

Chemical Composition and Antibacterial Properties Survey of *Salvia mirzayanii* Essential Oils in Different Ecological Conditions

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Abstract: Chemical composition and antimicrobial effects of essential oils derived from *Salvia mirzayanii* were determined using GC and GC/MS. In this research four habitats in Larestan region were determined (A-D). The results showed that ecological factors had significant effect on oil yields of *S. mirzayanii*. The components were various in different regions. Linalyl acetate (18.64%), 5-neo-cedranol (17.43%), α -Terpinyl acetate (10.81%), 7-epi- α -selinene (9.18%), Bicyclogermacrene (5.33), Linalool (4.77%), 1,8-Cineol (4.53%) were the main components. The antimicrobial activity of essential oil of *S. mirzayanii* was studied against three bacteria (*Shigella dysenteriae*, *Staphylococcus aureus* and *Escherichia coli*). Antimicrobial activities according to the disk diffusion method and MIC values were performed and showed that essential oils of *S. mirzayanii* revealed antimicrobial activity against bacteria in all regions. In all microorganisms MIC value was 0.625 μ l/ml and inhibition zones ranged from 8 to 17.67 mm. The maximum and minimum antimicrobial activity was observed on *Staphylococcus aureus* and *Escherichia coli*, respectively.

Keywords: Antimicrobial activity, essential oil, *Salvia mirzayanii*.

1. INTRODUCTION

Salvia is an important genus from the Lamiaceae with approximately 900 species and represented in Flora iranica by 58 species (1). *Salvia mirzayanii* Rech. f. & Esfand. ("Moor-e-Talkh" in local language) is one of the native species. This plant distributed in central and southern parts of Iran. *Salvia mirzayanii* is growing as a biennial or perennial flowering plants (2). Local people usually used *Salvia mirzayanii* for colds, hemostatic and infections (3). This species used as an astringent and tonic in Iranian folk medicine (4). In addition, several studies have shown the various biological activities of this plant including antiplasmodial, antidiabetic, antioxidant, antifungal, antibacterial activity, anti-HIV, antimalarial, cytotoxicity, antitumor and cardiotoxic properties (5).

The pharmacological effects of *Salvia* essential oils are based on the presence of more than 100 active elements which can be classified into Monoterpene hydrocarbons, Oxygenated monoterpenes, Sesquiterpene hydrocarbons, Diterpenes, Not iso-prenoid compounds and Oxygenated sesquiterpenes. The major components contain 1, 8-cineole, camphor, borneol, β -pinene, α -pinene, camphene and α -thujene (6). By Javidnia et al. (7) spathulenol, δ -cadinene, linalool, α -terpinyl acetate, α -cadinol, β -eudesmol, cubenol and linalyl acetate were reported to be the main components of *S. mirzayanii* essential oil. In another study, Yamini et al. (8) reported that, linalyl acetate, 1,8-cineole, linalool and 8-acetoxy linalool were the major components in *S. mirzayanii* essential oil.

The essential oils of extracted from *S. mirzayanii* have recently been investigated, showing strong antimicrobial activity. Sonboli et al. (9) demonstrated that *S. mirzayanii* essential oils exhibited antimicrobial activity. Results of other study showed *S. mirzayanii* was rich in 1,8-cineol and exhibited strong antimicrobial activity against tested microbes, in this study demonstrated that 1,8-cineol has significant antimicrobial activities (10). Haghghi et al. (11) reported that the major components in *S. mirzayanii* oil were 5-neocedranol, α -terpinyl acetate, 1,8-cineol, bicyclogermacrene, δ -cadinene, Globulol, α -cadinol, tau-cadinol, 7-epi- α -selinene, Linalyl acetate, Linalool, β -Elemene, γ -cadinene and α -guaiene and the oil showed good antimicrobial activity against *Fusarium solani*, *Staphylococcus aureus* and *Candida albicans*.

In the current study, we investigated the chemical composition of the essential oils from the aerial parts of *Salvia mirzayanii*. Furthermore, antimicrobial activity of the essential oils obtained from this plant was evaluated against *Sh. dysenteriae*, *E. coli O157* and *S. aureus*.

2. MATERIAL AND METHODS

2.1. Plant Material

The aerial parts of *S. mirzayanii* were collected in June 2012 at flowering stage from four habitats in Larestan region: (A) center of Lar, Sahraye nimeh, (B) north of Lar, Dehkuieh, (C) south of Lar, Kuh perdi, (D) northeast of Lar, Dresosaiban. Features of this region and soil properties listed in table 1 and 2. Plant materials were identified in the Herbarium of Medicinal and Aromatic Plants, Islamic Azad University, Jahrom, Iran. Harvested plants were dried at temperature below 30 °C for 15 days.

