabia, its aerial parts are hanged in the entrances of houses to repel insects and mosquitoes.

Reviewing the current literature, nothing could be traced dealing with the chemistry of the plant. On the other hand, triumferol (4-hydroxyisoxazole), a seed germination inhibitor and triumboidin, a flavone glycoside were isolated from another species, namely *T. rhomboidea* (Nakanishi 1980, Kusumi *et al.* 1981, Srinivasan and Subramanian 1981 and Nair *et al.* 1986). Concerning the study of essential oil, the only work on members of the Tiliaceae was that reported by

Guenther 1952 who gave the physico-chemical constants of blossoms of linder trees (*Tilia cordata* Mill., *T. tomentosa* and *T. platyphyllos* Scop.). However, a preliminary unpublished neuropharmacological screening, carried out in our laboratories, showed that the chloroformic extract of the aerial parts of *T. flavescens* produced slight hyperthermic effect along with tremors and excitability while the ethanolic extract produced depression of CNS and respiratory system accompanied by mild hypothermia of experimental animals. The latter extract showed as well a significant antimicrobial activity against *Bacillus subtilis*.

The present article deals with the investigation of the essential oil of *T. flavescens*, as a part of our continuous programme for studying the aromatic plants of Saudi Arabia with the aim of declarion of its possible economical and biological potentialities and to explain the folklore use of the plant in Saudi Arabia.

Experimental

Plant Material

Triumfetta flavescens was collected in the flowering stage (March 1983) from Al Madina area in the Western region of Saudi Arabia. Plant identity was confirmed through the Taxonomy section of the Medicinal, Aromatic and Poisonous Plants Research Centre, College of Pharmacy, King Saud University, Riyadh. A voucher specimen representing the collection was kept in the herbarium of the same institute.

The oil was prepared by steam distillation of the fresh aerial parts and the content was calculated as percent of the fresh weight (B.P. 1980).

Chromatographic Analysis

A Ribermag - Riber R10-10 GC/MS (France) apparatus was used and the oil was analyzed under the following operating conditions: Type of column and phase: FS-57 CB; column specifications: coiled glass, 25 m, 2-3 mm i.d.; carrier gas, He; input carrier gas pressure: 1 bar; flow rate: 2 ml/min. oven temperature: 70 to 220°C with a rate of 2°/min.; injection mode: split; ionization mode: electron impact (EI) at 70 eV; interface temperature: 300°C. Mass spectral analysis was monitored according to the Flame Ionization (FID) detected components.

Qualitative identification was based on comparing the resulted mass spectra with those of standards as well as by matching them with those given by the electronic data system and with reported spectra (Ryhage and von Sydow 1963, von Sydow 1963, 1964, 1965, Moshonas and Lund 1970, Nose *et al.* 1971, Enzell *et al.* 1972, and Stenhagen *et al.* 1974).

Quantitative analysis was based on the peak area measurements and the percentages were calculated relatively.

Results and Discussion

This is the first report on *T. flavescens* as one of the aromatic plants belonging to the Tiliaceae. The oil content reached up to 0.24% v/w calculated on fresh weight basis of the aerial parts including leaves, flowers and small branches. The oil of this species possesses a strong persistant and characteristic aroma.

The GC/MS analysis of the oil shows the presence of at least 91 compounds of which 63 have been identified and the chemical classes of the other 28 were recorded. These amounted to about 96% of the total content of the oil (Tables 1 and 2). The oil is rich in C_{15} compounds including sesquiterpene hydrocarbons and alcohols.

From the first class 21 compounds were identified representing about 33.5% of the total oil constituents. α-caryophyllene, muurolene, α-cubebene, α-himachelene, and L-aromadendrene are of prominent occurence. The sesquiterpene alcohols are represented by $C_{15}H_{24}O$ (M^+220) and $C_{15}H_{26}O$ (M^+222) compounds, amounting for 22.5 and 12.4% of the oil content respectively. α- and β-cedrol, ledol, α-copaen-11-01, widdrol, △-cadinol, selinen-4 α-01, palustrol and α-caryophyllene alcohol are the most abundant.

