

Chemical Composition of Essential Oil Extracted from *Urtica pilulifera* and Evaluation Its Biological Activity

Shaza Sitrallah, Joumaa Merza

Abstract— The essential oil has been extracted from the aerial parts of *Urtica pilulifera* from *Urticaceae* family collected in south west of Hama in Syria by hydro-distillation using Clavenger type apparatus. The composition, the qualitative and quantitative analysis of the essential oil was achieved and characterized by means of GC-MS and comparing with the references in the literature. A total of 45 compounds of the essential oil were characterized. The major compounds in the oil was dominated by four compounds representing the majority of the components: (-) Limonene (1.24%), 1,8-Cineole (8.20%), 3-Carene (3.76%), (+) Limonene (6.76%), Terpinene (2.41%), Vanillin (1.70%), Butyl acetate (3.23%), 1,2-Benzene dicarboxylic acid (13.50%), 7-Acetyl-6-ethyl-1,1,4,4-tetramethyltetralin (19.61%). The other compounds were mostly presented in low amounts. The biological effects of aromatic oil were studied on some pathogenic bacteria including: *Staphylococcus aureus*, *Escherichia coli*.

Index Terms— Biological effects, Clavenger, *Escherichia coli*, essential oil, GC-MS, hydro-distillation, *Staphylococcus aureus*, *Urticaceae*, *Urtica pilulifera*.

1 INTRODUCTION

THE *Urticaceae* is a common family of plants which produces allergenic substances causing edema and inflammation in humans [1]. It is composed of over 48 genera and nearly 1300 species [2]. The main varieties identified under the *Urtica* species are *Urtica dioica* L., *U. urens* L., *Urtica pilulifera* L., *U. cannabina* L., *U. membranacea* Poir., *U. kiovensis* Rogoff L. [2]. *Urtica* plant is useful for bladder disorder. *Urtica pilulifera* L. is a member of family *Urticaceae*. It is one of the most effective medicinal plant and widely used in folk remedy to treat hyperglycemia, hypertension and inflammation of some organs such as uvula and uterus, anemia, wound healing, as purifier and as toner tea and other ingredient as anhydrous lanolin and mint oil in preparation of wound-healing antimicrobial ointment [3-6]. Its extract reported as useful for bladder disorder and reduced postoperative blood loss, and prevented hemorrhagic and purulent inflammation following adenomectomy [7]. As a part of our study to characterize the chemical constituents of Syrian plants we have investigated the essential oil of *Urtica pilulifera* L. In the present communication we wish to report the extraction and studying the chemical composition of the essential oil of *Urtica pilulifera* and evaluate its biological activity.

2 TAXONOMIC DESCRIPTION OF EUPHORBIA URTICA PILULIFERA:

Urtica pilulifera Stinging nettle (*Urticaceae*) are annual and perennial herbs, a plant with a square leg, its leaves are large serrated in the heart, its thickness and the leg are thick, thin hairs that carry in its bases a liquid composed of several chemicals, most important of which is histamine and formic acid. These capillaries are opened at their pointed tops once The small nettle flowers are colored in clusters that hang down, the seeds are dark yellow, and the two species that spread in the Arab environment are: the small nettle, up to 50 cm high, the large nettle, rising about one and a half meters and the leaves slightly larger than the nettle, but the two types are similar in their chemical components and therapeutic properties [8]. As for its whereabouts, it is found everywhere in the world, and the types of this plant spread in most of the Arab Mashreq countries. The whole herb, including roots and seeds, is harvested from the beginning of July until the beginning of September, and if only the roots are required to be harvested before flowering [9].

3 EXPERIMENTAL PROCEDURE:

3.1 Plant Material

Aerial parts of *Urtica pilulifera*, were collected and dried in August 2017, from South-Est of Hama, Syria. The plant was authenticated by the Atomic Agent in Syria. A voucher specimen of plant was deposited in the laboratory of chemistry of natural products, Department of chemistry, Faculty of sciences, AL Baath University, Homs, Syria.

