

Effect of Geographic Location, Ontogenesis on Essential Oil Composition and Spontaneously Emitted Volatile Organic Compounds of *Inula viscosa* (L.) Greuter. (Astraceae) Grown in Jordan

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The present study aimed at investigating the variation in the chemical composition of emitted volatile organic compounds (VOCs) and essential oils obtained from fresh aerial parts of *Inula viscosa* (Asteraceae) as affected by growth stage and geographical location zone. VOCs were extracted by solid phase microextraction (SPME) method from whole aerial parts of *I. viscosa* during the vegetative (spring-August) period, and from fresh leaves, closed pre-flowering buds and fully expanded flowers during the full flowering season (October). The essential oils were extracted by hydrodistillation form fresh organs during the flowering stage. GC/MS analysis of VOCs during the vegetative ereid revealed sesquiterpene hydrocarbons as main components in the samples collected from the Mediterranean zone (MID-1: 64.72-27.48%, MID-2: 32.09-76.77%). The profile of the samples from the Irano-Turanian zone (IT) was quite different. Myrcenol was the main component in the leaves, pre-flowering buds and fully expanded flowers from all locations (0.61-39.01%). E-nerolidol was the main component in the hydrodistilled oil of all organs from the different locations (73.72-88.66%). Principle component and cluster analysis revealed that the chemical composition of the essential oils and VOCs belonging to MID-zone were similar and quite different when compared to the composition of the samples belonging to IT-zone. The results indicated the possible use of E-nerolidole as a stable chemotype marker in *I. viscosa* taxonomy.

Keywords: Inula viscosa L., Volatile organic compounds, Geographical variation, Principle component analysis, Cluster analysis.

INTRODUCTION

Inula viscosa (L.) Greuter. (known also as *Dittrichia viscosa*), belonging to the Asteraceae family (formerly known as Compositeae) is a perennial shrubby leafy aromatic plant, reported to grow wild in different geographical regions in Jordan including the Mediterranean (MID) and Irano-Turanian (IT) zones [1]. Flowering occurs, during the period extending from late summer (August) until early winter season (December). This indigenous plant is found along the road sides and waste

places of Irbid, Ajloun, Jerash, Al-Salt, Amman, Madaba, Al-Karak, Tafila, Zarqa cities, Jordan valley and Dead sea.

I. viscosa is commonly used in the traditional medicine of different cultures [2-5] for the treatment of many ailments, especially arthritis and rheumatism. Moreover, in Jordan, a hot beverage prepared from the flowers is prescribed by traditional healers for the treatment of chronic cough and tumors [6-8]. Several studies described the phytochemical constituents and biological activities of *I. viscosa* [6-8].

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In the present study, the emission profiles of *I. viscosa* collected from three different locations belonging to two different bio-geographical zones in Jordan were evaluated by means of solid phase microextraction-gas chromatography/mass spectrometry (SPME-GC/MS) technique during the vegetative and flowering seasons of the plant. Additionally, the chemical composition of the hydrodistilled essential oil obtained from different I. visocsa organs was also investigated. However, principle component analysis (PCA) and cluster analysis (CA) were performed to all data to investigate similarities and differences in the chemical composition of the emission profiles and essential oils as affected by growth stage, geographical zone and organ. To the best of our knowledge, the characterization VOCs obtained from fresh aerial parts of I. viscosa plant during the ontogenetic evolution has never been mentioned before.

EXPERIMENTAL

In the present investigation, equal amounts of the aerial parts of *I. viscosa* were collected during the vegetative stage (April-August) and full flowering stage (October) from the two different locations belonging to the MID zone (Al-Salt governorate: (BAU/COMP-35-2015S), Northern Jordan Valley: (BAU/COMP-35-2015NJV), and from the IT zone (Area surrounding Al-Karak governorate (BAU/COMP-35-2015K). The plant material was identified by Prof. Dr Hala I. Al-Jaber, Department of Chemistry, Al-Balqa Applied University, Al-Salt, Jordan. Voucher specimens were deposited in the Department of Chemistry, Faculty of Science, Al-Balqa Applied University, Al-Salt, Jordan.

Solid-phase-micro-extraction (SPME) and hydrodistillation of essential oils: Both experiments were performed under the same conditions described earlier [7,9,10]. α and β-Pinenes, *p*-cymene, 1,8-cineol, α-terpineol, limonene, ocimene, linalool, sabinene, thujone, borneol, humulene, alloarmadendrene, caryophyllene, caryophyllene oxide and E-nerolidol were purchased from Sigma-Aldrich (Buchs, Switzerland). GC-grade hexane and analytical reagent grade anhydrous Na₂SO₄ were purchased from Scharlau (Barcelona, Spain) and UCB (Bruxelles, Belgium), respectively.

GC-MS and GC-FID analysis: Both experiments were performed according to the procedure described in the literature [9,10-14] and using the same instruments and chromatographic conditions. Identification of compounds was achieved based on the built in libraries (NIST and Wiley, USA) and through comparing the calculated retention indices (RI) of the identified compounds with their literature values measured relative to same standard under same chromatographic conditions [15] or by co-injection of authentic samples. The relative peak areas were measured and then used to calculate the concentration of the detected compounds.

Statistical analysis: Statistical analysis was performed on R 3.6.1 (The R Foundation of Statistical Computing) with devtools, ggbiplot, psych and GPA rotation packages were used for the data processing and chemometric analysis.

RESULTS AND DISCUSSION

Emission profile during the vegetative period: Using static-SPME followed by GC/MS analysis, investigation of the chemical composition of the spontaneously emitted aroma from the aerial parts of the plant during vegetative period led to the identification of 61 compounds from the three locations. The identified compounds and their % concentrations are listed in Table-1.

The plant collected from Al-Salt area (MID-1) showed an emission profile rich in sesquiterpene hydrocarbons (64.72%).

