Original Article:

Chemical Composition of Methanolic Extracts of *Scutellaria orientalis* L.: Digitoxin and Neocurdione Detection by Gas Chromatography/Mass Spectrometry

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ABSTRACT

Background: *Scutellaria orientalis* subsp. virens is a species of Scutellaria genus (the Lamiaceae family). The aqueous root extract of *S. orientalis* has been traditionally used by Iranians to treat neurological disorders, dermatitis, and bronchitis. The active compounds in the roots and shoots of *S. orientalis* are mainly flavones such as baicalin, wogonoside, scutellarin, and their aglycons. These compounds have anticancer, anti-inflammatory, antiviral, and antibacterial activities.

Objectives: This study aimed to identify the root and shoot methanolic extracts components of *S. orientalis*.

Methods: The Gas Chromatography/Mass Spectrometry (GC/MS) method was used to identify components of *S. orientalis*.

Results: GC/MS analysis of methanolic root extract identified 63 components, such as flavonoids, fatty acids, ketones, glucosides, amino acids, steroids, and alkaloids, with flavonoids like wogonin (12.6%) as the main components. *S. orientalis* methanolic shoot extract analysis also identified 79 components such as ketones, alcohols, sesquiterpenes, fatty acids, steroids, vitamins, and diterpenoids, with digitoxin and neocurdione as the main components. In addition, the Fourier Transform Infrared (FTIR) analysis of roots and shoots confirmed the presence of normal polymeric O-H stretch, aliphatic alkenes, aromatic alkenes, amides, ammonium ions, alcohols, ethers, carboxylic acids, esters, amines, and alkanes in the structure of identified compounds by GC/MS analysis.

Conclusion: The obtained results revealed the presence of biologically active compounds that can be used for various herbal formulations.

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Introduction

Scutellaria genus, as a member of the Lamiaceae family, has been distributed worldwide with over 350 species [1]. Twenty species and two hybrids of this genus exist in Iran [2]. *S. orientalis* is a perennial herb and medicinal plant which grows in Tabriz City, Iran. It grows on soils with little clay, at the height of 1650 m above sea level, and an annual rainfall of 250 to 300 mm. All parts of this plant, including leaves, fruits, roots as well as seeds, are widely used in Iranian traditional medicine to treat constipation, wounds, stress, neurological disorders, dermatitis, bronchitis, and inflammation [3]. Pharmacological properties of the Scutellaria genus are due to the presence of 4′-deoxyflavones such as chrysin, baicalein, wogonin, and their glycosides (baicalin, wogonoside) [4].

Baicalein, wogonin, chrysin, pinocembrin, baicalin, skullcapflavon II, and wogonoside are the main flavonoids of *S. orientalis* species [5, 6]. *S. orientalis* essential oil contains different classes of terpenes, such as monoterpenes and sesquiterpenes, which among them, oxygenated monoterpenes (49.8%) constitutes the major part [7]. Acteoside, verbascoside, allysonoside, and martynoside are the main phenylethanoid glycosides determined from *S. orientalis* root and shoot [6]. Identifying natural plant compounds has advanced from product quality assurance to fundamental research. Regardless of the considerable progress in analytical techniques in the past few decades, identifying unknown compounds has remained a complex problem. Analytical methods such as Gas Chromatography (GC) and Gas Chromatography/Mass Spectrometry (GC/MS) are frequently used to recognize plant extract components. Measurement of mass fragmentation ions (m/z) of chemical compounds and comparison with available mass spectrum collections of known compounds based on similar fragmentation patterns is the usual approach to confirm compound identification. This study aimed to collect *S. orientalis* from its habitat, including limited populations in Tabriz, Iran, and identify and quantify its phytochemical profiles by GC/MS analysis.

Materials and Methods

Plant material collection

S. orientalis roots and shoots were collected from Marand City, Iran, during the summer (July 2018). The plant was identified by Dr Talebpour (plant taxonomy department, University of Tabriz, Tabriz, Iran). A voucher specimen was deposited in the herbarium of the East-Azerbaijan Agricultural and Natural Resources Research and Education Center, Tabriz, Iran (No. 2488).

Preparation of methanolic extracts

Roots and shoots of *S. orientalis* were used for phytochemical studies. Approximately 10 g of plant roots and shoots was extracted using 30 mL of methanol solvent. The extraction was carried out at room temperature by the percolation method for 48 hours. The extract was subsequently filtered. The methanol solvent was concentrated to 1 mL. About 1 µL of the methanolic extract was used for GC/MS analysis to identify phytochemical compounds.

Gas Chromatography/Mass Spectrometry (GC/MS)

The qualitative assessment of methanolic extracts was carried out by GC/MS. GC analysis was conducted in a Hewlett-Packard (HP, Palo Alto, CA) HP 7890A gas chromatograph system. Helium (99.999%) was utilized as the carrier gas at the flow rate of 1 mL/min. The injection port temperature was set at 240°C; column temperature was firstly held at 40°C for 1 min, and then slowly increases to 240°C at the rate of 3°C/min.

Identification and quantification of the compounds

The chemical components of the methanolic extracts were determined by comparing their mass spectra with those of the GC/MS library (WILEY 7n D. 04.00 and NIST). The relative percentage of methanol extract components was quantified based on GC peak areas.

Fourier Transform Infrared (FTIR) spectrophotometer

The *S. orientalis* root and shoot methanolic extracts were subjected to Fourier Transform Infrared (FTIR) analysis to identify functional groups. FTIR spectra were recorded in the infrared region of 400 to 4000 cm^{-1} using a Bruker FT-IR spectrophotometer as KBr (Potassium Bromide) disks [8].

Results and Discussion

The extraction and analysis of plant compounds are crucial for developing, improving, and quality control of herbal formalization. Therefore this study was directed to determine the bioactive compounds of methanolic extracts of *S. orientalis* by GC/MS analysis. The

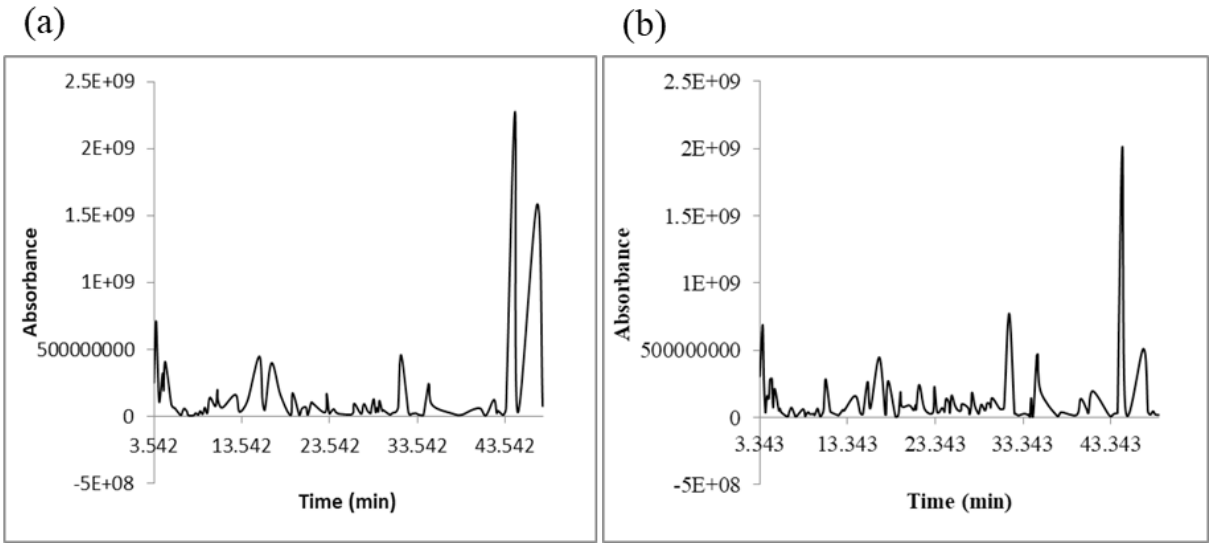

Figure 1. GC/MS chromatogram of methanolic root and shoot extracts of *S. orientalis*

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bioactive constituents, along with their percentage (%), are presented in Tables 1 and 2. GC/MS chromatograms of *S. orientalis* methanolic root and shoot extract are shown in Figure 1.