Table1. Collection site information (Meteorological Center, Larestan, Iran)

Site	Latitude	Longitude	Elevation (M)	Average Of Relative Humidity (%)	Total Annual Evaporation (Mm)	Mean Annual Rainfall (Mm)	Max Temperature (°C)	Min Temperature (°C)
A	54° 23 01.94	27° 33 34.30	1115	42	3321.5	203	47.8	-4.8
B	54° 25 39.18	27° 56 09.17"	1315	47	3248.5	211.2	45	-7
C	54° 16 44.27"	27° 28 09.32	2100	50	3102	223	35	-12
D	55° 33 09.45"	27° 53 29.06	987	37	4015	185	50	-2

2.2. Essential Oil Extraction

Essential oil was extracted from dried and powdered aerial parts (100 g) of *S.mirzayanii* by hydro-distillation using glass Clevenger type apparatus during approximately 3 hours. The distilled essential oil was dried using anhydrous sodium sulfate. The essential oil was weighted and stored in refrigerator at 4°C until analysis (12).

2.3. Essential Oil Analysis

Gas chromatographic analysis was performed using an Agilent Technologies gas chromatographer (model 6890A, USA) equipped with a flame ionization detector (FID) and a DB-5 fused silica capillary column (30 m × 0.32 mm, film thickness 0.25 µm). The oven temperature was 140°C. Helium was used as the carrier gas. The samples were injected using split sampling technique by a ratio of 1.50. GC-MS analysis was carried out using an Agilent Technologies GC-MS (model 6890 C) equipped with fused silica capillary DB-5 column (30 m × 0.25 mm, 0.25 µm film thickness) with Helium as the carrier gas and a split ratio of 1.50(13).

Table2. Physico- chemical properties of the experimental soil (Soil Science Laboratory, Shiraz, Iran)

Region	Soil properties													
	Acidity (pH)		Electrical conductivity (EC)		Humidity (%)		Phosphorous		Potassium (K)		Carbon (OC)		Nitrogen (N)	
	mean	F-value	mean	F-value	mean	F-value	mean	F-value	mean	F-value	mean	F-value	mean	mean
A	7.61	0.02 ^{ns}	1.42	52.37 ^{**}	33.3	75.24 ^{**}	7.72	579 ^{**}	154	249 ^{**}	1.32	43 ^{**}	0.12	9.06 ^{**}
B	7.63		1.08		43		0.72		160		0.93		0.09	
C	7.57		1.55		38.33		8.67		243		0.98		0.1	
D	7.52		1.55		57.33		26.33		232		2.51		0.25	

Region	Soil properties							
	Sand		Silt		Clay		TNV	
	F-value	F-value	mean	F-value	mean	F-value	mean	F-value
A	75	265 ^{**}	25	169 ^{**}	0.01	104 ^{**}	16.6	310 ^{**}
B	51		41		7.97		21.42	
C	39		53		7.87		39.43	
D	47		47		6.07		52.43	

*, ** = Significant at 5 % and 1%, respectively, ns= Non-significant

2.4. Microorganisms

The antimicrobial activities of essential oils were tested against standard strain of Gram-positive bacteria *Staphylococcus aureus* (ATCC 6538), *Shigella dysenteriae* (RI366), *Escherichia coli* (O157:H7 EPEC (M). This strain was obtained from Persian Type Culture Collection (PTCC) in Iranian Research Organization for Science and Technology.

2.5. Antimicrobial Activity Survey

Antimicrobial activity of essential oil of *S. mirzayanii* was evaluated by using disc diffusion method, according to the National Committee for Clinical Laboratory Standards (14). The inoculants of the microbial strains were prepared from freshly cultured bacteria that were adjusted to 0.5 McFarland standard turbidity (15). Tested bacteria strains were suspended in the agar media and the plates (treated wells and untreated controls) were incubated in a humid atmosphere at 30°C for 24 hours (16). The essential oil was dissolved in dimethyl sulfoxide (DMSO), and diluted in a twofold manner to make the concentrations of 0.625, 1.25, 2.5, 5, 10 and 20 µl/disc. These plates were incubated for 24 hours at 35- 37 °C. After the incubation period, the diameter of inhibition zone (IZ) was measured in millimeters. The minimum inhibitory concentration (MIC) of the essential oil was determined through a serial dilution method. MICs is visually determined and defined as the lowest concentration of the essential oil producing no visible growth. Each experiment was performed in triplicate.

Analysis of variance was performed by ANOVA by the software SAS. Significant differences between means were determined by Duncan's new multiple-range test. Correlations among data were calculated using Pearson's correlation coefficient.

3. RESULTS AND DISCUSSION

3.1. Chemical Composition of the Essential Oil

The essential oil of the aerial parts of *S. santolinifolia* was obtained as a light yellow liquid with a yield of 1.6% (w/w), based on dry weight. The results showed that ecological factors had significant effect on oil yields of *S. mirzayanii* (Table 3). Identification of chemical constituents of the essential oils showed that the components were various in different regions (Table 4). GC/FID and GC/MS analysis identified forty-six and forty-three components in the essential oil of *S. mirzayanii* that represented 95.26% and 94.94% of the oils in B region (max) and A region (min), respectively. Also, forty-four and forty-five components were identified in the essential oil of *S. mirzayanii* in C region and D region, respectively.