According to the interpretation of the mass spectra and the main fragments of each one, some of the resolved peaks are classified as sesquiterpene compounds by referring to the published work on the general behaviour of sesquiterpenes in mass spectrometry; none of them, however, coincide with the available spectra and thus are presented in the tables as a class and not as identified compounds. These include 6 sesquiterpene hydrocarbons, 17 sesquiterpene alcohols and 5 azulene derivatives. Moreover, terpenes and other common volatile oil constituents *viz* α- and β-linalool, 4-terpineol, *cis*-piperitol, cuminol, piperitone, ionones, farnesyl acetate and civetone (known to be one of the main constituents in aromatic products from certain animals), show themselves in moderate amounts in the oil under investiation.

In conclusion, most of the identified compounds in the oil of *T. flavescens* Hochst ex A. Rich are belonging to the azulene series and some of them are related to benzene and naphthalenone moieties, a fact which explains the folk use of this herb as an efficient and long acting natural insect-and mosquitoes-repellant as well as its significant anti-microbial effect. Also, according to the chemical nature of the oil constituents and the moderate oil content of the plant, it can economically serve as natural aroma-fixative in high grade perfumes.

Table 1. GC/MS analysis of essential oil of *T. flavescens*

Peak No.	RT	Mass spectra			Formula	Compound	% (rel.)
		M^+	100%	Main fragments			
1	8.6	142	68	57,85,41	$C_9H_{10}O$	Nonaldehyde (pelargonic aldehyde)	2.1
2	11.55	204	105	119,161	$C_{15}H_{24}$	α-cubebene	1.7
3	13.1	204	105	161,119,93	$C_{15}H_{24}$	α-Copaene	0.5
4	14.1	204	81	80,123,161	$C_{15}H_{24}$	β-Bourbonene	Tr.
5	15.1	204	161	105,91,119	$C_{15}H_{24}$	β-cubebene	Tr.
7	16.5	154	71	93,55,69	$C_{10}H_{18}O$	α-Linalool	0.1
8	17.17	204	69	93,79,133	$C_{15}H_{24}$	Bicyclo -7.2.0-undeca-4-ene, 4,11,11-trimethyl, 8-methylene	3.7
9	18.06	154	71	111,93,69	$C_{10}H_{18}O$	4-Terpineol	2.1
10	18.33	204	93	121,107,161	$C_{15}H_{24}$	α-Guaiene	0.5
11	19.05	154	43	93,69,139	$C_{10}H_{18}O$	β-Linalool	0.2
12	19.16	204	93	80,121,147	$C_{15}H_{24}$	α-Humulene (α-Caryophyllene)	2.8
13	19.55	204	161	105,119,93	$C_{15}H_{24}$	α-Himachalene	0.8
14	20.22	204	161	93,105,136	$C_{15}H_{24}$	Muurolene	3.7
15	20.41	204	161	93,105,119	$C_{15}H_{24}$	γ-Cadinene	0.3
16	20.47	204	41	81,79,107,133	$C_{15}H_{24}$	Alloaromadendrene	Tr.
17	21.01	204	105	59,93,81	$C_{15}H_{24}$	Muurolene isomer	1.1
18	21.1	204	93	121,107,161	$C_{15}H_{24}$	Benzocycloheptene derivative	0.5