SEASONAL AND GEOGRAPHICAL VARIATION OF THE CHEMICAL COMPOSITION OF VOCs EMITTED FROM THHE AERIAL PARTS OF <i>I. viscosa</i> GRWOING WILD IN JORDAN													
	R	I ^a			MID-1			MID-2		IT			
No.	Lit.	Exp.	Constituents	April (%)	June (%)	August (%)	April (%)	June (%)	August (%)	April (%)	June (%)	August (%)	b
1	939	935	α-Pinene	-	2.37	2.17	3.26	-	4.38	6.23	2.87	0.87	1, 2, 3
2	924	932	3-Methyl-4-heptanone	0.97	-	-	-	-	-	-	-	1.89	1,2
3	985	980	trans-Isolimonene	-	-	-	1.00	1.04	-	0.62	0.59	-	1,2
4	979	979	β-Pinene	0.70	9.54	1.78	12.39	12.24	-	9.80	2.27	3.66	1,2, 3
5	991	990	Dehydro-1,8-cineol	-	-	-	-	-	-	-	5.34	2.89	1,2
6	975	966	Sabinene	1.96	0.18	1.48	-	-	-	0.46	0.32	-	1,2, 3
7	1005	1005	cis-3-Hexenyl acetate	-	1.37	6.14	2.57	1.55	3.16	-	3.15	-	1,2
8	1025	1024	<i>p</i> -Cymene	4.56	-	-	-	-	-	0.24	19.45	-	1,2, 3
9	1029	1029	Limonene	3.86	9.80	1.31	13.17	9.24	3.86	32.20	-	-	1,2, 3
10	1031	1032	1,8-Cineol	5.16	-	-	-	4.10	-	-	-	-	1,2, 3
11	1050	1045	trans-Ocimene	-	-	-	1.03	1.71	0.68	0.24	-	-	1,2, 3
12	1089	1094	Terpinolene	-	-	3.53	-	-	-	0.53	4.91	-	1, 2
13	1091	1093	Methylbenzoate	-	1.89	-	-	3.21	-	-	-	-	1, 2
14	1097	1100	Linalool	-	0.53	2.19	-	-	-	0.52	-	-	1, 2, 3
15	1101	1105	<i>n</i> -Nonanal	-	-	-	0.82	0.82	0.82	0.51	0.82	0.82	1, 2
16	1123	1113	Myrceneol	6.39	11.02	28.51	19.12	4.68	-	14.07	38.15	13.98	1, 2
17	1169	1171	Borneol	-	-	1.23	-	-	-	-	-	-	1, 2
18	1192	1191	Methyl salicylate	1.47	1.26	-	-	6.38	-	0.32	1.05	1.23	1, 2
19	1189	1196	α-Terpineol	-	-	-	-	-	-	0.58	0.70	-	1, 2, 3
20	1200	1200	Dodecane	0.99	-	-	-	-	-	-	-	-	1, 2

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21	1202	1206	n-Decanal	0.66	-	-	-	-	-	0.38	-	-	1, 2
22	1221	1219	cis-Sabinene hydrate acetate	-	-	-	1.69	-	-	-	-	-	1, 2
23	1235	1228	Thymol methyl ether	2.52	-	2.00	-	-	-	-	-	-	1, 2
24	1244	1235	2-E-Hexenyl isovalerate	-	0.36	2.80	-	-	-	-	-	-	1, 2
25	1264	1260	2-E-Decenal	-	-	1.15	-	-	-	-	-	-	1, 2
26	1319	1302	cis-Cycloisolongifol-5-ol	1.89	-	-	-	-	-	-	0.34	-	1, 2
27	1319	1320	3-Z-Hexenyl tiglate	-	0.72	3.50	-	-	-	-	-	-	1, 2
28	1351	1366	α-Cubebene	2.52	1.84	0.12	-	0.46	3.32	-	-	-	1, 2
29	1377	1373	α-Copaene	25.31	16.85	7.91	3.10	7.34	23.50	3.49	-	-	1, 2
30	1391	1386	7-Episesquithujene	-	0.37	-	1.15	1.92	3.34	0.31	-	-	1,2
31	1391	1391	β-Elemene	-	0.37	3.74	-	-	0.71	15.37	0.99	-	1,2
32	1401	1399	B-Longifolene	-	-	_	_	1.06	-	-	-	3.18	1.2
33	1409	1412	<i>cis</i> -Carvonhyllene	-	-	1.96	-	_	_	-	0.44	1.25	1.2.3
34	1419	1418	trans-Carvophyllene	14.83	11.04	1.55	13.28	8.81	7.62	-	1.16	4.30	1, 2, 3
35	1435	1431	trans_0_Bergamotene	-	-	-	-	-	1.57	-	-	-	1.2
36	1436	1446	Nervl acetone	0.67	_	_	_	_	1.13	_	0.45	1.05	1,2
37	1450	1/152	B E Estraçono	1.03	_	_	_	_	1.15	1.26	-	1.05	1,2
38	1455	1450	p-E-Pariesene	1.75	1 57		1.84	1 30	3 73	0.31	0.48		1,2
20	1455	1457	Alle annuellene	-	2.41	-	2.15	2.25	6.01	0.31	0.40	-	1, 2, 3
39	1400	1457	Alloarmadendrene	4.81	3.41	1.22	2.15	2.25	0.81	0.48	-	-	1, 2, 3
40	1482	1459	γ-Irone	-	-	1.26	-	-	-	-	0.07	-	1, 2
41	1477	1471	γ-Gurjunene	-	0.76	3.19	1.61	1.08	-	0.67	0.32	2.64	1, 2
42	1496	1474	Valencene	-	-	-	-	-	-	-	-	0.51	1, 2
43	1480	1472	γ-Muurolene	0.24	-	-	-	1.91	-	0.58	0.99	3.35	1,2
44	1483	1476	γ-Curcumene	-	-	-	-	1.98	4.84	-	-	-	1, 2
45	1481	1479	ar-Curcumene	-	1.71	-	-	1.65	2.60	-	-	-	1, 2
46	1490	1486	β-Selinene	0.92	1.80	3.98	2.24	5.30	7.29	0.98	0.71	5.62	1, 2
47	1493	1493	δ-Selinene	2.91	5.33	3.81	6.72	4.81	5.22	0.97	2.09	14.96	1, 2
48	1500	1496	α-Muurolene	3.14	1.13	-	-	3.06	3.07	-	-	-	1, 2
49	1506	1502	E.E-α-Farnesene	-	0.80	-	-	0.97	-	0.53	-	0.84	1,2
50	1506	1506	B-Bisabolene	_	-	-	_	0.91	0.72	-	-	_	1.2
51	1516	1507	B Curcumene	_	_	_	_	-	0.69	_	_	_	1 2
52	1514	1515	y Cadinana	8 11	1 70	_	_	177	1.84	0.97	_	_	1 2
52	1550	1519	p-Caulifelie Dimathyl azalata	0.11	1.79		_	4.77	7.07	0.77	1 21		1,2
53	1550	1562	E Nerolidol	-	- 8.40	-	2 24	-	-	-	5.16	27.04	1, 2 1 2 3
55	1564	1566	ani Longininanol	-	0.40	10.71	2.24	-	-	4.00	5.10	27.94	1, 2, 3
56	1560	1560	Longipinanol	-	0.63	-	-	-	-	0.07	-	-	1, 2
57	1583	1578	Carvonhyllene oxide	0.77	1.34	1 / 2		0.87	3.28	1.56	0.45	2 32	1,2
58	1601	1580	Cedrol	0.77	1.54	1.42	_	0.87	5.20	1.50	5.25	2.32	1, 2, 3
50	1682	1683	Ishwarone	-	0.30	-	-	-	-	-	5.25	2 /8	1,2
60	1724	1724	Methyl tetradecanoate	2 11	0.57							2.40	1, 2 1 2
61	1807	1700	2 Ethylbevyl salicylate	2.11	1 / 1	-	-	-	-	-	0.26	288	1,2
Mon	1007	hydroca	2-Eurymexyr sancyrau rhons (1, 3, 4, 6, 9, 11, 12)	6.52	21.90	10.27	30.85	24.23	802	50.08	10.20	4.53	1, 2
Ovv	serpene	nyuloca	(1, 3, 4, 0, 3, 11, 12)	12 22	11 55	31.03	20.81	24.23 8.78	1.13	15 17	10.90	17.02	
Seco	uiternene	hydrocy	(10, 10, 14, 10, 17, 19, 22, 50)	64 72	11.55	27.48	20.01	0.70 17 03	76 77	25.02	7 1 8	36.65	
Ovv	renated s	esquiter	$\frac{100118}{28-35}, \frac{57-59}{57-59}, \frac{41-44}{40-52}$	2 66	10.76	12 13	2 24	0.87	3.28	6.80	11 25	30.05	
Aliph	patic hyd	rocarbor	& their derivatives	4 73	2 45	13.59	3 39	2 37	3.98	0.89	5 18	2 71	
(2 7)	15.20 [′]	21.24.2	5 27 53 60)	4.75	2.45	15.57	5.57	2.57	5.70	0.07	5.10	2.71	
Phen	vl propa	noids (8)	13 18 23 45 61)	8.55	4.38	2.00	-	8.03	2.6	0.56	20.76	4.11	
Total	identifie	ed (%)	10, 10, 20, 10, 01)	99.4	98.09	97.4	89.38	92.21	96.68	99.51	99.92	98.66	
1014				· · · ·	10.07	27.1	07.00	2.21	20.00	77.01	,,,,,	20.00	