Table3. Variance analysis of *S. mirzayanii* essential weight and yield

S.O.V	Df	Mean Squares	
		Essential Weight	Essential Yield
Treatment	3	0.367**	0.06**
Experimental error	8	0.002**	0.003**
Coefficient of variation (%)	-	5.45	7.79

*,** and ns is significant at probability level of 1 and non-significant at 5 %, respectively.

Table4. Essential weight and yield of *S. mirzayanii* in different region

Region	Essential weight	Essential yield
A	0.51c	0.78b
B	0.68c	1.23c
C	1.3a	1.33a
D	0.71b	0.69b

In a column, means with the same letters are not significantly different

The major components were 5- neo-cedranol (17.43%), Linalyl acetate (15.89%), 7-epi- α -selinene (9.18%), Spatolenol (7.46%), α -terpinyl acetate (4.66%), bicyclogermacrene (4.59%), Linalool (4.37%), Germacrene – d- 4- ol (4.30%) (A region), Linalyl acetate (15.89%), 5- neo-cedranol (11.42%), α -terpinyl acetate (10.81%), Spatolenol (5.81%), Linalool (4.77%), bicyclogermacrene (4.59%), 1,8-cineol (4.53%), 7-epi- α -selinene (4.17) (B region), Linalyl acetate (13.10%), 5- neo-cedranol (12.83%), Spatolenol (8.18%), bicyclogermacrene (5.33%), 7-epi- α -selinene (4.88%), α -Terpineol (4.71%), α -cadinol (4.36%) (C region) and Linalyl acetate (18.64%), 5- neo-cedranol

(9.75%), Spatolenol (8.68%), 7-epi- α -selinene (7.28%), Linalool (6.17%), α -terpinyl acetate (6.16%) (Dregion). Other components were present in amounts less than 4% (Table 5-8). The main components observed in the essential oil of *S. mirzayanii* in each region were different. The peaks in the chromatogram showed major components in essential oil in each region (Figure 1-4).

Table5. Chemical composition of the essential oil in A region

No	Compounds	RI	Oil percentage
1	a-Thujene	926.4	0.029
2	a-Pinene	933.7	0.090
3	Sabinene	973.7	0.039
4	b-Pinene	978.2	0.137
5	Myrcene	990.5	0.462
6	n-octanal	996.4	0.740
7	Limonene	1029	0.202
8	1,8-Cineol	1036	2.312
9	trans-b-Ocimene	1046	0.297
10	trans-linalool oxide	1078	0.438
11	Terpinolene	1089	0.141
12	Linalool	1105	4.373
13	trans-Pinocarveol	1130	0.105
14	trans-verbenol	1158	0.492
15	P-mentha-1,5-dien-8-ol	1172	0.328
16	Terpinene-4-ol	1182	0.206
17	a-Terpineol	1197	1.731
18	trans-Carveol	1212	0.341
19	Nerol	1233	0.355
20	Linalyl acetate	1261	9.114
21	n-Decanol	1275	0.483
22	d-Elemene	1338	0.693
23	a-Terpinyl acetate	1355	4.661
24	Neryl acetate	1368	0.461
25	Geranyl acetate	1388	1.003
26	b-Elemene	1394	1.446
27	a-Gurjunene	1412	0.846
28	(E)-Caryophyllene	1422	0.607
29	a-Guaiene	1441	0.938
30	g-Muurolene	1478	0.877
31	b-Selinene	1489	0.884
32	Bicyclogermacrene	1501	6.848
33	a-Muurolene	1517	1.436
34	7-epi-a-selinene	1528	9.189
35	d-Cadinene	1533	2.697
36	Germacrene D-4-ol	1583	4.305
37	Spathulenol	1588	7.460
38	Globulol	1592	0.783
39	epi-a-Cadinol	1647	2.119
40	epi-a-Muurolol	1649	1.440
41	b-Eudesmol	1659	2.581
42	a-Cadinol	1664	4.137
43	5-neo-cedranol	1705	17.437
	Oil Yield (%w/w)		0.78
	Total		95.263

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Table6. Chemical composition of the essential oil in **B** region