S. orientalis methanolic root extract components

GC/MS chromatogram of *S. orientalis* methanolic root extract is shown in Figure 1a. Sixty-three components including flavonoids (15.3%), esters (12.5%), fatty acids (10.5%), glycoside (7.37%), ketones (5.9%), alcohols (3.44%), steroids (3.4%), acetates (2.25%), amides (0.46%), aldehydes (0.27%), ethers (0.24%), amino acids (0.15%), and alkaloids (0.08%) were determined in the methanolic root extract of *S. orientalis* (Table 1).

By comparative inspection, the main components in terms of their relative abundance were wogonin; propanoic acid,2-oxo-,methyl ester; n-hexadecanoic acid; 5-hydroxymethylfurfural; 2-propanone; 1-hydroxy-,ethyl iso-allocholate; 2-methoxy-4-vinylphenol; ethanethioic acid; and s-(dihydro-2,5-dioxo-3-furanyl) ester, corresponding to 18.1%, 12.6%, 4.75%, 4.2%, 3.55%, 3.44% 3.39%, 3.2% and 2.55%, respectively. A molecular docking and dynamics study by Malathi et al. revealed that ethyl iso-allocholate isolated from a medicinal rice variety, Karungkavuni, can serve as a potent inhibitor for dihydropteroate synthase activity in Escherichia coli [9]. Figure 2 shows the mass spectrum of some root-specific biologically active compounds determined in *S. orientalis* methanolic extract.

In the same context described above, the minor components were within a range of 0.05% to 2%. Specifically, 1-nitropyrrolidine, γ-chlorobutyrophenone, and 1,2-cyclohexanedione represented the components with the lowest amounts in methanolic extract of *S. orientalis* root. The obtained results revealed the presence of several bioactive components.

S. orientalis shoot methanolic extract components

GC/MS chromatogram of *S. orientalis* methanolic shoot extract is shown in Figure 1b. GC/MS analysis of *S. orientalis* methanolic shoot extract identified 79 components. These components belonged to multiple chemical groups, including fatty acids (21.6%) with palmitic acid (5.38%) as the main acid, esters (9.16%) with 9,12,15-octadecatrienoic acid, 2,3-dihydroxypropyl ester, (Z,Z,Z)- (3.3%) as the main ester, ketones (6.7%) with methylglyoxal (2.13%) as the main ketone, aldehyde (3.9%), steroids (3.33%), alcohols (3%), glycosides (4.94%), acetates (1.12%), amides (2.4%), sesquiterpenes (1.98%), diterpenes (phytol; 1%), carotenoid (0.42%) and vitamins (0.2%) (Table 2). The main components in terms of relative abundance were palmitic acid, dibenz(a,c)cyclohexane, 2,4,7-trimethoxy, ethyl iso-allocholate, 9,12,15- octadecatrienoic acid, 2,3-dihydroxypropyl ester, (Z,Z,Z)-, 9,12-octadecadienoic acid (Z,Z)-, 2-methoxy-4-vinylphenol and o-guaiacol, corresponding to 5.38%, 3.56%, 3.33%, 3.27%, 3.2%, 3.12% and 3.07%, respectively.

Methylglyoxal was identified as the most influential antibacterial component of manuka honey, derived from the Manuka tree (*Leptospermum scoparium*) in New Zealand [9]. Methylglyoxal has antibacterial activity against Gram-positive bacteria, including methicillin-re-

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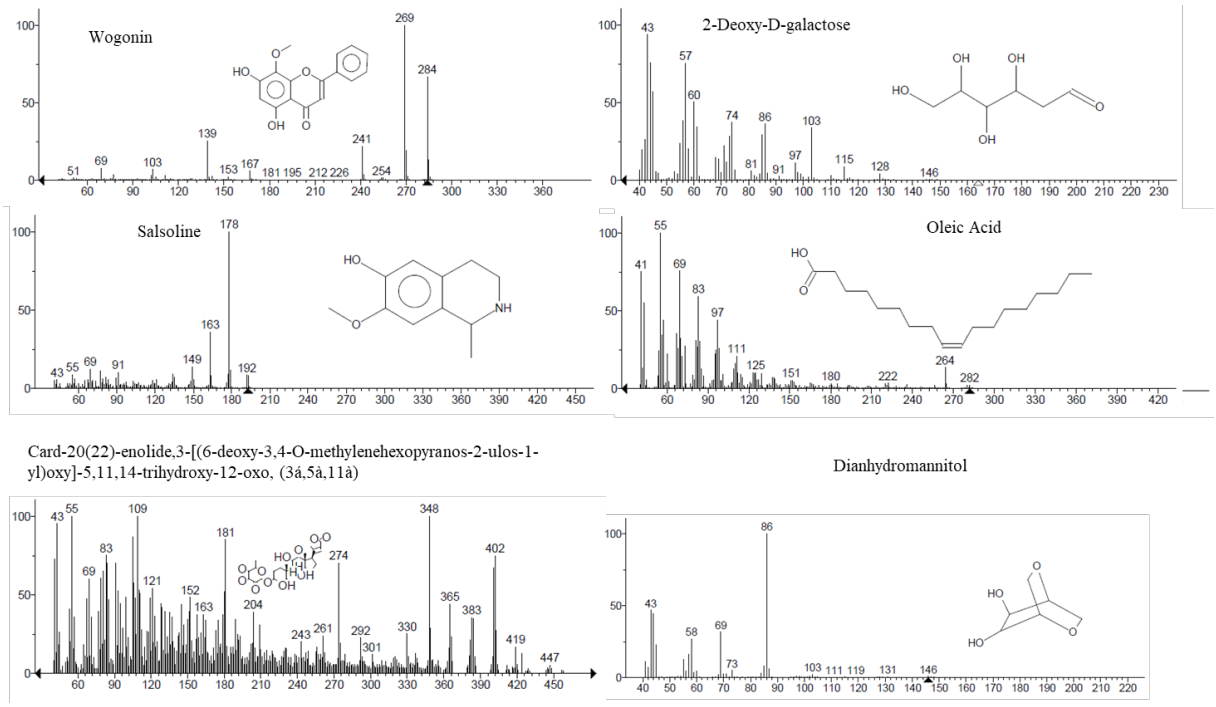

Figure 2. Mass spectrum of main biologically active compounds from *S. orientalis* methanolic root extract

sistant *Staphylococcus aureus* and vancomycin-resistant Enterococcus. Hayashi et al. reported the effect of methylglyoxal against Multidrug-Resistant *Pseudomonas aeruginosa* (MDRP) using 53 clinically isolated strains [10]. They also assessed the impact of omitting the five multidrug efflux systems in *P. aeruginosa*, as well as the efflux systems in *Escherichia coli* and *Salmonella enterica* serovar Typhimurium, on minimum inhibitory concentrations of methylglyoxal. Their results revealed that methylglyoxal inhibits the growth of MDRP at concentrations of 128–512 µg/mL (1.7–7.1 mM) and was not recognized by drug efflux systems [10]. Figure 3 shows the mass spectrum of some shoot-specific biologically active compounds recognized in *S. orientalis* methanolic extract. Conversely, the minor components among the entire components of *S. orientalis* shoot methanolic extract were those with a relative abundance range of 0.052%-2.28%, representing 1-nitropyrrolidine, γ-chlorobutyrophenone, and 1,2-cyclohexanedione as the lowest ones. GC-MS analysis of *S. orientalis* shoot extract identified several bioactive components.