Table 1. GC/MS analysis of essential oil of *T. flavescens*

Peak No.	RT	Mass spectra			Formula	Compound	% (rel.)
		M_i,	100%	Main fragments			
19	21.18	152	82	110,137,95	$C_{10}H_{16}O$	Piperitone	
20	21.52	204	161	105,119,91	$C_{15}H_{24}$	γ-Muurolene	0.8
21	21.56	204	161	119,105,134	$C_{15}H_{24}$	α-Cubebene isomer	0.9
22	22.18	204	79	161,107,93	$C_{15}H_{24}$	α-Guaiene isomer	Tr.
23	22.28	154	84	93,139,91	$C_{10}H_{18}O$	*cis*-Piperitol	0.1
24	22.35	204	93	119,109,80	$C_{15}H_{24}$	α-Humulene isomer	0.4
26	23.51	204	121	93,105,81	$C_{15}H_{24}$	L-Alloaromadendrene	3.7
27	24.06	174	159	69,105	$C_{13}H_{18}$	α-Ionene	1.2
28	25.28	150	43	135,91	$C_{10}H_{14}O$	Cuminol	0.3
29	26.31	204	157	161,142,105	$C_{15}H_{24}$	β-Cadinene	1.0
30	26.56	206	79	43,106,91	$C_{15}H_{26}$	Patchoulane	0.9
31	27.13	222	161	119,105,120,43	$C_{15}H_{26}O$	α-Cedrol	0.5
32	27.18	182	126	111,70,84	$C_{11}H_{18}O_2$	1-Oxaspiro 4,4 nonan-4-one, 2-isopropyl	
33	27.48	192	177	43,135,157	$C_{13}H_{20}O$	β-Ionone	0.5
34	28.12	222	161	109,105,119	$C_{15}H_{25}O$	β-Cedrol	0.7
38	29.47	204	93	80,109,121,138	$C_{15}H_{24}$	β-Selinene	Tr.
40	30.46	208	81	123,79,161	$C_{14}H_{24}O$	Luciferin aldehyde	0.2
42	31.11	222	161	119,179,204	$C_{15}H_{26}O$	β-Cadinol	0.6
43	31.20	222	119	161,179,204	$C_{15}H_{26}O$	α-Cedrol isomer	0.5
45	32.08	222	43	69,81,109,161	$C_{15}H_{26}O$	D. Ledol	0.5
48	32.59	206	59	93,79,149	$C_{13}H_{18}O_2$	3 Keto β-Ionone	0.2
49	33.35	220	43	91,119,205	$C_{15}H_{24}O$	α-Copaen-11-01	0.9

Table 1. (Continued)

Peak No.	RT	Mass spectra			Formula	Compound	% (rel.)
		M^+	100%	Main fragments			
50	33.49	206	43	79,81,93,122	$C_{15}H_{26}$	Isopatchoulane	0.1
51	43.11	222	179	161,55,119	$C_{15}H_{26}O$	Widdrol	0.7
52	34.59	204	161	43,81,105	$C_{15}H_{24}$	γ-Muurolene	2.2
53	35.31	222	95	43,121,161	$C_{15}H_{26}O$	△-Cadinol	2.2
57	36.55	222	95	121,161,204	$C_{15}H_{26}O$	△-Cadinol isomer	3.7
58	37.21	222	81	71,135,204	$C_{15}H_{26}O$	Selinen 4-α01	0.3
59	38.29	220	159	177,91,81	$C_{15}H_{24}O$	α-Copaen-11-01	0.4
64	40.03	206	41	69,91,122,149	$C_{13}H_{18}O_2$	Ionone derivative	0.7
66	40.31	290	159	109,79,202	$C_{19}H_{30}O_2$	Fatty acid methyl ester	0.8
67	40.47	240	157	200,142,43	$C_{18}H_{24}$	Aromatic compound	2.6
72	45.27	192	43	153,135,111	$C_{13}H_{20}O$	β-Ionone isomer	0.9
73	45.35	264	69	81,93,107	$C_{17}H_{28}O_2$	Trans Farnesyl acetate	0.9
76	46.46	220	43	93,107,162	$C_{15}H_{24}O$	Naphthalenone derivative	0.4
77	46.58	264	41	139,123,91	$C_{17}H_{28}O_2$	Sesquiterpene acetate	0.8
78	47.16	206	41	55,81,191	$C_{15}H_{26}$	Isopatchulane	3.2
80	47.49	206	71	43,55,69	$C_{17}H_{30}O$	Civetone	0.5
81	48.05	302	43	123,79,107	$C_{19}H_{26}O_3$	Allyl cinerin	2.6
83	50.37	222	122	55,148,166	$C_{15}H_{26}O$	Palustrol	0.3
84	52.11	248	81	55,107,121	$C_{15}H_{20}O_3$	Confertin	0.4
85	52.44	220	43	55,81,107	$C_{15}H_{24}O$	Naphthalenone derivative	3.0