No	Compounds	RI	Oil percentage
2	a-Pinene	933.9	0.141
3	Sabinene	973.9	0.141
4	b-Pinene	978.4	0.239
5	Myrcene	990.5	0.605
6	n-octanal	996.8	1.211
7	a-terpinene	1016	0.166
8	<i>p</i> -Cymene	1025	0.142
9	Limonene	1029	0.317
10	1,8-Cineol	1037	4.534
11	trans-b-Ocimene	1047	0.320
12	trans-linalool oxide	1079	0.689
13	Terpinolene	1089	0.180
14	Linalool	1105	4.770
15	trans-Pinocarveol	1144	0.201
16	trans-verbenol	1159	0.527
17	P-mentha-1,5-dien-8-ol	1172	0.648
18	Terpinene-4-ol	1182	0.285
19	a-Terpineol	1197	2.274
20	trans-Carveol	1213	0.422
21	Nerol	1234	0.433
22	Linalyl acetate	1263	15.891
23	n-Decanol	1276	0.730
24	d-Elemene	1338	0.334
25	a-Terpinyl acetate	1358	10.817
26	Neryl acetate	1368	0.514
27	Geranyl acetate	1388	1.003
28	b-Elemene	1394	0.937
29	a-Gurjunene	1412	0.513
30	(E)-Caryophyllene	1422	0.506
31	a-Guaiene	1444	1.355
32	allo-Aromadendrene	1461	0.331
33	g-Muurolene	1478	0.702
34	b-Selinene	1489	0.556
35	Bicyclogermacrene	1501	4.595
36	a-Muurolene	1517	0.905
37	g-Cadinene	1525	2.110
38	7-epi-a-selinene	1528	4.178
39	d-Cadinene	1532	1.151
40	Germacrene D-4-ol	1583	2.730
41	Spathulenol	1588	5.810
42	Globulol	1592	0.414
43	epi-a-Muurolol	1649	2.866
44	b-Eudesmol	1659	2.384
45	a-Cadinol	1664	3.892
46	5-neo-cedranol	1704	11.421
	Oil Yield (%w/w)		0.052
	Total		94.942

Table7. Chemical composition of the essential oil in C region

No	Compounds	RI	Oil percentage
1	a-Thujene	926.4	0.036
2	a-Pinene	933.8	0.180
3	Sabinene	973.8	0.069
4	b-Pinene	978.4	0.315
5	Myrcene	990.2	0.477
6	n-octanal	996.4	0.939
7	Limonene	1029	0.340
8	1,8-Cineol	1036	1.681
9	trans-b-Ocimene	1046	0.302
10	trans-linalool oxide	1078	0.529
11	Linalool	1105	3.955
12	trans-Pinocarveol	1130	0.187
13	trans-verbenol	1158	0.752
14	P-mentha-1,5-dien-8-ol	1172	0.389
15	a-Terpineol	1197	1.737
16	trans-Carveol	1213	0.489
17	Nerol	1234	0.384
18	Linalyl acetate	1262	13.104
19	n-Decanol	1275	0.573
20	d-Elemene	1338	0.537
21	a-Terpinyl acetate	1355	4.714
22	Neryl acetate	1368	0.472
23	a-Copaene	1381	0.401
24	Geranyl acetate	1387	1.013
25	b-Elemene	1394	1.208
26	a-Gurjunene	1412	0.666
27	(E)-Caryophyllene	1421	0.623
28	a-Guaiene	1444	2.201
29	allo-Aromadendrene	1461	0.674
30	g-Muurolene	1478	0.918
31	b-Selinene	1489	0.748
32	Bicyclogermacrene	1500	5.334
33	a-Muurolene	1517	1.238
34	g-Cadinene	1525	3.415
35	7-epi-a-selinene	1528	4.881
36	d-Cadinene	1532	2.039
37	Germacrene D-4-ol	1583	3.258
38	Spathulenol	1588	8.180
39	Globulol	1592	0.674
40	epi-a-Cadinol	1647	1.826
41	epi-a-Muurolol	1649	1.640
42	b-Eudesmol	1659	3.579
43	a-Cadinol	1663	4.362
44	5-neo-cedranol	1703	12.835
	Oil Yield (%w/w)		0.690
	Total		93.874

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Table 8. Chemical composition of the essential oil in D region

No	Compounds	RI	Oil percentage
1	a-Thujene	926.3	0.070
2	a-Pinene	933.8	0.189
3	Sabinene	973.7	0.105
4	b-Pinene	978.2	0.314
5	Myrcene	990.3	0.817
6	n-octanal	996.4	0.785
7	p-Cymene	1025	0.143
8	Limonene	1029	0.349
9	1,8-Cineol	1036	2.637
10	trans-b-Ocimene	1046	0.458
11	g-Terpinene	1058	0.069
12	trans-linalool oxide	1078	0.748
13	Terpinolene	1089	0.187
14	Linalool	1106	6.178
15	trans-Pinocarveol	1130	0.172
16	trans-verbenol	1158	0.671
17	P-mentha-1,5-dien-8-ol	1172	0.349
18	Terpinene-4-ol	1182	0.163
19	a-Terpineol	1197	2.227
20	trans-Carveol	1213	0.424
21	Nerol	1234	0.466
22	Linalyl acetate	1263	18.643
23	n-Decanol	1276	0.672
24	d-Elemene	1338	0.494
25	a-Terpinyl acetate	1356	6.166
26	Neryl acetate	1368	0.699
27	a-Copaene	1382	0.560
28	Geranyl acetate	1388	1.389
29	b-Elemene	1394	0.987
30	a-Gurjunene	1412	0.563
31	(E)-Caryophyllene	1422	0.577
32	a-Guaiene	1444	2.385
33	allo-Aromadendrene	1461	0.696
34	g-Muurolene	1478	0.767
35	b-Selinene	1489	0.652
36	Bicyclogermacrene	1501	4.888
37	a-Muurolene	1517	1.210
38	7-epi-a-selinene	1528	7.282
39	d-Cadinene	1532	1.559
40	Spathulenol	1588	8.685
41	Globulol	1592	0.493
42	epi-a-Cadinol	1647	2.950
43	b-Eudesmol	1659	2.233
44	a-Cadinol	1663	3.127
45	5-neo-cedranol	1703	9.759
	Oil Yield (%w/w)		1.310
	Total		94.957