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3.2 Essential oils analysis

The analysis of the essential oil was performed with Shimadzu Bruker Ultra Shield 400MHz gas chromatograph with a capillary column DB5 (30m × 0.25µm) With an internal character (0.25µm). Temperature program was as follows: 3 min at 40°C, increased to 100°C at a rate of 5°C min, then, increased to 120°C at a rate of 5°C min and held at that temperature for 1 min, increased to 180°C at a rate of 6°C min, increased to 200°C at a rate of 20°C min, increased to 220°C at a rate of 30°C min, then increased to 280°C at a rate of 40°C min and held at that temperature for 1 min. Injection temperature was 230°C. Injection volume was 1.0 µL. Helium was used as a carrier gas (1 mL/min). the identification of the constituents was performed by comparing the spectra obtained with database of Wiley Spectral Library Collection and NSIT library database. Quantitative data were obtained from the electronic integration of the FID peak areas.

3.3 Extraction the essential oil

The extraction of essential oil was carried out by hydro-distillation using Clavenger type apparatus. 200 gr of *Urtica pilulifera* was boiled in water during 4 hours and the yield of essential oil was 1.42% (w/w). The essential oils obtained has brown pale color with characteristic odor. The oils were stored in a refrigerator until the analysis by GC-MS.

3.4 Evaluation the biological activity

The biological efficacy of the aromatic oil extracted from the *Urtica pilulifera* was studied in a manner of spreading the tablets to two bacteriostatic strains: *Staphylococcus aureus*, *Escherichia coli*. The lobster was highly effective on bacterium Effectiveness has been compared with gentamicin anti-inflammatory drug. Transfer (0.1) cm³ from the diluted bacterial suspension to the center of nutritious Nutrient agar and spread on the surface of the center in a homogeneous manner and incubated for 30 minutes at a temperature of 37 °C for the purpose of sowing. In the meantime, the tablets were filled with oil extract and active ingredients. The discs were prepared from the filter paper with a perforation of the leaves and a diameter of 5 mm. These tablets were treated with different concentrations of the oil extract (100%, 50%, 25%). The steroid tablets containing the nutrient medium are then sterilized with sterile concentrates. At this time, the Gentamicin filter paper is coated with a concentration of 500 µg/cm³. It is determined by the different concentrations of the oil extract and DMSO, all in one dish on the feeding medium and incubated at a temperature of (37) °C for a period of (16) hours.

4 RESULTS AND DISCUSSION

4.1 Volatile Chemical Composition

The figure 2 presents a typical chromatographic profile of essential oil from *Urtica pilulifera* by hydrodistillation (HD) us-

ing Clavenger type apparatus. The individual components of the oils were identified by GC/Mass, The Peaks identification and relative amounts of the various compounds present in the volatile fraction appear in Table 1. A total of 45 compounds. The oil was dominated by some compounds representing the majority of the components: (-) Limonene (1.24%), 1,8-Cineole (8.20%), 3-Carene (3.76%), (+) Limonene (6.76%), Terpinene (2.41%), Vanillin (1.70%), Butyl acetate (3.23%), 1,2-Benzenedicarboxylic acid (13.50%), 7-Acetyl-6-ethyl-1,1,4,4-tetramethyltetralin (19.61%). The other compounds were mostly presented in low amounts, most of the components of *Urtica* oil are found in references [10-14], and they are the main components of the species in other parts of the world, and some other components were not mentioned in the references. In other words, the aromatic oil content shows a diversity in the proportions of its components compared to the references. The chemical composition of the oil and their percentage in the oil extracted from *urtica pilulifera* grown in Syria is compared to the composition of oil studied in the references and that is due to several factors, such as the climatic difference and geographical conditions in each country such as soil type and conditions in which the leaves were harvested [12,13].

The analyses of table 1 shows that most of compounds are mentioned in other species, but with difference in proportions.

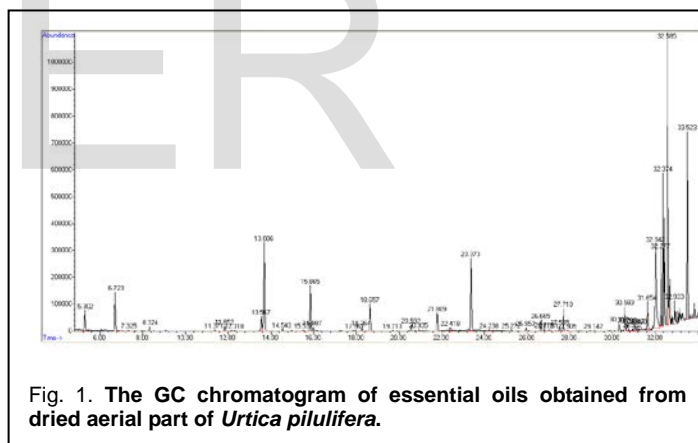


TABLE 1
CHEMICAL COMPOSITION OF ESSENTIAL OIL EXTRACTED OF
URTICA PILULIFERA.