The amount of volatile oil compounds, including sesquiterpenes, was comparatively less than other bioactive compounds (Table 2). However, Gharari et al. study on phytochemical screening of the methanolic root extract of *Scutellaria orientalis* subsp. *bornmuelleri* revealed the presence of several groups of terpenes, including sesquiterpenes and monoterpenes, which among them sesquiterpene hydrocarbons (12.66%) were more abundant [11]. Among different examined extracts of *Scutellaria orientalis* subsp. *bornmuelleri*, including n-hexane, CH_2Cl_2, EtOAc fractions, and aqueous and methanol extracts, the methanol extract had the most antiproliferative activity against HCT-116 and SW-480 cells at 48 h with IC_{50} values of 614.5 and 592.3 µg/mL, respectively [11]. Some of the identified compounds such as methylglyoxal (2.130%), phytol (1.008%), neocurdione (0.599%), spathulenol (0.48%), β-copaene (0.647%) and vanillin (1.322%) have strong pharmacological activities. Phytol is diterpene alcohol from chlorophyll. Phytol and its derivative, phytanic acid, exert various biological properties. It is an important essential oil used as a food additive and in medicinal fields as a potential candidate for an extended range of applications in the pharmaceutical industry. There is a wealth of evidence that phytanic acid has a critical function in developing pathophysiological states. Recent investigations with phytol indicated antinociceptive, antioxidant, anxiolytic, cytotoxic, metabolism-modulating, autophagy-inducing, apoptosis-inducing, immune-modulating, anti-inflammatory, and antimicrobial effects [12, 13]. PPARs- and NF-κB-mediated activities are responsible mechanisms for some of the bioactivities of phytol [14]. De Moraes et al.'s research on the application of phytol against neglected tropical disease schistosomiasis revealed its promising antischistosomal properties in vitro and a mouse model of *Schistosomiasis mansoni* [15]. Several studies demonstrated that spathulenol has antioxidant, anti-inflammatory, antiproliferative, and antimycobacterial activities [16]. Vanillin, another

Table 1. The chemical composition of methanolic root extracts of *S. orientalis*

No.	Compounds	Mw	Formula	Negative Ion Mode ([M-H]-) at m/z	%	RT (min)
1	Methylazoxymethanol acetate	132.12	$C_4H_8N_2O_3$	42, 43, 46, 55, 60, 73, 90	2	3.533
2	Hydroxyacetone	74.08	$C_3H_6O_2$	42, 43, 45, 53, 55, 57, 73, 74, 75	3.44	3.947
3	Arabinose	150.13	$C_5H_{10}O_5$	43, 55, 60, 73, 85, 96, 119, 126, 133, 149	1.04	4.129
4	Ethanethioic acid, S-(dihydro-2,5-dioxo-3-furanyl) ester	174.17	$C_6H_6O_4S$	43, 55, 87, 102, 174	2.55	4.501
5	Propanoic acid, 2-oxo-, methyl ester	102.09	$C_4H_6O_3$	43, 55, 60, 73, 79, 86, 96, 102	4.75	4.788
6	Cyclohexanone, 3-hydroxy-	114.14	$C_6H_{10}O_2$	43, 55, 60, 68, 71, 81, 86, 96, 99, 114	0.72	5.509
7	2-Furanmethanol	98.10	$C_5H_6O_2$	41, 53, 69, 81, 98	0.51	5.733
8	Guanosine	283.24	$C_{10}H_{13}N_5O_5$	43, 57, 60, 69, 73, 78, 86, 97, 114, 124, 133, 151, 161, 175, 194, 205	0.07	6.584
9	2-Propenamide, N-(1-cyclohexylethyl)-	181.27	$C_{11}H_{19}NO$	41, 44, 55, 67, 72, 84, 98, 110, 166, 182	0.5	7.030
10	2-Decenal, (E)-	154.25	$C_{10}H_{18}O$	41, 55, 70, 83, 98, 110, 121, 136, 154	0.3	7.307
11	1,5-Hexadiene-3,4-diol, 3,4-dimethyl-	142.20	$C_8H_{14}O_2$	43, 55, 71, 84, 91, 109, 124	0.06	7.445
12	9-Oxa-bicyclo(3.3.1)nonane-1,4-diol	158.19	$C_8H_{14}O_3$	43, 58, 70, 86, 97, 112, 130, 140, 158	0.08	8.614
13	1-Hexanol, 2-ethyl-	130.23	$C_8H_{18}O$	41, 57, 70, 83, 98, 112	0.3	8.795
14	l-Gala-l-ido-octose	240.21	$C_8H_{16}O_8$	43, 57, 61, 73, 83, 97, 106, 124, 145, 163, 175, 193, 211	0.64	9.329
15	2-Deoxy-D-galactose	164.16	$C_6H_{12}O_5$	43, 57, 60, 74, 81, 86, 91, 97, 103, 115, 128, 146	0.18	9.624
16	Acetophenone	120.15	C_8H_8O	43, 51, 63, 77, 91, 105, 120	1.12	9.921
17	Furaneol	128.13	$C_6H_8O_3$	43, 57, 60, 67, 72, 85, 94, 100, 109, 128	0.6	10.474
18	Formic Acid, 4-methoxyphenyl ester	152.15	$C_8H_8O_3$	41, 53, 63, 71, 81, 95, 109, 124, 152	1.6	10.750
19	o-Guaiacol	124.14	$C7H_8O_2$	53, 63, 77, 81, 95, 109, 124	0.9	10.841
20	Imidazole, 2-amino-5-[(2-carboxy)vinyl]-	153.14	$C_6H7N_3O_2$	44, 55, 69, 82, 96, 109, 125, 135, 153	0.52	11.282
21	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl- (DDMP)	144.13	$C_6H_8O_4$	43, 55, 58, 72, 85, 101, 115, 130, 144	2.34	12.812
22	3-Trifluoroacetoxydodecane	282.34	$C_{14}H_{25}F_3O_2$	41, 55, 69, 83, 97, 111, 125, 140, 168	0.26	13.290
23	Glucosamine, N-acetyl-N-benzoyl-	325.31	$C_{15}H_{19}NO7$	43, 51, 60, 68, 72, 77, 83, 88, 100, 105, 114, 122, 130, 142, 154, 167, 176, 185, 192, 204, 216	0.92	14.151
24	5-Hydroxymethylfurfural	126.11	$C_6H_6O_3$	41, 53, 69, 81, 97, 109, 126	3.55	15.533
25	6-O-acetyl-β-d-mannose	222.19	$C_8H_{14}O7$	43, 53, 60, 73, 81, 91, 97, 109, 126, 144, 165, 175, 192	1.29	15.990
26	2-Methoxy-4-vinylphenol	150.17	$C_9H_{10}O_2$	51, 63, 77, 89, 107, 135, 150	3.2	16.925
27	Syringol	154.16	$C_8H_{10}O_3$	40, 51, 65, 79, 96, 111, 125, 139, 154	1.27	17.988
28	[1,1'-Bicyclopropyl]-2-octanoic acid, 2'-hexyl-, methyl ester	322.52	$C_{21}H_{38}O_2$	45, 55, 67, 73, 81, 95, 109, 165, 192, 207, 224, 291, 332	0.06	19.104
29	Vanillin lactoside	476.43	$C_{20}H_{28}O_{13}$	43, 60, 73, 77, 81, 91, 109, 123, 137, 152, 163, 191, 204	1.38	19.359
30	Isochavibetol	164.20	$C_{10}H_{12}O_2$	51, 55, 65, 77, 91, 103, 121, 131, 149, 164	0.44	20.369
31	Cyclopropane tetradecanoic acid, 2-octyl-, methyl ester	394.67	$C_{26}H_{50}O_2$	45, 55, 69, 73, 83, 97, 112, 129, 147, 165, 178, 193, 206, 219, 241, 259	0.54	20.837
32	N-(O-Nitrophenylthio)-L-leucine	284.33	$C_{12}H_{16}N_2O_4S$	43, 51, 69, 77, 86, 98, 125, 138, 154, 163, 178, 206, 222, 238, 252, 284	0.08	21.081