^aLinear retention index on a DB-5 column; ^bIdentification method: 1 = linear retention index; 2 = identification based on comparison of mass spectra; 3 = Co-injection with standard compounds. Serial numbers of components detected in each class are listed beside the Class name.

The main compound detected in this fraction was α -copaene (25.31%). The content of sesquiterpene hydrocarbons decreased gradually in the emitted aroma of the plant as a function of growth stage, reaching a minimum value of 27.48% prior to the flowering season. The decrease in the concentration of the main class was accompanied also by a decrease in the concentration of α -copaene, which accounted for 7.91% of the total emitted aroma content during this period.

Sesquiterpene hydrocarbons also dominated the emission profile of the aerial parts of *I. viscosa* collected from northern Jordan valley region (MID-2) and their contents increased gradually with maturation degree (32.09 to 76.77%). During

the early vegetative stage, *trans*-caryophyllene was the main component (13.28%) and its content decreased gradually to 7.62% prior to flowering period. On the contrary, the content of α -copaene increased gradually to 23.50% of the total composition during the same period.

The aerial parts of *I. viscosa* collected from IT zone had a variably interesting emission profile that was rich mainly in monoterpene hydrocarbons in the early vegetative stage (50.08%), oxygenated monoterpenes in early summer season (44.74%) and sesquiterpene hydrocarbons in late summer (36.65%). During this period of time, these three classes were dominated by limonene (32.20%), myrcenol (38.15%) and

 δ -selinene (14.96%), respectively. Interestingly, the aroma of the plant collected from this location contained higher amounts of the oxygenated sesquiterpene E-nerolidol during the summer season, which amounted to 27.94% of the total content.

Volatile organic compounds (VOCs) during the flowering period: The chemical composition of the spontaneously emitted VOCs of *I. viscosa* aerial organs including fresh leaves (L), pre-flowering buds (PF) and full inflorescences (F) were also determined during the full flowering period (October). Results are summarized in Table-2.

Sesquiterpenoids were the main class detected in the fresh leaves samples collected from the various locations, but with