Peak No.	RT	Mass spectra			Formula	Compound	% (rel.)
		M_i,	100%	Main fragments			
86	55.02	266	43	195,81,159	$C_{15}H_{22}O_4$	Ambrosiol	1.1
87	56.06	222	43	175,109,193	$C_{15}H_{26}O$	α-Caryophyllane alcohol (11-Apollanol)	1.1
89	58.30	208	123	81,93,107	$C_{14}H_{24}O$	Luciferin aldehyde	2.1
90	1.00	248	43	55,71,162,190	$C_{15}H_{20}O_3$	Tomentosin	1.2
6,25, 36,54, 56,61	variable according to peak number	204		Difference characteristic fragments	$C_{15}H_{24}$	Unidentified sesquiterpene hydrocarbons	6.9
37,39, 55,60, 62,63, 65,68, 69,70, 71,74, 75,79, 82,88, 91	variable according to peak number	220		Difference characteristic fragments	$C_{15}H_{24}O$	Unidentified sesquiterpene alcohols	18.8
35,41, 44,46, 47	variable according to peak number	206 222		Difference characteristic fragments	$C_{15}H_{26}$, $C_{15}H_{26}O$	Unidentified azulene compounds	5.5

* m/z = 3-4 diagnostic fragments arranged according to their relative intensity.
RT = Retention time measured in minutes.

Table 2. Major classes of constituents in oil of *T. flavescens*

Class of compounds	Mol. formula	Number	% rel.
Sesquiterpene hydrocarbons	$C_{15}H_{24}$	21	33.5
Sesquiterpene alcohols	$C_{15}H_{24}O$	22	22.5
	$C_{15}H_{26}O$	16	12.4
Others	Different formulaes	more than 32	31.5

References

B.P. (1980) The British Pharmacopoeia., London, Her Majesty's Stationary Office. A 108.

Enzell, C.R., Appleton, R.A. and **Wahlberg, I,** (1972) Terpenes and terpenoids, in: **Waller, G.R.** (ed.) *Biochemical Application of Mass Spectrometry*, Wiley Interscience, N.Y. pp. 352-369.

Guenther, E. (1952) *The Essential Oils*, D. Van Nostrand Comp. INC., Toronto, Vol. 5, p. 421.

Kusumi, T., Chang, C.C., Wheeler, M., Kubo, S., Nakanishi, K., and **Naoki, H.** (1981) Isolation, Structure, and synthesis of 4-hydroxyisoxazole (Triumferol), a seed germination inhibitor, from an African Plant, *Tetrahedron lett.* **22**(36): 3451-3454.

Migahid, A.M. (1978) *Flora of Saudi Arabia*, Riyadh University Publication, 2nd ed., 113.

Moshonas, M.G. and **Lund, E.D.** (1970) The mass spectra of sesquiterpene hydrocarbons, *The Flavor Industry* 375-378.

Nair, A.G.R., Seetharaman, T.R., Voirin, B., and **Favre-Bovin, J.** (1986) True Structure of Triumboidin, a flavone glycoside from Triumfetta rhomboidea, *Phytochem.* **25**(3): 768-769.

Nakanishi, K. (1980) Isolation and structures of insect and plant growth regulators: Appliations of droplet counter-current chromatography, 2-D proton NMR, and a new circular dichroic correction, *Chem. Nat. Prod. Proc. Sino-Am. Symp.* 9-18.