No.	Compounds	Mw	Formula	Negative Ion Mode ([M-H]-) at m/z	%	RT (min)
33	5-Hepten-3-yn-2-ol, 6-methyl-5-(1-methylethyl)-	166.26	$C_{11}H_{18}O$	43, 55, 81, 123, 133, 151, 166	0.83	21.506
34	Benzoic acid, 2-fluoro-4,5-dimethoxy-	200.16	$C_9H_9FO_4$	45, 65, 77, 97, 109, 139, 157, 167, 185, 200	0.73	21.740
35	Pyrrolizidine-3-one-5-ol, ethyl ether	169.22	$C_9H_{15}NO_2$	44, 55, 69, 84, 98, 110, 124, 142, 157, 182, 197	0.24	23.016
36	Pyrrolizin-1,7-dione-6-carboxylic acid, methyl(ester)	197.19	$C_9H_{11}NO_4$	43, 55, 57, 73, 77, 84, 97, 105, 125, 140, 166, 182, 200, 241, 267, 281	0.69	23.066
37	2H-1-Benzopyran-3,4-diol, 2-(3,4-dimethoxyphenyl)-3,4-dihydro-6-methyl-, (2α,3α,4α)-	316.35	$C_{18}H_{20}O_5$	119, 137, 151, 165, 180, 270, 298, 316	1.34	23.207
38	Digitoxin	764.94	$C_{41}H_{64}O_{13}$	43, 58, 69, 73, 81, 95, 113, 124, 131, 147, 162, 175, 203, 219, 231, 243, 259, 277, 303, 323, 339	0.23	23.451
39	Cyclohexane, 1,2,3,4-bis(epoxy)-2,6,6-trimethyl-1-(pent-2-en-4-one-2-yl)-	236.31	$C_{14}H_{20}O_3$	43, 55, 69, 77, 91, 109, 125, 137, 161, 175, 193, 218, 236	0.23	23.473
40	Melezitose	504.44	$C_{18}H_{32}O_{16}$	43, 60, 69, 73, 85, 97, 113, 126, 145, 163, 187, 199	0.92	24.068
41	Tetraacetyl-d-xylonic nitrile	343.29	$C_{14}H_17NO_9$	44, 55, 60, 67, 73, 95, 112, 133, 149, 161, 176, 197, 209, 221, 233, 245, 270, 344	0.15	24.631
42	Dodecanoic acid, 3-hydroxy-	216.32	$C_{12}H_{24}O_3$	43, 55, 69, 83, 96, 112, 126, 138, 151, 180, 200, 213	0.1	25.896
43	10-Heptadecen-8-ynoic acid, methyl ester, (E)-	338.44	$C_{19}H_{30}O_5$	41, 57, 67, 71, 79, 85, 91, 105, 119, 133, 150, 164, 175, 189, 205, 219, 232, 247, 278	0.23	26.236
44	1,3-Benzodioxole, 5-(1-(2-(2-butoxyethoxy)ethoxy)butyl)-		$C_{19}H_{30}O_5$	41, 45, 51, 57, 67, 71, 79, 85, 91, 107, 119, 135, 149, 159, 176, 194, 222, 296, 338	0.76	26.417
45	1b,4a-Epoxy-2H-cyclopenta[3,4]cyclopropa[8,9]cycloundec[1,2-b]oxiren-5(6H)-one, 7-(acetyloxy)decahydro-2,9,10-trihydroxy-3,6,8,8,10a-pentamethyl	424.48	$C_{22}H_{32}O_8$	43, 55, 69, 85, 97, 109, 123, 137, 152, 181, 192, 210, 237, 261, 279, 317, 346, 364, 424	0.14	27.108
46	(E)-4-(3-Hydroxyprop-1-en-1-yl)-2-methoxyphenol	180.20	$C_{10}H_{12}O_3$	65, 77, 91, 103, 124, 137, 147, 180	0.62	27.352
47	9-Octadecenoic acid, (2-phenyl-1,3-dioxolan-4-yl)methyl ester, cis-	444.64	$C_{28}H_{44}O_4$	45, 55, 73, 83, 91, 105, 122, 149, 179, 197, 221, 264, 283, 305, 338, 367	0.73	27.543
48	Paromomycin	615.63	$C_{23}H_{45}N_5O_{14}$	42, 57, 67, 80, 94, 109, 124, 162, 191, 210, 230, 257, 277, 303, 323	0.32	27.841
49	1-Methyl-1-(2-ethylhexyloxy)-1-silacyclohexane	242.47	$C_{14}H_{30}OSi$	41, 55, 77, 103, 129, 143, 171, 199, 227, 242	1.02	28.617
50	2-Myristynoyl pantetheine	484.69	$C_{25}H_{44}N_2O_5S$	42, 55, 71, 83, 100, 143, 184, 213, 250, 283, 354, 407, 440, 484	1.92	28.968
51	9-Octadecenoic acid, (2-phenyl-1,3-dioxolan-4-yl)methyl ester, cis		$C_{28}H_{44}O_4$	45, 55, 73, 83, 91, 105, 122, 149, 179, 221, 264, 283, 305, 338, 367		29.659
52	Cyclopropane butanoic acid, 2-[[2-[[2-[(2-pentylcyclopropyl)methyl)cyclopropyl)methyl]cyclopropyl)methyl]-, methyl	374.60	$C_{25}H_{42}O_2$	41, 55, 67, 74, 81, 95, 107, 121, 135, 149, 168, 185, 199, 227, 270, 298, 334	0.19	30.700
53	9-Hexadecenoic acid		$C_{16}H_{30}O_2$	41, 55, 69, 83, 97, 111, 138, 194, 219, 236, 254		31.359
54	n-Hexadecanoic acid	254.41	$C_{16}H_{32}O_2$	43, 60, 73, 83, 97, 115, 129, 157, 185, 213, 227, 256	4.2	31.593
55	9-Octadecenoic acid, (2-phenyl-1,3-dioxolan-4-yl)methyl ester, trans-	444.64	$C_{28}H_{44}O_4$	43, 57, 73, 91, 105, 149, 179, 207, 223, 238, 264, 282, 338, 367	0.13	32.837
56	9,12-Octadecadienoic acid (Z,Z)-	280.44	$C_{18}H_{32}O_2$	41, 55, 60, 67, 81, 95, 109, 123, 150, 182, 209, 233, 262, 280	1.92	34.792
57	Oleic Acid	282.46	$C_{18}H_{34}O_2$	41, 55, 69, 83, 97, 111, 125, 151, 180, 222, 264, 282	1.86	34.898
58	Octadecanoic acid, 2-hydroxy-1,3-propanediyl ester	625.01	$C_{39}H7_6O_5$	43, 57, 73, 84, 98, 116, 129, 143, 185, 199, 241, 267, 284, 297, 341, 359, 395, 437	0.66	35.249
59	7,8-Epoxylanostan-11-ol, 3-acetoxy-	502.77	$C_{32}H_{54}O_4$	44, 57, 69, 83, 95, 109, 123, 149, 159, 191, 227, 244, 269, 304, 330, 393, 424, 440	0.14	37.694
60	Salsoline	193.24	$C_{11}H_{15}NO_2$	43, 55, 69, 91, 149, 163, 178, 192	0.08	38.576
61	Butanoic acid, 1a,2,5,5a,6,9,10,10a-octahydro-5,5a-dihydroxy-4-(hydroxymethyl)-1,1,7,9-tetramethyl-11-oxo-1	418.52	$C_{24}H_{34}O_6$	43, 55, 71, 91, 107, 122, 135, 151, 177, 213, 241, 299, 330, 369, 387, 418	0.05	41.382
62	Docosanoic acid, 1,2,3-propanetriyl ester	1059.79	$C_{69}H_{134}O_6$	41, 57, 69, 83, 97, 111, 129, 149, 171, 185, 197, 219, 241, 275, 297, 323, 340, 381, 451, 465	0.6	42.796