TABLE-2 VARIATION OF THE SPME-VOCs CONTENT OF LEAVES (L), PRE-FLOWERING (PF) BUDS AND FULLY EXPANDED FLOWERS (F) DURING THE FLOWERING SEASON													
	R	Ia			MID-1			MID-2			IT		_
No.	T :+	Eve	Constituents	L	PF	F	L	PF	F	L	PF	F	b
	Lit.	Exp.		(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	
1	927	927	Tricyclene	-	0.67	0.82	-	-	-	-	0.92	-	1, 2
2	930	930	α-Thujene	-	-	-	-	-	-	-	-	2.55	1, 2
3	924	932	3-Methyl-4-heptanone	-	-	-	-	-	-	-	0.61	-	1, 2
4	939	933	α-Pinene	0.89	0.40	0.70	1.15	1.60	2.00	-	1.47	2.76	1, 2, 3
5	967	967	n-Heptanol	-	0.44	0.71	-	1.44	0.58	-	-	0.69	1, 2
6	975	980	Sabinene	1.17	2.34	2.38	0.56	-	1.13	-	-	-	1, 2, 3
7	986	980	6-Methyl-5-hepten-2-one	-	-	-	-	-	-	-	0.63	-	1, 2
8	985	984	trans-Isolimonene	-	-	-	-	1.08	0.44	-	-	-	1,2
9	979	986	β-Pinene	0.67	1.35	1.36	0.93	1.75	17.96	2.96	-	0.87	1, 2, 3
10	988	990	Dehydro-1,8-cineol	2.12	4.07	4.12	5.92	5.46	-	-	-	17.34	1,2
11	999	1003	<i>n</i> -Octanal	-	-	-	-	-	-	-	0.60	-	1,2
12	1005	1005	cis-3-Hexenyl acetate	1.62	-	-	1.53	-	1.73	-	-	-	1,2
13	1017	1010	α-Terpinene	0.18	-	-	-	-	-	-	-	-	1, 2, 3
14	1025	1024	<i>p</i> -Cymene	1.94	-	-	-	-	-	- 2.26	-	-	1, 2, 3
15	1029	1029	Limonene	1.98	1.09	1.21	11.11	-	3.17	5.50	-	0.89	1, 2, 3
10	1031	1032	cis Ocimene	0.71	0.45	0.82	-	0.12	1.20	-	0.82	1.20	1, 2, 3 1 2 3
17	1057	1035	trans_Ocimene	-	0.80		-	0.0	1.21	-	0.00	0.97	1, 2, 3 1 2 3
19	1050	1045	v Terninene	0.81	2.96	2.96	_	-	-	_	-	-	1, 2, 3
20	1068	1064	<i>n</i> -Octanol	-	2.90	2.90	_	_	_	_	1.06	_	1,2
21	1089	1004	Terminolene	0.98	1 38	1 46	_	_	0.80	_	2.89	2 91	1 2 3
22	1091	1090	Dehydrolinalool	-	-	-	_	-	1.09	_	-	-	1, 2, 3
23	1097	1099	Linalool	-	0.47	-	_	1.22	0.57	3.40	0.71	-	1, 2, 3
24	1102	1107	α-Thuione	0.64	_	-	-	_	-	-	_	-	1, 2, 3
25	1101	1105	<i>n</i> -Nonanal	-	-	0.53	0.56	-	-	-	0.72	-	1, 2
26	1123	1113	Myrcenol	17.29	37.07	36.74	0.61	20.92	30.36	26.03	39.09	33.88	1, 2
27	1192	1193	Methyl salicylate	-	-	-	1.14	-	-	-	-	-	1, 2
28	1189	1196	α-Terpineol	-	0.34	-	-	-	-	0.60	0.64	0.97	1, 2, 3
29	1202	1206	n-Decanal	0.39	0.42	-	-	-	-	0.74	0.84	-	1, 2
30	1197	1215	Butanoicacid, 2-methyl-4-	-	-	-	-	-	-	-	1.00	-	1, 2
			methylpentyl										
31	1229	1228	3-Z-Hexenyl 2-methyl butanoate	18.05	-	-	-	-	-	-	-	-	1,2
32	1235	1228	Thymol methyl ether	-	-	-	-	-	-	0.97	-	-	1,2
33	1232	1235	3-Z-Hexenyl 3-methyl butanoate	0.76	-	-	-	-	-	-	-	-	1,2
34 25	1290	1295	inymoi	0.44	-	-	-	-	-	-	-	-	1, 2, 3
35	1319	1302	Secamol	- 0.45	0.05	0.32	-	-	-	-	-	-	1, 2 1 2
30	1312	1312	Hexyl tiglate	1.32	_		_	_	-	-		-	1, 2 1 2
38	1351	1366	α -Cubebene	1.02	1 76	1 73	1.67	1 72	1 20	0.59	_	_	1, 2
39	1377	1373	a-Consene	7 29	13.04	12.89	15 44	11.72	9.13	-	_	_	1,2
40	1391	1386	7-Enisesquithuiene	-	-	-	2.94	-	-	_		_	1,2
41	1388	1391	B-Cubebene	-	1.67	1.66	-	-	-	-	-	_	1, 2
42	1391	1391	B-Elemene	4.21	1.08	1.07	-	1.35	1.38	1.10	-	_	1,2
/3	1/01	1302	B Longininono		-	-	_	-	-	8.94	1.96	1.02	1,2
43	1402	1403	jso-Italicene	_	_	_	2 55	_	_	0.54	1.50	1.92	1,2
45	1402	1412	<i>cis</i> -Carvophyllene	240	0.91	0.90	10.70	1.03	_	2.67	1 16	0.78	1, 2 1 2 3
46	1419	1418	trans-Carvophyllene	0.71	2.78	2,76	-	4.93	9.96	4.88	11.18	7.15	1, 2, 3
47	1435	1431	trans-\alpha-Bergamotene	-	-		1.62	-	0.41	-	-	-	1, 2, 3
48	1436	1446	Nervl acetone	_	0.63	0.63	0.42	1.29	_	2.22	_	_	1.2
49	1441	1440	Aromadendrene	0.70	0.83	0.82	-	-	-	-	-	-	1, 2
50	1454	1445	α-Neoclovene	-	-	-	-	-	-	-	-	0.81	1, 2
51	1457	1452	β-E-Farnesene	-	-	-	-	-	-	1.43	1.60	-	1, 2
52	1455	1453	α-Humulene	1.04	-	-	1.11	3.06	0.90	_	-	0.97	1, 2, 3
53	1460	1457	Alloarmadendrene	1.61	2.70	2.68	2.08	4.48	1.93	-	-	-	1, 2,3

Vol. 32, No. 10 (2020	 Effect of Geographic I 	Location, Ontogenesis of	on Essential Oil Composition of	I. viscosa (L.) Greuter.	(Astraceae) 2563
,	, U I	, 0	1		