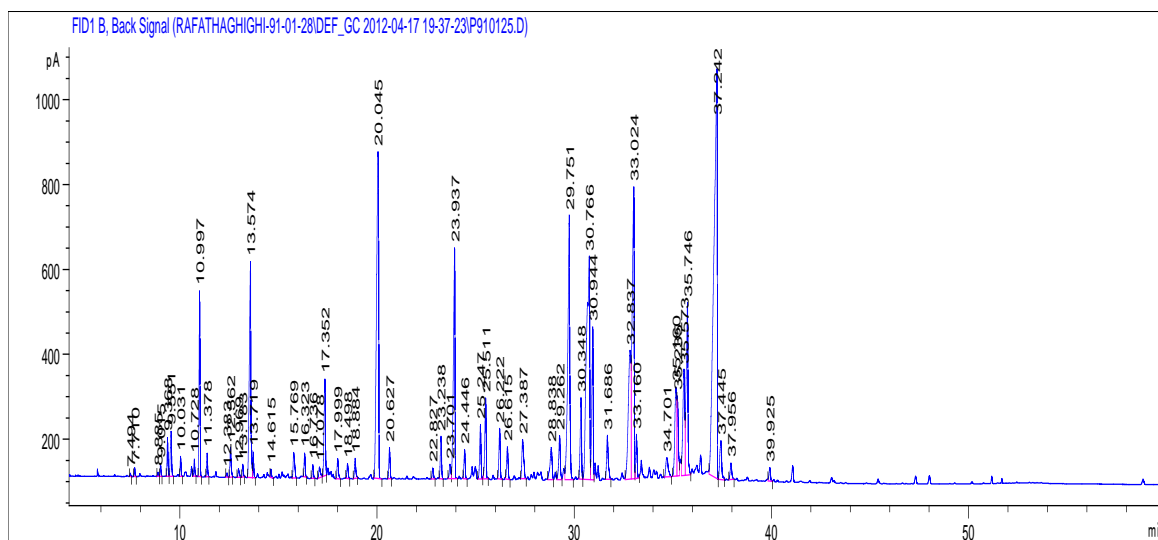


Figure1. GC-MS chromatogram of *S. mirzayanii* essential oil in A region

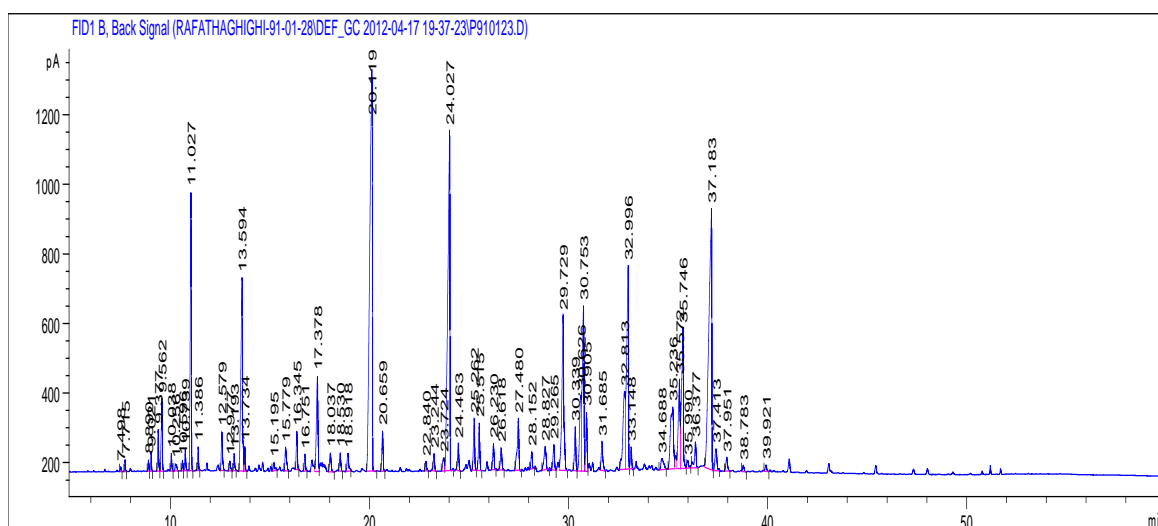


Figure2. GC-MS chromatogram of *S. mirzayanii* essential oil in B region

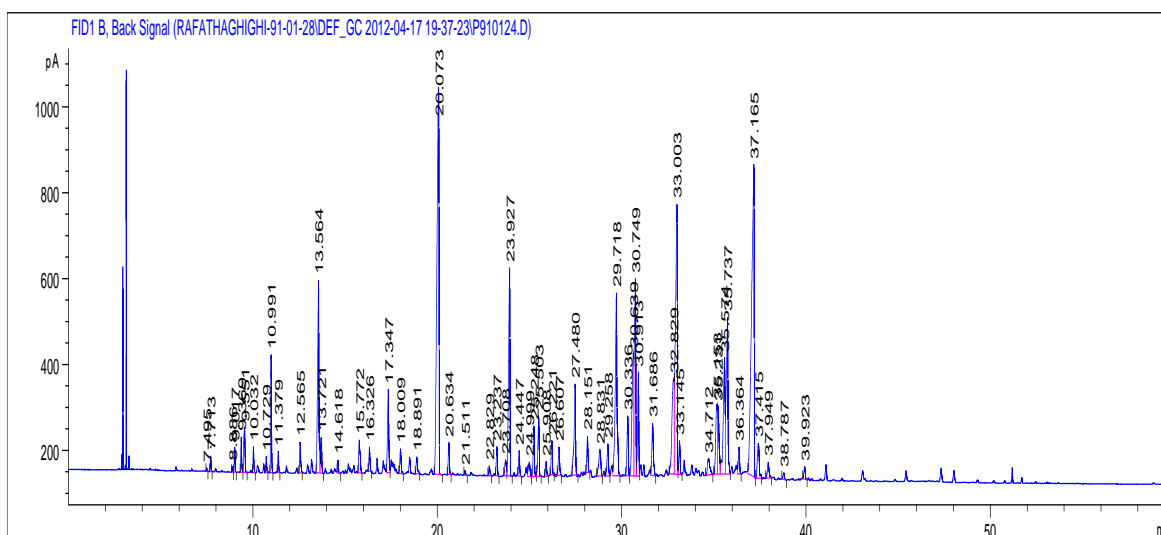


Figure3. GC-MS chromatogram of *S. mirzayanii* essential oil in C region

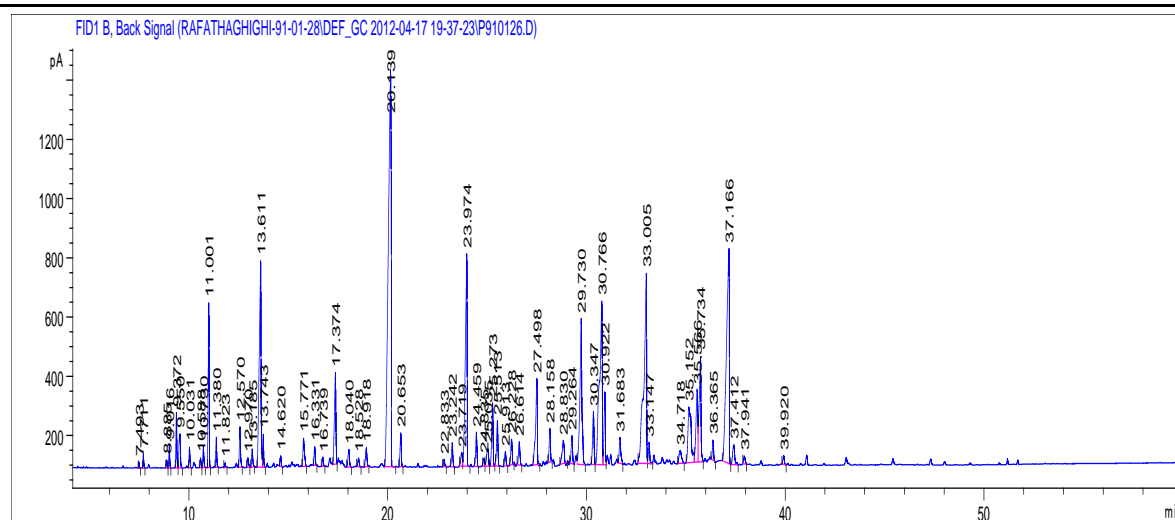


Figure4. GC-MS chromatogram of *S. mirzayanii* essential oil in D region

However, some major components as, linalool, linalyl acetate, 5- neo-cedranol, 7-epi- α -selinene, α -terpinyl acetate, 1,8-cineole, Spathulenol, α -cadinol were common between all. These differences in the essential oil compositions in different regions can be credited to several environmental agents such as climatic conditions, seasonal and geographical or ontogenesis variations(17). There are many reports in the literature showing the variation in the chemical composition of the essential oil with respect to geographical regions (18,19,20, and 21).