No.	Compounds	Mw	Formula	Negative Ion Mode ([M-H]-) at m/z	%	RT (min)
63	Card-20(22)-enolide,3-((6-deoxy-3,4-O-methylenehexopyranos-2-ulos-1-yl)oxy)-5,11,14-trihydroxy-12-oxo, (3á,5à,11à)	576.63	$C_{30}H_{40}O_{11}$	43, 55, 69, 83, 109, 121, 152, 163, 181, 204, 243, 261, 274, 292, 301, 330, 348, 365, 383, 402, 419, 447	0.47	43.635
64	Ethyl iso-allocholate	436.62	$C_{26}H_{44}O_5$	43, 55, 69, 81, 95, 121, 145, 171, 197, 223, 253, 272, 313, 382, 400, 418	3.39	42.278
65	Wogonin (Flavone, 5,7-dihydroxy-8-methoxy-)	284.26	$C_{16}H_{12}O_5$	69, 103, 139, 153, 167, 181, 195, 212, 226, 241, 254, 269, 284	12.6	47.100

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S. orientalis shoot methanolic extract compound, is a phenolic aldehyde with anti-carcinogenic [17], antimutagenic [18], and antioxidant [19] activities. It also has antifungal [20] and cytotoxic [21] properties.

One of the main differences between root and shoot content of *S. orientalis* methanolic extract was the presence of flavonoids (15.3%) and glycosides (monosaccharides and disaccharides; 7.37%) with high percentages in the root (Table 3). Wogonin, the major bioactive flavonoid of the Scutellaria genus, was found in root methanolic extract with a high amount (12.6%). Wogonin is an effective anti-inflammatory, antiviral, anticancer, and antibacterial compound [22]. In addition, it has shown protective effects against Alzheimer disease [23].cardiovascular disease [24], and ethanol-induced gastric mucosal damage in the rat model [25].

FTIR spectra of the plant root and shoot

FTIR analysis of dry methanolic root and shoot extract of *S. orientalis* demonstrated the presence of normal polymeric O-H stretch, aliphatic alkenes, aromatic alkenes, amides, ammonium ions, alcohols, ethers, carboxylic acids, esters, and amines with major peaks at 3404, 2923, 2851, 1664, 1407, 1286, and 1046 (Tables 3, 4), respectively in the structure of compounds. The FTIR spectrum of the *S. orientalis* roots and shoot extracts in the form of a KBr pallet is shown in Figure 4. The medium-strong bands at 3416.01 and 3404.07 cm^{-1} in FTIR spectra belong to –NH and OH stretching in primary amines and

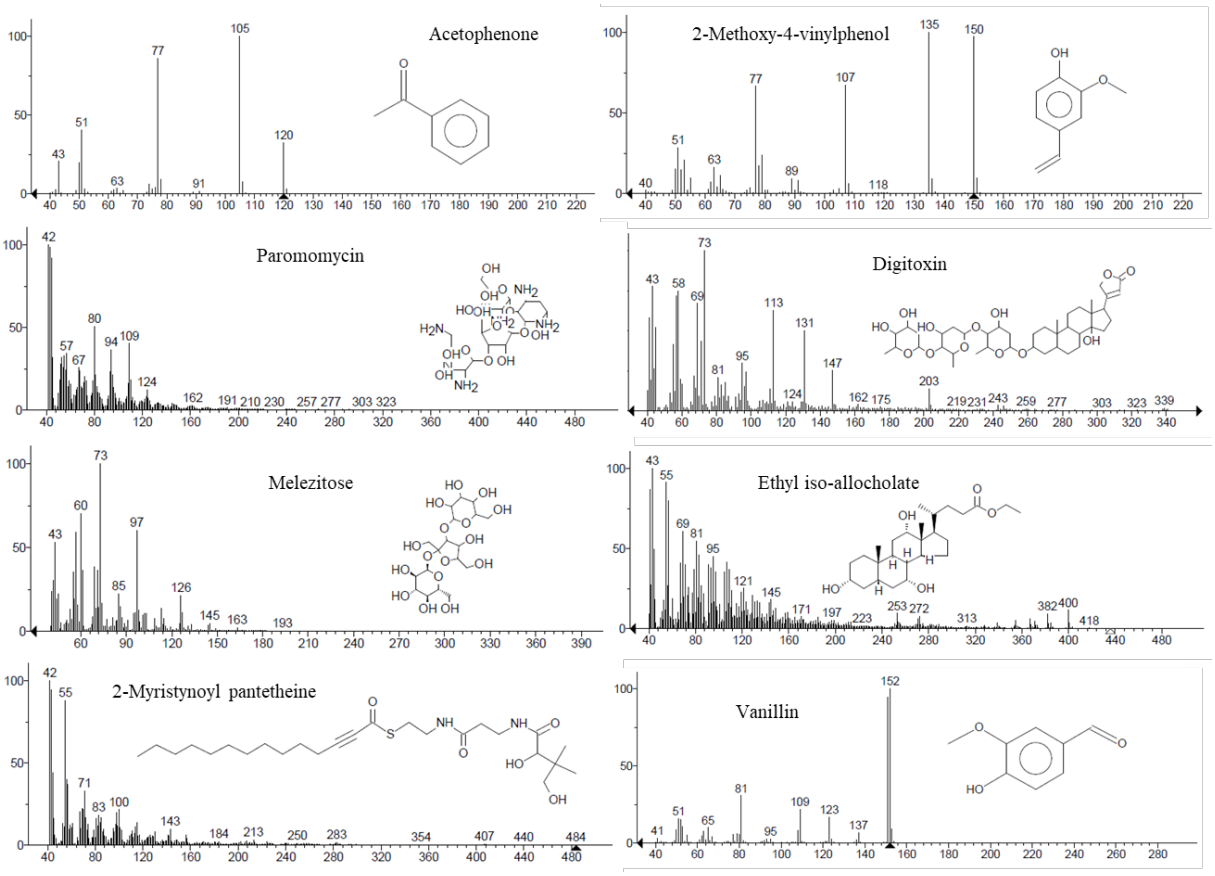

Figure 3. Mass spectrum of main biologically active compounds from *S. orientalis* methanolic shoot extract

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Table 2. The chemical composition of methanolic shoot extracts of *S. orientalis*