$ \begin{array}{cccccccccccccccccccccccccccccccccccc$														
55 1482 1462 γ -Frome 0.97 - - - - 7.45 2.65 - 1.2 56 1477 1471 γ -Gurjunene 1.27 1.40 1.39 0.63 - - 0.89 - 1.71 1.2 58 1496 1471 Valurolene - 2.05 - - 2.20 1.33 7.87 3.22 2.67 1.2 59 1483 1476 γ -Curcumene - - 13.55 - - - - 1.2 60 1481 1479 ar-Curcumene - - - 13.55 - - - - 1.2 61 1490 1486 β -Selinene 3.20 2.87 2.84 2.36 11.32 3.54 3.89 3.70 3.46 1.2 62 1493 1493 & Selinene 5.12 4.22 4.18 1.31 1.89 0.84 5.85 10.10 7.71 1.2 63 1500 1	54	1463	1461	Dehydroaromadendrane	-	0.80	0.80	0.40	1.04	-	-	-	-	1, 2
56 1477 1471 -FGurjunene 1.27 1.40 1.39 - 1.48 - 1.89 2.77 1.72 1.2 57 1480 1471 Y-Muurolene 2.34 - 2.03 0.63 - - 0.89 - 1.71 1.2 58 1496 1471 Valencene - 2.05 - - 2.00 1.33 7.87 3.22 2.67 1.2 59 1483 1476 y-Curcumene - - - 8.05 - - - - 1.27 60 1481 1479 ar-Curcumene - - - 8.05 - - - 1.2 61 1490 1486 β-Selinene 3.20 2.87 2.84 1.31 1.89 0.44 5.55 - - 1.2 63 1500 1496 o-Muurolene 0.71 0.92 0.91 1.28 3.39 2.05 0.55 - - 1.2 64 1516	55	1482	1462	γ-Irone	0.97	-	-	-	-	-	7.45	2.65	-	1, 2
57 1480 1472 r-Muurolene 2.34 - 2.03 0.63 - - 0.89 - 1.71 1.2 58 1496 1471 Valencene - 2.05 - - 2.20 1.33 7.87 3.22 2.67 1.2 59 1483 1476 r-Curcumene - - - 8.05 - - - 1.2 61 1490 1486 β-Selinene 3.20 2.87 2.84 2.36 11.32 3.54 3.89 3.70 3.46 1.2 62 1493 1496 α -Muurolene 0.71 0.92 0.91 1.28 3.39 2.05 0.55 - - 1.2 63 1500 1496 α -Muurolene 0.67 1.11 1.10 3.00 2.00 1.00 - - 1.2 65 1514 1516 r-Cardinene 0.67 0.67 - - 0.37 - 1.2 66 1535 1536 10-epi-Longip	56	1477	1471	γ-Gurjunene	1.27	1.40	1.39	-	1.48	-	1.89	2.77	1.72	1, 2
58 1496 1471 Valencene - 2.05 - - 2.20 1.33 7.87 3.22 2.67 1, 2 59 1483 1476 γ Curcumene - - - - - - - - 1, 2 60 1481 1479 ar-Curcumene 3.20 2.87 2.84 2.36 11.32 3.54 3.89 3.70 3.46 1, 2 61 1490 1486 β-Selinene 3.20 2.87 2.84 2.36 11.32 3.59 3.69 3.70 3.46 1, 2 62 1493 1496 α -Murolene 0.71 0.92 0.91 1.28 3.39 2.05 0.55 - - 1, 2 63 1500 1496 α -Curcumene - - 2.31 - - - - 1, 2 64 1516 1507 β -Curcumene 0.67 1.11 1.10 3.00 2.00 1.00 - - 1, 2 65 1514<	57	1480	1472	γ-Muurolene	2.34	-	2.03	0.63	-	-	0.89	-	1.71	1, 2
59 1483 1476 γ -Curcumene - - 13.55 - - - - 1, 2 60 1481 1479 ar-Curcumene - - 8.05 - - - - 1, 2 61 1490 1486 β -Selinene 3.20 2.87 2.84 2.36 11.32 3.59 3.89 3.70 3.46 1.2 62 1493 1496 α -Sumene 5.12 4.22 4.18 1.31 1.89 0.84 5.85 10.10 7.71 1,2 63 1500 1496 α -Muurolene 0.71 0.92 0.91 1.28 3.39 2.05 0.55 - - 1,2 64 1516 1507 β -Curcumene - - 2.31 - - - 1,2 65 1514 1516 requiveloption 0.67 0.67 - - - 1,2 3 36 1,2 3 55 7.45 3.67 1,2,3 3 68 <t< td=""><td>58</td><td>1496</td><td>1471</td><td>Valencene</td><td>-</td><td>2.05</td><td>-</td><td>-</td><td>2.20</td><td>1.33</td><td>7.87</td><td>3.22</td><td>2.67</td><td>1, 2</td></t<>	58	1496	1471	Valencene	-	2.05	-	-	2.20	1.33	7.87	3.22	2.67	1, 2
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	59	1483	1476	γ-Curcumene	-	-	-	13.55	-	-	-	-	-	1, 2
6114901486β-Selinene3.202.872.842.3611.323.543.893.703.461, 26214931493δ-Selinene5.124.224.181.311.890.845.8510.107.711, 26315001496 α -Muurolene0.710.920.911.283.392.050.551, 26415161507β-Curcumene2.311, 26515141516y-Cadinene0.671.111.103.002.001.001, 2661535153610-epi-Cubebol-0.670.671, 26715631562E-Nerolidol9.822.852.82-12.091.385.557.453.671, 2, 36815641566epi-Longipinanol0.500.530.870.562.381.200.781, 2, 37016411631Caryophylla-4(14).8(15)-dien-5- β-ol0.491, 27316821678Intermedel0.491, 27316821678Isbwarone0.510.33-1.273 <t< td=""><td>60</td><td>1481</td><td>1479</td><td>ar-Curcumene</td><td>-</td><td>-</td><td>-</td><td>8.05</td><td>-</td><td>-</td><td>-</td><td>-</td><td>-</td><td>1, 2</td></t<>	60	1481	1479	ar-Curcumene	-	-	-	8.05	-	-	-	-	-	1, 2
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	61	1490	1486	β-Selinene	3.20	2.87	2.84	2.36	11.32	3.54	3.89	3.70	3.46	1, 2
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	62	1493	1493	δ-Selinene	5.12	4.22	4.18	1.31	1.89	0.84	5.85	10.10	7.71	1, 2
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	63	1500	1496	α-Muurolene	0.71	0.92	0.91	1.28	3.39	2.05	0.55	-	-	1, 2
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	64	1516	1507	β-Curcumene	-	-	-	2.31	-	-	-	-	-	1, 2
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	65	1514	1516	γ-Cadinene	0.67	1.11	1.10	3.00	2.00	1.00	-	-	-	1,2
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	66	1535	1536	10-epi-Cubebol	-	0.67	0.67	-	-	-	-	-	-	1, 2
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	67	1563	1562	E-Nerolidol	9.82	2.85	2.82	-	12.09	1.38	5.55	7.45	3.67	1, 2, 3
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	68	1564	1566	epi-Longipinanol	0.50	0.53	-	-	-	-	0.37	-	-	1, 2
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	69	1583	1578	Caryophyllene oxide	1.35	-	0.53	-	0.87	0.56	2.38	1.20	0.78	1, 2, 3
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	70	1641	1631	Caryophylla-4(14),8(15)-dien-5-	-	-	-	0.49	-	-	-	-	-	1, 2
$\begin{array}{cccccccccccccccccccccccccccccccccccc$				β-ol										
7216671662Intermedeol0.451, 27316821678Ishwarone0.510.381, 2Monoterpene hydrocarbons (1, 2, 4, 6, 8, 9, 13, 15, 17, (8, 19, 21)6.6811.4810.8913.755.3328.346.326.2710.95Oxygenated monoterpenes (10, 16, 22, 23, 24, 26, 28, 48)20.7643.0142.316.9529.0133.2832.2541.2653.39Sesquiterpene hydrocarbon (38-47, 49-54, 56-65)32.3639.4739.1062.9551.1833.6741.2335.6928.90Oxygenated sesquiterpenes (55, 66-73)14.154.684.540.4912.961.9417.611.34.45Aliphatic hydrocarbons & their derivatives (3, 5, 7, 11, 22, 25, 29, 30, 31, 33, 37)2.83-9.19-0.97Phenyl propanoids (14, 32, 34, 36)2.839.19-0.97Total identified98.9299.5098.0895.4299.9299.5499.1199.9898.38	71	1648	1652	Agarospirol	1.0	-	-	-	-	-	1.02	-	-	
73 1682 1678 Ishwarone 0.51 - - - 0.38 - - 1, 2 Monoterpene hydrocarbons (1, 2, 4, 6, 8, 9, 13, 15, 17, 18, 19, 21) 6.68 11.48 10.89 13.75 5.33 28.34 6.32 6.27 10.95 Oxygenated monoterpenes (10, 16, 22, 23, 24, 26, 28, 48) 20.76 43.01 42.31 6.95 29.01 33.28 32.25 41.26 53.39 Sesquiterpene hydrocarbon (38-47, 49-54, 56-65) 32.36 39.47 39.10 62.95 51.18 33.67 41.23 35.69 28.90 Oxygenated sesquiterpenes (55, 66-73) 14.15 4.68 4.54 0.49 12.96 1.94 17.6 11.3 4.45 Aliphatic hydrocarbons & their derivatives (3, 5, 7, 11, 22.14 0.86 1.24 2.09 1.44 2.31 0.74 5.46 0.69 12, 20, 25, 29, 30, 31, 33, 37) Phenyl propanoids (14, 32, 34, 36) 2.83 - 9.19 - 0.97 - - Total identified 98.92 99.50 98.08 95.42 99.92 99.54 99.11 <	72	1667	1662	Intermedeol	-	-	-	-	-	-	0.45	-	-	1, 2
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	73	1682	1678	Ishwarone	0.51	-	-	-	-	-	0.38	-	-	1, 2
18, 19, 21)Oxygenated monoterpenes $(10, 16, 22, 23, 24, 26, 28, 48)$ 20.7643.0142.316.9529.0133.2832.2541.2653.39Sesquiterpene hydrocarbon $(38-47, 49-54, 56-65)$ 32.3639.4739.1062.9551.1833.6741.2335.6928.90Oxygenated sesquiterpenes $(55, 66-73)$ 14.154.684.540.4912.961.9417.611.34.45Aliphatic hydrocarbons & their derivatives $(3, 5, 7, 11, 22.14)$ 0.861.242.091.442.310.745.460.6912, 20, 25, 29, 30, 31, 33, 37)Phenyl propanoids $(14, 32, 34, 36)$ 2.839.190.97Total identified98.9299.5098.0895.4299.9299.5499.1199.9898.38	Mone	oterpene	hydroca	rbons (1, 2, 4, 6, 8, 9, 13, 15, 17,	6.68	11.48	10.89	13.75	5.33	28.34	6.32	6.27	10.95	
Oxygenated monoterpenes (10, 16, 22, 23, 24, 26, 28, 48) 20.76 43.01 42.31 6.95 29.01 33.28 32.25 41.26 53.39 Sesquiterpene hydrocarbon (38-47, 49-54, 56-65) 32.36 39.47 39.10 62.95 51.18 33.67 41.23 35.69 28.90 Oxygenated sesquiterpenes (55, 66-73) 14.15 4.68 4.54 0.49 12.96 1.94 17.6 11.3 4.45 Aliphatic hydrocarbons & their derivatives (3, 5, 7, 11, 22.14 0.86 1.24 2.09 1.44 2.31 0.74 5.46 0.69 12, 20, 25, 29, 30, 31, 33, 37) Phenyl propanoids (14, 32, 34, 36) 2.83 - 9.19 - 0.97 - - Total identified 98.92 99.50 98.08 95.42 99.92 99.54 99.11 99.98 98.38	18, 1	9, 21)												
Sesquiterpene hydrocarbon (38-47, 49-54, 56-65) 32.36 39.47 39.10 62.95 51.18 33.67 41.23 35.69 28.90 Oxygenated sesquiterpenes (55, 66-73) 14.15 4.68 4.54 0.49 12.96 1.94 17.6 11.3 4.45 Aliphatic hydrocarbons & their derivatives (3, 5, 7, 11, 22.14 0.86 1.24 2.09 1.44 2.31 0.74 5.46 0.69 12, 20, 25, 29, 30, 31, 33, 37) 2.83 - - 9.19 - 0.97 - - Total identified 98.92 99.50 98.08 95.42 99.92 99.54 99.11 99.98 98.38	Oxyg	enated n	nonoterp	benes (10, 16, 22, 23, 24, 26, 28, 48)	20.76	43.01	42.31	6.95	29.01	33.28	32.25	41.26	53.39	
Oxygenated sesquiterpenes (55, 66-73) 14.15 4.68 4.54 0.49 12.96 1.94 17.6 11.3 4.45 Aliphatic hydrocarbons & their derivatives (3, 5, 7, 11, 22.14 0.86 1.24 2.09 1.44 2.31 0.74 5.46 0.69 12, 20, 25, 29, 30, 31, 33, 37) 2.83 - - 9.19 - 0.97 - - Total identified 98.92 99.50 98.08 95.42 99.92 99.54 99.11 99.98 98.38	Sesqu	iterpene	e hydroca	arbon (38-47, 49-54, 56-65)	32.36	39.47	39.10	62.95	51.18	33.67	41.23	35.69	28.90	
Aliphatic hydrocarbons & their derivatives (3, 5, 7, 11, 22.14 0.86 1.24 2.09 1.44 2.31 0.74 5.46 0.69 12, 20, 25, 29, 30, 31, 33, 37) 2.83 - - 9.19 - - 0.97 - - Phenyl propanoids (14, 32, 34, 36) 2.83 - - 9.19 - - 0.97 - - Total identified 98.92 99.50 98.08 95.42 99.92 99.54 99.11 99.98 98.38	Oxyg	enated s	esquiter	penes (55, 66-73)	14.15	4.68	4.54	0.49	12.96	1.94	17.6	11.3	4.45	
12, 20, 25, 29, 30, 31, 33, 37) Phenyl propanoids (14, 32, 34, 36) 2.83 - 9.19 - 0.97 - - Total identified 98.92 99.50 98.08 95.42 99.92 99.54 99.11 99.98 98.38	Aliph	natic hyd	rocarbor	ns & their derivatives (3, 5, 7, 11,	22.14	0.86	1.24	2.09	1.44	2.31	0.74	5.46	0.69	
Phenyl propanoids (14, 32, 34, 36) 2.83 - 9.19 - 0.97 - - Total identified 98.92 99.50 98.08 95.42 99.92 99.54 99.11 99.98 98.38	12, 20, 25, 29, 30, 31, 33, 37)													
Total identified 98.92 99.50 98.08 95.42 99.92 99.11 99.98 98.38	Phenyl propanoids (14, 32, 34, 36)			2.83	-	-	9.19	-	-	0.97	-	-		
	Total	identifie	ed		98.92	99.50	98.08	95.42	99.92	99.54	99.11	99.98	98.38	