According to the results, by increasing of moisture content, silt, clay and lime percentage increased essential oil of *S. mirzayanii*, but essential oils of this plant are reduced by increasing of electrical conductivity, phosphorous, nitrogen and sand percentage. Potassium levels had no effect on the amount of essential oil. Percentage of essential oil increased by increasing elevation, but Linalyl acetate (one of the major constituents of the essential oil) was reduced.

In this study the number of components (forty-six) in *S. mirzayanii* essential oil were higher than previous reports in this plant. In present study, linalyl acetate and 5- neo-cedranol were principle component in *S. mirzayanii*, while Mirza et al. (22) reported that, linalool, linalyl acetate, 1,8-cineol, terpinenyl acetate were the main constituents of the *S. mirzayanii* essential oil. Javidnia et al.(7) reported that spathulenol, δ -cadinene, linalool, α -terpinyl acetate, α -cadinol, β -eudesmol, cubenol and linalyl acetate were the main components of *S. mirzayanii* essential oil. In another study by Yamini et al.(8) linalyl acetate, 1,8-cineole, linalool and 8-acetoxy linalool were reported to be the major components in *S. mirzayanii* essential oil.

3.2. Antimicrobial Activity of Essential Oils

The antimicrobial activity of *S. mirzayanii* essential oils were evaluated against standard strain of *Staphylococcus aureus* (ATCC 6538), *Shigella dysenteriae* (RI366), *Escherichia coli* (O157:H7 EPEC (M) by using disc diffusion method. Our results showed that, *S. mirzayanii* extract has significant antimicrobial properties against these microorganisms. The effectiveness of leaf essential oil is demonstrated by the size of the inhibition zone around the filter paper disk on microbial growth, which is typically expressed as the diameter of the inhibition zone in millimeter (23). This results showed different inhibition zone against *Sh. dysenteriae* (8, 9, 10, 12, 13.33, 15.67 mm), *E. coli* (8.67, 8.67, 10, 11.33, 12.33 and 14.33 mm) and *S. aureus* (8, 10.33, 11.33, 12.67, 14 and 17.67 mm) with different concentrations of *S. mirzayanii* essential oil (0.625, 1.25, 2.5, 5, 10 and 20), respectively.

In this study, *Escherichia coli* was the most resistant microbe and *Shigella dysenteriae* was the most sensitive (Figure 5). In all treatments, inhibitory effect of the essential oil was increased by increasing concentration of essential oil. Minimum inhibitory concentrations (MICs) were determined and the MIC value was 0.625 μ l/ml in all microorganisms. This activity is generally correlated to the chemical composition of the oil (11). These results suggest that antimicrobial activity in *S. mirzayanii* results from the phenolic compounds. The diameters of inhibition zones ranged from 8 to 17.67 mm including the diameter of paper disc (6 mm). The results are summarized in Table 9.

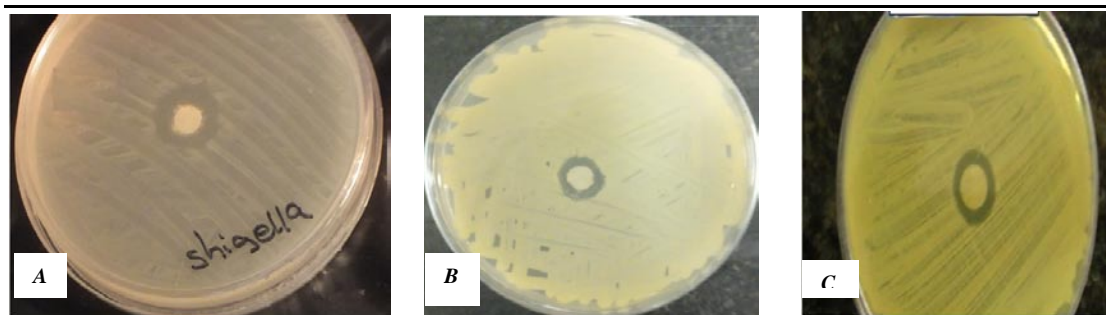


Figure5. Antibacterial activity of *S. mirzayanii* leaf essential oil against 3 selected bacteria (A: *Sh. Dysenteriae*, B: *S. aureus* and C: *E. coli*) by disc diffusion.

Table9. Antibacterial activities of extracts of *S. mirzayanii* against bacterial test organism

Microorganism	zone of inhibition (mm)					
	essential oil of <i>S. mirzayanii</i> (ul/ml)					
	0.625	1.25	2.5	5	10	20
<i>Shigella dysenteriae</i>	8b	9a	10a	12a	13.33a	15.67a
<i>Escherichia coli</i>	8.67b	8.67b	10a	11.33b	12.33b	14.33b
<i>Staphylococcus aureus</i>	8b	10.33	11.33b	12.67a	14c	17.67c

Each value in the table was obtained by calculating the average of three experiments.