No.	Compounds	Mw	Formula	Negative Ion Mode ([M-H]-) at m/z	%	Rt (min)
1	Methylglyoxal	72.06	$C_3H_4O_2$	41, 42, 43, 45, 72	2.13	3.385
2	Hydroxyacetone	74.08	$C_3H_6O_2$	42, 43, 45, 53, 55, 58, 73, 74, 75	2.02	3.797
3	3-Ethoxy-1,2-propanediol	120.15	$C_5H_{12}O_3$	42, 43, 44, 59, 61, 74, 89, 102	0.27	3.976
4	Glyceraldehyde	90.08	$C_3H_6O_3$	42, 43, 45, 57, 61, 73, 90	0.44	4.044
5	n-Propyl acetate	102.13	$C_5H_{10}O_2$	41, 42, 43, 61, 73	1.12	4.161
6	Furan, 2,5-dimethyl-	96.13	C_6H_8O	41, 43, 51, 53, 67, 81, 95, 96	0.98	4.374
7	Propanoic acid, 2-oxo-, methyl ester	102.09	$C_4H_6O_3$	42, 43, 59, 74, 102	2	4.703
8	Dianhydromannitol	146.14	$C_6H_{10}O_4$	43, 44, 57, 58, 86, 103, 111, 119, 131, 146	0.52	4.862
9	2-Propenoic acid, 2-methyl-	86.09	$C_4H_6O_2$	40, 41, 45, 58, 68, 69, 86	1.49	5.021
10	Cyclohexanone, 3-hydroxy-	114.14	$C_6H_{10}O_2$	42, 43, 55, 58, 60, 68, 71, 81, 86, 96, 99, 114	0.36	5.489
11	2-Furanmethanol	98.10	$C_5H_6O_2$	41, 42, 53, 69, 70, 81, 97, 98	0.47	5.638
12	Acetoxyacetone	116.11	$C_5H_8O_3$	42, 43, 60, 73, 86, 101, 116	0.28	5.787
13	1-Nitropyrrolidine –	116.12	$C_4H_8N_2O_2$	41, 42, 43, 55, 69, 85, 100, 116	0.05	6.499
14	4,5-Dihydro-2-methylimidazole-4-one	98.10	$C_4H_6N_2O$	42, 55, 71, 78, 83, 98	0.51	6.966
15	1,5-Hexadiene-3,4-diol, 3,4-dimethyl-	142.20	$C_8H_{14}O_2$	43, 55, 77, 84, 91, 109, 124, 142	0.22	7.243
16	Phenyl-β-D-glucoside	256.25	$C_{12}H_{16}O_6$	43, 57, 66, 73, 85, 94, 107, 127, 145, 162, 256	0.44	8.295
17	1,2-Cyclohexanedione	112.13	$C_6H_8O_2$	43, 55, 70, 83, 97, 112	0.07	8.497
18	9-Oxa-bicyclo[3.3.1]nonane-1,4-diol	158.19	$C_8H_{14}O_3$	43, 58, 70, 86, 97, 112, 130, 140, 158	0.13	8.571
19	1-Hexanol, 2-ethyl-	130.23	$C_8H_{18}O$	41, 43, 53, 55, 57, 67, 70, 77, 83, 98, 112, 130	0.29	8.826
20	Cycloheptanol, 2-chloro-, trans-	148.63	$C_7H_{13}ClO$	41, 44, 58, 68, 79, 84, 95, 98, 113, 130, 148	0.18	9.039
21	3-O-Benzyl-d-glucose	270.28	$C_{13}H_{18}O_6$	43, 51, 57, 61, 73, 77, 79, 91, 102, 108, 163, 239, 270	0.2	9.475
22	1,6-Anhydro-2,4-dideoxy-β-D-ribo-hexopyranose	130.14	$C_6H_{10}O_3$	41, 43, 57, 58, 84, 100, 113, 129	0.13	9.602
23	Acetophenone	120.15	C_8H_8O	43, 50, 51, 63, 74, 77, 91, 105, 120	0.47	9.911
24	γ-Chlorobutyrophenone	182.65	$C_{10}H_{11}ClO$	41, 51, 77, 105, 120, 133, 147, 164, 182	0.06	10.208
25	Furaneol	128.13	$C_6H_8O_3$	43, 57, 60, 72, 85, 100, 110, 128	0.34	10.538
26	o-Guaiacol	124.14	$C_7H_8O_2$	53, 63, 77, 81, 95, 109, 124	3.07	10.846
27	Imidazole, 2-amino-5-[(2-carboxy)vinyl]-	153.14	$C_6H_7N_3O_2$	44, 55, 69, 82, 96, 109, 125, 135, 153	0.32	11.324
28	l-Gala-l-ido-octose	240.21	$C_8H_{16}O_8$	43, 57, 61, 73, 83, 97, 106, 124, 145, 163, 175, 193, 211	0.26	11.526
29	d-Glycero-d-ido-heptose	210.18	$C_7H_{14}O_7$	43, 60, 73, 85, 103, 133, 149, 179, 193	0.12	12.313
30	β-D-Glucopyranose, 4-O-β-D-galactopyranosyl-	342.30	$C_{12}H_{22}O_{11}$	43, 60, 73, 85, 97, 113, 131, 145, 163, 173, 191, 201	0.41	12.812
31	Glucosamine, N-acetyl-N-benzoyl-	325.31	$C_{15}H_{19}NO_7$	43, 51, 60, 72, 77, 83, 100, 105, 114, 122, 130, 154, 176, 204, 216, 229, 247,266	1.13	14.194
32	Paromomycin	615.63	$C_{23}H_{45}N_5O_{14}$	42, 57, 67, 80, 94, 109, 124, 145, 162, 173, 191, 201, 211, 223, 243, 257, 267, 277, 287	0.23	14.810