^aLinear retention index on a DB-5 column; ^bIdentification method: 1 = linear retention index; 2 = identification based on comparison of mass spectra; 3 = Co-injection with standard compounds. Serial numbers of components detected in each class are listed beside the Class name.

some qualitative and quantitative differences regarding the types of sesquiterpenoids (hydrocarbons or oxygenated).

While sesquiterpene hydrocarbons dominated the emission profile of the fresh leaves collected from waste lands of MID-1 (32.36%), IT (41.23%) and MID-2 (62.95%), other classes afforded the most abundant compounds detected in the emission profiles of *I. viscosa* leaves. These included the aliphatic hydrocarbon 3-*Z*-hexenyl-2-methyl butanoate (18.05%, MID-1) and the oxygenated monoterpene myrcenol (26.03%, IT zone). α -Copaene (15.44%) was the main sesquiterpene hydrocarbon detected in the emitted aroma of leaves samples collected from MID-2 region.

The emission profiles of the flowers at the pre-flowering stage revealed a total of 39, 26 and 26 constituents detected in the spontaneously emitted VOCs from the three locations, and accounting for 98.92% (MID-1), 99.92% (MID-2), 99.98% (IT) of the total emission content. Oxygenated monoterpenes dominated the emission profiles of the PF buds of MID-1 and IT samples (43.01% and 41.26%, respectively) mostly represented by myrcenol (37.07% and 39.09%, respectively). On the other hand, sesquiterepene hydrocarbons dominated the aroma of the PF buds collected from MID-2 region (51.18%) followed by oxygenated monoterpenoids (29.01%), with myrcenol (20.92%) as the main component. The main sesquiterpene hydrocarbons detected included β -selinene (11.32%) and α -copaene (11.29%).