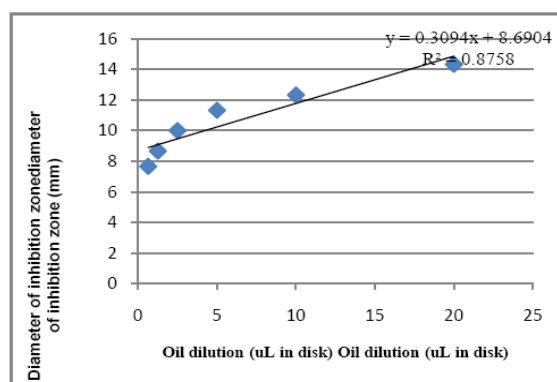
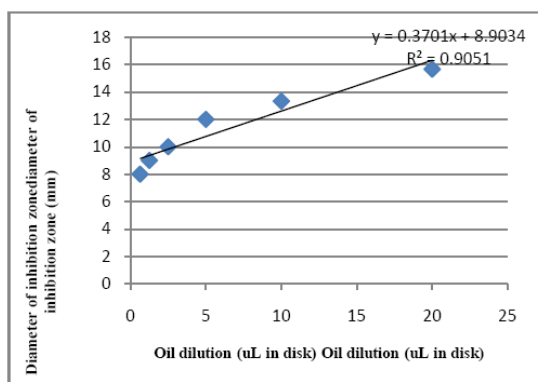
Diameter of inhibition zone including disc diameter of 6mm

Good antimicrobial activity of *S. mirzayanii* extract against experimental bacteria could be attributed to high amount of major components as 5-neo-cedranol, linalool, linalyl acetate, α -terpinyl acetate, 1,8-cineol and other valuable components in oil. Several researches on the antimicrobial activity of the essential oils of many *Salvia* species have been carried out, demonstrating that among the components of the oil, some of them showed strong antimicrobial activity (24). These results suggest that *S. mirzayanii* extract has high antimicrobial activity. This is in line with the observation of other authors who found antimicrobial properties of the essential oils of this plant. The essential oil of *Salvia tomentosa* showed antibacterial activity against eight microorganisms (25).

Yousefzad et al. (26) reported *S. chloroleuca* oil exhibited moderate to high antimicrobial activity, especially for *Bacillus subtilis*, *Staphylococcus epidermidis* and *S. aureus*. Results of Bouaziz et al. (27) confirmed that *S. officinalis* had inhibitive effect on bacterial strains including Gram+ and Gram- of rods and cocci.

S. przewalskii oil also showed an antimicrobial activity against *Staphylococcus aureus* and *S. epidermidis* strains (26). Kabouche et al. (28) demonstrated that essential oil obtained from roots of *Salvia jaminiana* has antibacterial activity. The essential oils of *S. santolinifolia*, *S. hydrangea* and *S. mirzayanii* have the antimicrobial property. Furthermore, the antimicrobial property of *S. mirzayanii* oil was superior to others (9). Results of Bahadori et al. (29) showed that the volatile oil of *S. santolinifolia* could be considered as a rich source of natural agents for several uses as antibiotics against human pathogenic microbes.

The strong positive correlations found between inhibitory concentrations of *S. mirzayanii* essential oil and diameter of inhibition zone of *Sh. dysenteriae* ($r^2 = 0.9051$), *E. coli* ($r^2 = 0.8758$) and *S. aureus* ($r^2 = 0.9081$) confirm the above remark (Figure 6).



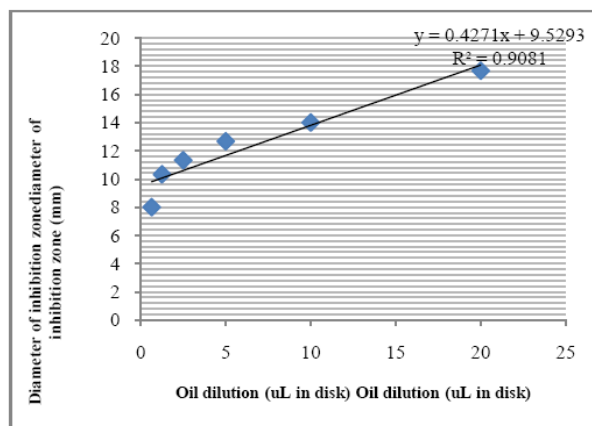


Figure 6. Correlations between inhibitory concentrations of *S. mirzayanii* essential oil and diameter of inhibition zone of *Sh. Dysenteriae* (Top-left), *S. aureus* (Top-right) and *E. coli* (Bottom)

4. CONCLUSION

The findings of this study indicated that *S. myrzayanii* has an antimicrobial essential oil. These essential oils of *S. myrzayanii* can be possibly used as an antimicrobial agent in control of oral pathogens. This oil could be applied for formulation of new natural antibiotics. Differences observed in the chemical composition and oil yield, between different regions may be due to climatic conditions and soil properties. From these results it can be concluded that *S. myrzayanii* cultivation in areas with moist, silt, clay and lime soil and increasing elevation can be enhanced essential oil of *S. myrzayanii*.

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