Nose, M., Naktani, Y. and **Yamanishi, T.** (1971) Studies on the Flavor of Green Tea, Part IX, Identification and composition of intermediate and high boiling constituents in green tea, Agr. Biol. Chem. **33:** 261-271.

Ryhage, R. and **von Sydow, E.** (1963) Mass spectrometry of terpenes, 1-monoterpene hydrocarbons, *Acta Chemica Scand.* **17:** 2025-2035.

Srinivasan, K.K. and **Subramanian, S.S.** (1981) Triumboidin, a new flavone glycoside from the leaves of *Triumfetta rhomboidea, Fitoterapia* **52**(6): 285-287.

Stenhagen, E., Abrahamsson, S. and **McIlafferty, F.W.** (1974) Registry of Mass Spectral Data, Vol. 1 & 2, Wiley, New York, pp. 1025-1027.

von Sydow, E. (1963) Mass Spectrometry of Terpenes, II. Monoterpene Alcohols, *Acta Chemica Scand.* **17:** 2504-2512.

von Sydow, E. (1964) Mass Spectrometry of Terpenes, III Monoterpene Aldehydes and Ketones, *ibid* **18:** 1099-1104.

von Sydow, E. (1965) Mass Spectrometry of Terpenes, IV. Esters of Monoterpene Alcohols, *ibid* **19:** 2083-2088.

(Received 05/09/1987; in revised form 28/02/1988)

النباتات العطرية في المملكة العربية السعودية الجزء التاسع ـ تحليل الزيت العطري لنبات الرشا

جابر بن سالم موسى و إبراهيم بن عبد الرحمن المشعل
و محمد بن عبد العزيز اليحيى و محمد سعيد حفناوي

قسم العقاقير ومركز أبحاث النباتات الطبية والعطرية والسامة ـ كلية الصيدلة ـ جامعة الملك سعود
ص. ب ٢٤٥٧ الرياض ١١٤٥١ ـ المملكة العربية السعودية

يعتبر نبات الرشا من النباتات العطرية ذات الاستعمال الشعبي في عدد كبير من مناطق المملكة حيث تعلق الأجزاء الهوائية للنبات في مداخل البيوت لطرد الذباب والبعوض. ويشمل هذا البحث دراسة وتحليل الزيت العطري لهذا النبات كجزء من الدراسات المستمرة في هذا المجال على النباتات العطرية للمملكة العربية السعودية ـ وقد جمع النبات من منطقة المدينة المنورة وتم تحضير الزيت بالتقطير بالبخار حيث وصلت نسبته إلى حوالي ٠,٢٤٪ بالنسبة للوزن الطازج للأجزاء الهوائية (الأوراق والأزهار والأغصان الصغيرة).

وبتحليل الزيت بوساطة جهاز كروماتوجرافيا الغاز المدمج مع مطياف الكتلة تم التعرف على ٩١ نوعاً من المركبات المميزة لهذا النبات. ووصلت نسبة المواد التي تم التعرف عليها أو على المجموعات الكيميائية التي تنتمي إليها إلى حوالي ٩٦٪ من المحتويات الكلية للزيت العطري حيث وجد أن المواد السيسكوتيربينية والأزولينات تمثل الجزء الأساسي في عطر نبات الرشا، وهذه المواد تمتاز بقوة رائحتها وثبات تركيبها وقد يعزى إليها التأثيرات الشعبية لهذا النبات من ناحية استعماله كطارد للحشرات. وكذلك فإن لهذه المواد تأثيرات طبية كمواد مضادة لبعض الميكروبات. وإلى جانب المواد السابقة فقد تم التعرف أيضاً على بعض المواد التربينية الشائعة في الزيوت العطرية مثل اللينالول والتربينول والبيربتول.

ويشير البحث في النهاية إلى إمكانية استعمال الزيت العطري لنبات الرشاد السعودي كبديل لبعض المثبتات الحيوانية في العديد من المستحضرات العطرية وذلك لطبيعة ما يحتويه من مواد عطرية.