Oxygenated monoterpenes dominated the emission profile of the inflorescences during the full flowering stage collected in IT (53.39%) and MID-2 (33.28%) samples. Samples from MID-1 area were rich in sesquiterpene hydrocarbons (39.10%). Despite this variation, myrcenol remained the main component in the emissions of all samples from the three different locations (MID-1: 36.74%; MID-2: 30.36%, and IT: 33.88%).

Chemical composition of the hydrodistilled oils obtained from I. viscosa fresh leaves and flowers at the pre- and full flowering stages: Analysis of the hydrodistilled oils obtained from the leaves and the inflorescences during the pre-flowering and full flowering stages resulted in the identification and characterization of a total of 48 compounds (Table-3). All samples from the two geographical zones were characterized by high content of oxygenated sesquiterpenoids amounting to more than 67% of the total content. In most cases, E-nerolidol had the highest contribution to the distilled oils obtained from leaves, pre-flowering buds and fully flowering blooms of MID-1(50.2%, 70.4% and 54.7%, respectively), MID-2 (88.7%, 40.1 and 41.2%, respectively) and IT (37.7%, 44.4% and 47.1%). In addition, other compounds belonging to this class were detected in appreciable amounts. Table-3 lists the main constituents detected in the different hydrodistilled organs.

Principal component analysis (PCA) and cluster analysis (CA): In the current work, PCA has been applied to the data obtained from the GC/MS analysis (Tables 1-3) and was then used to investigate similarities or differences in the chemical composition of emission profiles of the aerial parts and organs as affected by growth stage and origin (MID-1, MID-2 and IT). Therefore, only the first few PCs were studied, best result was obtained upon using a two-dimensional PCAs score models that for about 40% (Table-1), 42% (Table-2) and 21% (Table-3) of the total variation in the data sets using the first two PCs.

TABLE-3 CHEMICAL COMPOSITION OF THE HYDRO-DISTILLED OILS OBTAINED FROM THE LEAVES (L), PRE-FLOWERING BUDS (PF) AND FULLY EXPANDED FLOWERS (F) OF *I. viscosa* GRWOING WILD IN JORDAN

	R	Ia		MID-1				MID-2			П			
No.			Constituents		PF	F	L.	PF	F	I.	PF	F	b	
	Lit.	Exp.		(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)		
1	979	979	β-Pinene	-	-	-	-	-	-	0.63	-	-	1, 2	
2	991	990	Dehydro1,8-cineol	-	-	-	-	-	0.50	-	-	1.30	1, 2	
3	1189	1196	α-Terpineol	-	-	-	-	-	1.01	-	-	0.69	1, 2, 3	
4	1290	1293	Thymol	0.54	-	-	-	-	-	0.63	-	_	1. 2. 3	
5	1377	1373	a-Consene	2.23	0.66	2.09	0.49	2.40	2.42	-	-	_	1, 2, 3	
6	1391	1392	ß Elemene	2 69	-		_			2 67	0.87	1.67	1 2	
7	1/00	1/12	cis-Carvonbyllene	2.09	_	_	_	_	_	2.67	0.74	1.07	1 2 3	
8	1419	1418	trans-Caryophyllene	0.50			0.44	1 91	1.98	1.05	0.63	1.43	1, 2, 3 1 2 3	
0	1/136	1446	Nervl acetone	0.50			0.77	1.71	1.90	3.48	0.05	2.68	1, 2, 3	
10	1454	1445	Neoclovene	-						5.40		1.83	1, 2 1 2	
11	1455	1454	α-Humulene	1.83	_	_	0.13	2 14	_	_	_	-	1,2	
12	1455	1457	Alloarmadendrene	1.05	_	_	0.15	2.14	1 12	_	_	_	1, 2, 3	
12	1482	1468	Anoamiadendrene	1.22	1 / 3	3 3/	0.20	2.60	1.12	-	3 66	-	1,2	
13	1402	1400	y-none	4.57	1.45	5.54	0.45	2.00	1.56	5.02	2.16	1.05	1, 2	
14	14//	1471	γ-Gurjunene	-	-	-	-	-	-	5.05	2.10	1.05	1, 2	
15	1480	1472	γ-Muurolene	5.30	0.92	-	0.61	-	-	5.03	1.50	3.98	1, 2	
16	1481	14/9	ar-Curcumene	0.59	-	-	0.61	-	-	-	-	-	1,2	
17	1490	1486	β-Selinene	2.46	1.15	1.38	0.19	3.63	3.85	1.40	0.76	5.70	1,2	
18	1493	1493	δ-Selinene	1.72	-	2.07	0.28	1.87	1.90	-	1.86	-	1, 2	
19	1496	1489	Valencene	-	-	-	-	-	-	1.22	3.82	2.19	1, 2	
20	1500	1496	α-Muurolene	0.53	-	0.54	0.19	-	1.11	-	-	-	1, 2	
21	1514	1515	γ-Cadinene	1.23	0.62	0.87	0.53	1.52	1.56	-	-	-	1, 2	
22	1539	1536	α-Copaen-11-ol	0.48	-	1.77	0.29	1.51	1.55	-	-	-	1, 2	
23	1563	1562	E-Nerolidol	50.16	70.42	54.71	88.66	40.14	41.19	37.72	44.38	47.07	1, 2, 3	
24	1569	1569	Longipinanol	-	-	-	-	-	-	1.24	0.73	1.48	1, 2	
25	1564	1566	epi-Longipinanol	1.32	-	0.93	-	-	-	0.07	-	0.54	1, 2	
26	1583	1579	Caryophyllene oxide	3.56	2.24	4.11	2.20	6.65	6.81	5.13	3.32	7.10	1, 2, 3	
27	1582	1585	Globulol	-	-	-	-	-	-	0.94	-	-	1, 2	
28	1596	1592	ar-Dihydroturmerone	-	3.65	-	-	-	-	-	-	-	1, 2	
29	1596	1593	Fokineol	2.30	-	-	0.71	5.77	-	2.94	3.39	2.32	1, 2	
30	1601	1603	Guaiol	-	-	1.02	-	-	-	0.67	-	-	1, 2	
31	1608	1606	Humulene epoxide II	-	-	-	0.09	1.40	-	-	-	-	1, 2	
32	1608	1613	cis-Isolongifolanone	-	-	-	-	-	-	0.45	0.89	-	1, 2	
33	1619	1614	Isolongifolan-7a-ol	-	-	-	0.12	1.70	1.76	-	-	-	1, 2	
34	1637	1621	Gossonol	-	-	-	-	-	-	6.54	-	-	1, 2	
35	1613	1623	trans-Isolongifolanone	0.91	1.01	1.92	-	-	-	0.84	3.11	1.39	1, 2	
36	1641	1631	Alloarmadendrene epoxide	1.64	1.06	1.73	0.62	1.62	-	-	-	-	1, 2	
37	1641	1632	Caryophylla-4-(14)8(15)-dien-5-	1.14	0.88	1.21	-	1.67	1.11	2.98	1.40	0.59	1, 2	
			β-ol											
38	1651	1635	β-Eudesmol	3.04	1.48	7.59	-	-	-	1.34	1.30	1.04	1, 2	
39	1654	1649	α-Eudesmol	-	-	-	-	2.17	2.20	-	-	-	1, 2	
40	1648	1652	Agarospirol	3.30	5.24	5.21	1.11	4.91	5.09	10.66	11.79	7.11	1, 2	
41	1667	1662	Intermedeol	-	-	-	0.17	-	-	-	-	0.84	1, 2	
42	1661	1664	E-Amyl-cinnamic alcohol	-	-	0.51	-	-	-	-	-	-	1, 2	
43	1682	1683	Ishwarone	0.96	1.76	2.32	0.48	2.41	3.55	1.28	2.75	5.04	1, 2	
44	1691	1698	z-α-trans-Bergamotol	-	0.61	-	0.49	-	-	-	2.09	-	1, 2	
45	1745	1740	γ-Costol	-	1.55	-	0.41	2.86	2.81	0.66	2.08	-	1, 2	
46	1760	1761	Benzyl benzoate	-	-	-	0.13	-	-	-	-	-	1, 2	
47	1766	1762	β-Costol	0.76	-	1.64	-	6.19	6.15	1.24	-	-	1, 2	
48	1773	1765	α-Costol	1.02	4.58	4.22	-	4.91	4.87	-	4.78	1.44	1, 2	
Mone	oterpene	hydroca	rbons (1)	-	-	-	-	-	-	0.63	-	-		
Oxygenated monoterpenes (2, 3, 9)					-	-	-	-	1.51	3.48	-	4.67		
Sesqu	Sesquiterpene hydrocarbons (5-8, 10-12, 14, 15, 17-21)				3.35	6.95	3.06	13.47	13.94	19.07	12.34	19.33		
Oxygenated sesquiterpenes (22-27, 29-33, 35-41, 43-45,					92.26	91.72	95.78	86.51	78.47	68.16	85.67	75.96		
47-48	3)													
Phen	yl propa	noids (4,	16, 28, 34, 42, 46)	1.13	3.65	0.51	0.74	-	-	7.17	-	-		