No.	Compounds	Mw	Formula	Negative Ion Mode ([M-H]-) at m/z	%	Rt (min)
33	5-Hydroxymethylfurfural	126.11	$C_6H_6O_3$	41, 53, 69, 81, 97, 109, 126	1.85	15.608
34	6-Acetyl-β-d-mannose	222.19	$C_8H_{14}O_7$	43, 53, 60, 73, 81, 91, 97, 109, 126, 144, 165, 175, 192	0.48	16.011
35	2-Methoxy-4-vinylphenol	150.17	$C_9H_{10}O_2$	51, 63, 77, 89, 107, 135, 150	3.12	16.926
36	Octanamide, N-(2-mercaptoethyl)-	203.34	$C_{10}H_{21}NOS$	43, 57, 72, 85, 98, 112, 119, 127, 132, 144, 156, 170, 185, 203	0.15	17.627
37	Syringol	154.16	$C_8H_{10}O_3$	40, 51, 65, 79, 93, 111, 125, 139, 154	1.89	17.978
38	Vanillin	152.15	$C_8H_8O_3$	51, 65, 81, 95, 109, 123, 137, 152	1.32	19.349
39	10-Heptadecen-8-ynoic acid, methyl ester, (E)-	278.43	$C_{18}H_{30}O_2$	41, 57, 67, 71, 79, 85, 91, 105, 119, 133, 150, 164, 175, 189, 205, 219, 229, 247, 278	0.82	20.390
40	Desulphosinigrin	279.31	$C_{10}H_{17}NO_6S$	43, 60, 73, 85, 103, 127, 145, 163, 187, 213, 262	0.51	20.826
41	β-copaene	204.35	$C_{15}H_{24}$	41, 55, 67, 79, 91, 105, 119, 133, 147, 161, 176, 189, 204	0.65	21.071
42	3-Hydroxy-α-ionene	208.30	$C_{13}H_{20}O_2$	55, 69, 81, 91, 95, 109, 125, 147, 175, 190	0.42	21.188
43	1,4-Dimethoxy-2,3-dimethylbenzene	166.22	$C_{10}H_{14}O_2$	41, 53, 65, 77, 91, 108, 121, 137, 151, 166	1.7	21.485
44	2-(3,4-Dimethoxyphenyl)-6-methyl-3,4-chromandiol	316.35	$C_{18}H_{20}O_5$	-	1.6	23.218
45	Spathulenol	220.35	$C_{15}H_{24}O$	43, 55, 67, 79, 91, 105, 119, 131, 145, 159, 177, 187, 205, 220	0.48	23.462
46	Caryophyllene oxide	220.35	$C_{15}H_{24}O$	43, 55, 69, 79, 93, 109, 121, 138, 161, 177, 205, 220	0.25	23.590
47	Tetraacetyl-d-xylonic nitrile	343.29	$C_{14}H_{17}NO_9$	44, 55, 60, 67, 73, 95, 112, 133, 149, 161, 176, 197, 209, 221, 233, 245, 270	0.97	24.589
48	Melezitose	504.44	$C_{18}H_{32}O_{16}$	43, 60, 69, 73, 85, 97, 113, 126, 145, 163, 189	1.05	24.823
49	Digitoxin	764.94	$C_{41}H_{64}O_{13}$	43, 58, 69, 73, 81, 113, 124, 131, 147, 162, 175, 203, 219, 231, 243, 259, 277, 303	0.23	24.993
50	Cinnamic acid, 4-hydroxy-3-methoxy-, (5-hydroxy-2-hydroxymethyl-6-[2-(4-hydroxy-3-methoxyphenyl)ethoxy]-	652.64	$C_{31}H_{40}O_{15}$	43, 55, 77, 91, 105, 121, 137, 151, 168, 177, 194, 215, 229, 268, 283, 296, 316, 330, 339, 354, 368	2.21	25.216
51	1-Heptatriacotanol	537.00	$C_{37}H_{76}O$	43, 55, 69, 81, 95, 107, 121, 133, 147, 161, 175, 190, 203, 229, 243, 257, 272, 285	1.26	25.567
52	1b,4a-Epoxy-2H-cyclopenta[3,4]cyclopropa[8,9]cycloundec[1,2-b]oxiren-5(6H)-one, 7-(acetyloxy)decahydro-2,9,10-trihydroxy-3,6,8,8,10a-pentamethyl	424.48	$C_{22}H_{32}O_8$	43, 55, 73, 85, 95, 109, 123, 137, 163, 181, 207, 223, 242, 260, 283, 300, 329, 348	0.98	27.108
53	Acetamide, N-methyl-N-[4-(3-hydroxypyrrolidinyl)-2-butynyl]-	210.27	$C_{11}H_{18}N_2O_2$	43, 56, 68, 81, 91, 117, 124, 137, 155, 167, 192, 210	2.24	27.565
54	Neocurdione	236.35	$C_{15}H_{24}O_2$	41, 55, 69, 82, 95, 109, 121, 137, 167, 180, 193, 216, 236	0.6	27.905
55	24(R), 25-Dihydroxyvitamin D3	416.63	$C_{27}H_{44}O_3$	43, 55, 59, 69, 81, 91, 105, 118, 129, 136, 145, 158, 176, 189, 207, 221, 235, 253, 271, 291	0.2	28.309
56	Dasycarpidan-1-methanol, acetate (ester)	326.43	$C_{20}H_{26}N_2O_2$	60, 69, 73, 83, 97, 111, 124, 137, 149, 167, 180, 194, 208, 222, 237, 256, 284, 308, 326	0.74	29.276
57	2-Myristynoyl pantetheine	484.69	$C_{25}H_{44}N_2O_5S$	42, 55, 71, 83, 100, 115, 130, 143, 156, 171, 184, 213, 250, 283, 299	2.28	29.542
58	9-Octadecenoic acid, (2-phenyl-1,3-dioxolan-4-yl)methyl ester, cis-	444.64	$C_{28}H_{44}O_4$	45, 51, 55, 61, 73, 77, 83, 91, 105, 122, 135, 149, 162, 179, 197, 221, 239, 264, 283, 296, 309, 325, 338, 353	0.61	29.659
59	3-hydroxy-Dodecanoic acid-	216.32	$C_{12}H_{24}O_3$	43, 55, 69, 83, 96, 112, 138, 151, 180, 200	1.77	29.818
60	Cyclopropane butanoic acid, 2-[[2-[[2-[(2-pentylcyclopropyl)methyl]cyclopropyl]methyl]cyclopropyl]methyl]-, methyl ester	374.60	$C_{25}H_{42}O_2$	41, 55, 67, 74, 81, 95, 121, 135, 161, 199, 227, 270, 298, 334	0.44	30.721
61	Palmitic acid	256.42	$C_{16}H_{32}O_2$	43, 60, 73, 83, 97, 115, 129, 143, 157, 171, 185, 199, 213, 227, 239, 256	5.38	31.678
62	Dihydroagathic acid	336.46	$C_{20}H_{32}O_4$	41, 55, 67, 81, 95, 109, 121, 139, 167, 175, 189, 221, 235, 257, 275, 290, 300, 318, 336	0.19	32.539

No.	Compounds	Mw	Formula	Negative Ion Mode ([M-H]-) at m/z	%	Rt (min)
63	Ethanone, 1,1'-(5-hydroxy-2,2-dimeth-ylbicyclo[4.1.0]heptane-1,7-diyl)bis-, (1α,5β,6α,7α)-	224.30	$C_{13}H_{20}O_3$	43, 55, 67, 81, 95, 108, 163, 181, 191, 209, 224	0.11	32.837
64	Hexadecanoic acid, 1-(hydroxymethyl)-1,2-ethanediyl ester	568.91	$C_{35}H_{68}O_5$	43, 57, 73, 83, 98, 129, 185, 213, 239, 256, 269, 313, 331, 354, 401, 423	0.19	33.411
65	Linoleic acid ethyl ester	308.50	$C_{20}H_{36}O_2$	41, 55, 67, 81, 95, 109, 123, 150, 178, 220, 263, 279, 308	0.13	33.910
66	Phytol	296.53	$C_{20}H_{40}O$	43, 57, 71, 81, 95, 111, 123, 137, 151, 165, 179, 196, 208, 221, 235, 249, 263, 278, 296	1	34.271
67	Cyclopropanedodecanoic acid, 2-octyl-, methyl ester	366.62	$C_{24}H_{46}O_2$	41, 55, 69, 83, 97, 118, 132, 166, 194, 266, 292, 334, 366	0.08	34.495
68	9,12-Octadecadienoic acid (Z,Z)-	280.44	$C_{18}H_{32}O_2$	41, 55, 67, 81, 95, 109, 123, 182, 222, 237, 256, 280	3.2	34.909
69	9,12,15-Octadecatrienoic acid, 2,3-dihy-droxypropyl ester, (Z,Z,Z)-	352.51	$C_{21}H_{36}O_4$	41, 57, 67, 79, 95, 109, 121, 155, 163, 191, 232, 261, 277, 296, 321, 352	3.27	35.005
70	Octadecanoic acid	284.48	$C_{18}H_{36}O_2$	43, 60, 73, 83, 97, 115, 129, 143, 171, 185, 199, 227, 241, 255, 284	1.37	35.324
71	9-Octadecenoic acid, (2-phenyl-1,3-dioxo-lan-4-yl)methyl ester, cis-	444.64	$C_{28}H_{44}O_4$	45, 55, 73, 83, 91, 105, 122, 149, 179, 197, 221, 264, 283, 305, 338, 367	0.09	37.237
72	Eicosanoic acid	312.53	$C_{20}H_{40}O_2$	43, 57, 73, 85, 97, 111, 129, 185, 213, 241, 269, 312	0.97	39.905
73	Butanoic acid, 1a,2,5,5a,6,9,10,10a-octa-hydro-5,5a-dihydroxy-4-(hydroxymethyl)-1,1,7,9-tetramethyl-11-oxo-1	418.52	$C_{24}H_{34}O_6$	43, 55, 71, 91, 107, 122, 135, 151, 177, 213, 241, 269, 284, 299, 312, 330, 348, 369	0.61	40.585
74	Ethyl iso-allocholate	436.62	$C_{26}H_{44}O_5$	43, 55, 69, 81, 95, 107, 121, 145, 159, 171, 185, 197, 213, 225, 237, 253, 272, 289, 313, 339	3.33	41.285
75	2,4,6-Decatrienoic acid, 1a,2,5,5a,6,9,10,10a-octahydro-5,5a-dihydroxy-4-(hydroxymethyl)-1,1,7,9-tetramethyl	496.63	$C_{30}H_{40}O_6$	41, 55, 67, 79, 91, 122, 135, 149, 161, 189, 213, 241, 284, 312, 330, 347, 368, 400, 432, 465	0.45	44.018
76	Dibenz[a,c]cyclohexane, 2,4,7-trimethoxy-	284.35	$C_{18}H_{20}O_3$	43, 57, 71, 85, 97, 127, 165, 211, 241, 284	3.6	47.058
77	Dodecanoic acid, 1a,2,5,5a,6,9,10,10a-octahydro-5a-hydroxy-4-(hydroxymethyl)-1,1,7,9-tetramethyl-6,11-dioxo	530.73	$C_{32}H_{50}O_6$	43, 57, 69, 79, 93, 107, 122, 135, 177, 241, 284, 312, 330, 361, 385, 437, 461, 512	0.81	47.855
78	Octanoic acid, 1a,2,5,5a,6,9,10,10a-octa-hydro-5,5a-dihydroxy-4-(hydroxymethyl)-1,1,7,9-tetramethyl-11-oxo-1	474.63	$C_{28}H_{42}O_6$	43, 57, 69, 93, 107, 122, 135, 162, 189, 213, 241, 269, 312, 330, 348, 365, 405, 443	0.33	48.152
79	4aα,4bβ-Gibbane-1α,10β-dicarboxylic acid, 4a-formyl-7-hydroxy-1-methyl-8-methy-lene-, dimethyl ester	390.47	$C_{22}H_{30}O_6$	41, 55, 69, 79, 95, 121, 135, 159, 173, 199, 225, 241, 253, 270, 302, 330, 334, 358, 390	0.15	48.546

amides [26]. The weak bands at 2924.25 and 2923.15 cm^{-1} attributed to the stretching vibration of $–CH_3$ and $–CH_2$ groups in alkanes, which indicates the presence of some alkane compounds, including chlorophyll groups [27] in medicinal plants [28]. The band at 2119 cm^{-1} was assigned to the cyanide -CN and alkyne groups [29]. The strong bands around 1664.05 and 1663.81 cm^{-1} are due to the amide, aldehyde, and ketone regions that are characteristic of proteins, enzymes, and sugars. The strong bands at 1616.37 and 1611.69 cm^{-1} showed alkene C=C stretch. The band at 1412.44 and 1407.88 cm^{-1} showed CH_2 and CH_3 bending vibrations.

The weak bands at 1289.81 and 1286.3 cm^{-1} represent the stretching vibrations of C-O due to the presence of alcohol and ether. Also, the C-N (aryl) stretching was assigned to peaks observed at 1289.81 and 1286.3 cm^{-1}. The strong 1053.67 and 1046.43 cm^{-1} bands predict the presence of amines glycoside/C–OH bands in the polysaccharide/protein structures. The presence of Si-O- and aromatic CH bonds has been detected by means of the low-intensity band located at 920 cm^{-1} [30]. The IR spectra of methanolic root extract exhibited C-X (X=F, Cl, S) locations in the spectral region of 866.56-815 cm^{-1} [31]. The FTIR analysis of methanolic root and shoot extracts revealed the presence of functional groups in the structure of biologically active compounds.

Conclusions

In conclusion, the presence of different biologically active components in S. orientalis methanolic extracts indicates its great potential for future pharmaceutical investigations.

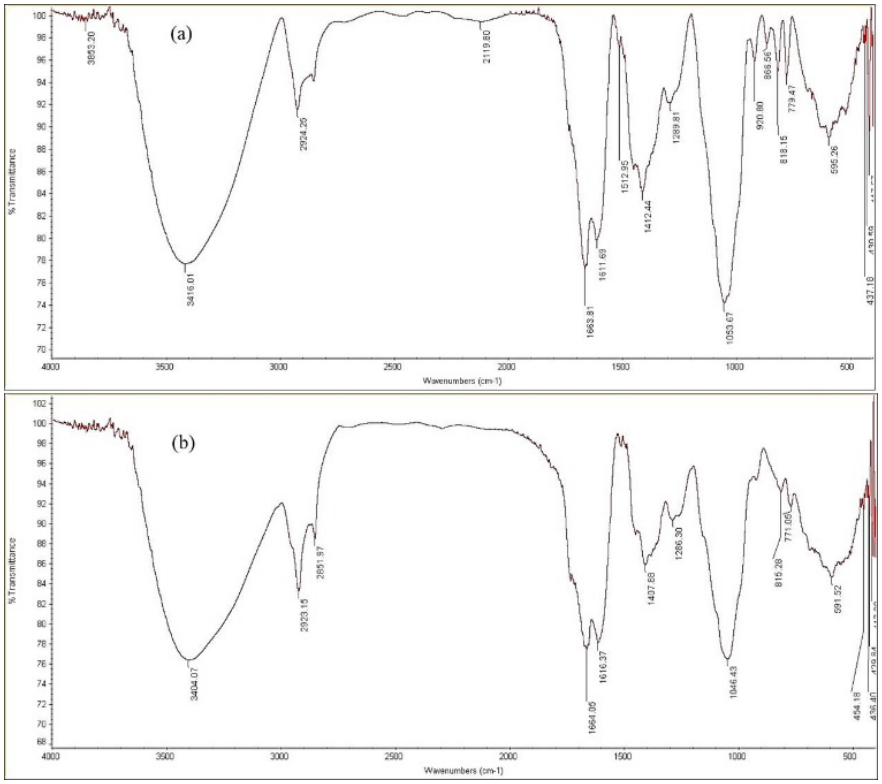

Figure 4. The FTIR spectrum of the *S. orientalis* roots (a) and shoots (b) of methanolic extracts

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Table 3. FTIR peak values of *S. orientalis* methanolic root extract

No.	Peak (Wave Number cm^{-1})	Intensity	Bond	Functional Group Assignment	Group Frequency
1	3416	77.59	N-H, O-H	Normal polymeric O-H stretch	3200-3400
2	2924	91.49	C-H	Aliphatic Alkanes	2850-3000
3	2119	99.40	C≡C, C≡N	Alkyne, Cyanide	2100-2250
4	1663	77.10	RC(O)NHR'	Amide	1640-1670
5	1611	79.73	C=C	Alkenes	1600-1680
6	1412	84.1	-	Ammonium ions	1390-1430
7	1289	92.09	C-O	Alcohols, Ethers, Carboxylic acids, Esters	1000-1300
8	1053	74.10	C-N	Amines	1000-1350
9	920	95.81	C-H	Alkenes	675-995
10	866	97.44	C-H	Alkenes	675-995
11	816	95.07	C-H	Alkenes	675-995
12	779	99.43	C-H	Alkenes	675-995
13	595	88.97	Unknown	-	-

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Table 4. FTIR peak values of *S. orientalis* methanolic shoot extract

No.	Peak (wave number cm^{-1})	Intensity	Bond	Functional Group Assignment	Group Frequency
1	3404	76.31	OH, NH_2	Normal polymeric O-H stretch	3200-3400
2	2923	83.19	C-H	Aliphatic Alkenes	2850-3000
3	2851	88.44	C-H	Aromatic Alkenes	2850-3000
4	1664	77.36	RC(O)NHR′	Amide	1640-1670
5	1616	77.9	C=C	Alkenes	1600-1680
6	1407	85.81	-	Ammonium ions	1390-1430
7	1286	90.3	C-O	Alcohols, Ethers, Carboxylic acids, Esters	1000-1300
8	1046	76.41	C-N	Amines	1000-1350
9	815	93.13	C-H	Alkenes	675-995
10	771	91.67	C-H	Alkenes	675-995
11	591	84.54	Unknown	-	-

Ethical Considerations

Compliance with ethical guidelines

There were no ethical considerations to be considered in this research.

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Authors' contributions

Conceptualization and Supervision: Khadijeh Bagheri and Ali Sharafi; Methodology: Zahra Gharari; Investigation, Writing – original draft, and Writing – review & editing: All authors; Data collection: Zahra Gharari; Data analysis: Zahra Gharari; Funding acquisition and Resources: Khadijeh Bagheri, Ali Sharafi.

Conflict of interest

The authors declared no conflict of interest.

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PBR

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