^aLinear retention index on a DB-5 column; ^bIdentification method: 1 = linear retention index; 2 = identification based on comparison of mass spectra; 3 = Co-injection with standard compounds. Serial numbers of components detected in each class are listed beside the Class name.



Fig. 1. PCA scores plot, PCA bi-plots and dendrogram based on Table-1 data

Fig. 1 represents the resulted PCA scores plot, PCA biplots and dendrogram corresponding to the VOCs emissions obtained from the whole aerial parts of the plant during the vegetative state (Table-1).

Two crossed clusters were obtained, one corresponding to the plant data collected from MID-1 and the other corresponded to the plant samples collected from MID-2, indicating similarities in the chemical composition of the emission profiles from the two locations, the third location which belongs to the collections from a different bio-geographical zone (IT) was quite different. Every point in Fig. 1 of the three clusters represents the data obtained from the collection of the plant in particular month from the three different locations and the shown clusters in this figure clearly reveals no significant differences in the chemical compositions extracted from different months in MID-1 and MID-2. Further investigation of the data has been carried out in an attempt to identify the major chemical constituents responsible for the observed behaviour during the vegetative state of the plants' life cycle. Hence, bi-plots (scores and loadings) PCA were created. As can be concluded, γ muurolene, E,E- α -farnesene, methyl salicylate, γ -gurjunene, E-nerolidol and myrceneol had the greatest impact on the similarity in the first two PCs. For more analysis, CA was applied to the same data matrix used for the PCA. The resulted dendrogram of this test is shown in Fig. 1 using complete linkage distances between the samples (organs). The obtained results were very consistent to those obtained from the PCA application revealing strong relation between the emitted VOCs belonging to MID collection during the vegetative period.

PCA scores plot, PCA bi-plots and dendrogram were generated for the data given in Table-2 (Fig. 2). The results during the vegetative period were in agreement with the two crossed clusters correspond to obtained MID-1 and MID-2 collections, revealing good similarities in the composition of their emission profiles while the third location showed a different emission profile.

The main constituents responsible for this behaviour during the flowering period that had the greatest impact on the similarity included β -selinene, *n*-heptanol, dehydroaromadendrane, γ -muurolene, alloarmadendrene, α -cubebene and γ -cadinene and to a lower impact each of *cis*-3-hexenyl acetate and *cis*caryophyllene. Also, it was noticed that there is a strong relations between all IT organs emissions supporting its separation in a different cluster.

The variation in the chemical composition of the hydrodistilled essential oils was also statistically investigated, the results of the PCA, bi-plots (scores and loadings) PCA and dendrogram are shown in Fig. 3. In agreement with present results obtained from the emission profiles during vegetative and flowering stages, the two crossed clusters corresponding



Fig. 2. PCA scores plot based on Table-2 data



to MID-1 and MID-2 collections revealed quite similar chemical composition. Despite the observed intersection of IT with both MID-1 and MID-2, it can be noticed easily that most of the points belonging to the IT collections (PF buds (7) and F (8)) are out of the intersection area of MID-1 and MID-2. Components having the high impact on the observed similarity of the first two principal components included α -costol, γ -costol, α -copaen-11-ol, alloarma-dendrene, ar-curcumene, epilongipinanol, *cis*-caryophyllene, β -elemene and γ -muurolene, while agarospirol, E-nerolidol and *trans*-isolongifolanone despite their less impact still had important role in the observed similarity.

Conclusion

The current investigation is the first report presenting the geographical and seasonal variation effects on the chemical composition of I. viscosa using two different extraction methods SPME and hydrodistillation. The current results, revealed that plants collected from the Mediterranean (MID-1 & 2) and Irano-Turanian (IT) bio-geographical zones, exhibited quite different qualitative and quantitative variations in their content (emitted scent and essential oil), as confirmed from the statistical analysis results. Such variations could be mainly attributed to exogenous factors (environmental) like soil nature, light, water and other related climatic conditions. It was noticed that E-nerolidol was the main component detected in the hydrodistilled oils of the different organs. Careful inspection of the literature indicates that there are several chemotypes of I. visocsa. Regarding the current findings, E-nerolidol is the main chemotype of I. viscosa growing wild in different bio-geographical zones of Jordan.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

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