NO	Compound	RT	%
1	Methylbenzene	5.302	1.6415
2	Butyl acetate	6.7208	3.2399
3	Furan-3-aldehyde	7.325	0.1458
4	Xylene	8.3229	0.3848
5	Benzaldehyde	11.371	0.1391
6	2-β-Pinene	11.853	0.3957
7	2-Methoxy-4-vinylphenol	12.3214	0.1087
8	(-)-Limonene	13.5638	1.2463
9	1,8-Cineole	13.686	8.2085
10	Terpinene	14.5413	0.1705
11	p-Guaiacol	15.5392	0.1521
12	3-Carene	15.8651	3.7624
13	n-Nonanal	15.9873	0.3288
14	Octyl heptafluorobutyrate	17.9696	0.1347
15	γ-Terpinen	18.2615	0.3824
16	γ-Terpinene	18.6552	2.415
17	Citronellol	19.7143	0.1388
18	Ocimene	20.59	0.6869
19	Carvone	20.9362	0.1721
20	Vanillin	21.812	1.7906
21	Trans-2,3-dimethylbicyclo[2.2.2]octane	22.4161	0.3454
22	(+)-Limonene	23.3733	6.7658
23	1-(4'-pentenyl)-1,2-epoxycyclopentane	24.2355	0.1271
24	Bicyclo[10.1.0]trideca-4,8-diene-13-carboxamide	25.2741	0.1325
25	Geranyl acetone	25.953	0.3483
26	alpha-Cetone	26.6658	0.9039
27	-Ionon	26.822	0.1714
28	1,4-Diazepine	27.0528	0.3009
29	1-Penten-3-one	27.5551	0.3782
30	Lilyal	27.7113	1.8666
31	Benzofuranone	27.9014	0.1183
32	Anozol	29.1437	0.1346
33	Methyl dilydrojasmonate	30.3181	0.8451
34	1-(4-Isopropylphenyl)-2-methylpropyl acetate	30.5829	2.062
35	Benzoic acid	30.7118	0.873
36	diethoxylated tridecyl alcohol	30.9494	0.1828
37	Vinyl	31.0415	0.3751
38	Ethylhexyl benzoate	31.187	0.3837
39	Octanal	31.6555	2.0563
40	1,2-Benzenedicarboxylic acid	32.0424	13.5056
41	Bisomel	32.28	3.7872
42	Neophytadiene	32.3751	5.2683
43	7-Acetyl-6-ethyl-1,1,4,4-tetramethyltetralin	32.5855	19.618
44	2-Propenoic acid	5.302	2.2418
45	Hexatriacontane	6.7208	11.5631

The essential oil exhibit significant activity against the pathogenic bacteria including: *Staphylococcus aureus*, *Escherichia coli*. in comparing to the gentamicin. The damping area is measured by a graduated ruler and the results are recorded in the table 2.

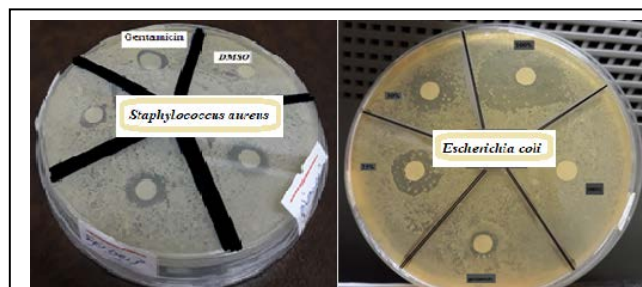


Fig. 2. The zones of inhibition by essential oil of *Urtica pilulifera* against to the growing of *Staphylococcus aureus* and *Escherichia coli*. in different concentration

TABLE 2
THE ACTIVITY OF ESSENTIAL OIL OF *URTICA PILULIFERA*, IT IS CALCULATED BY MEASURING THE DIAMETER OF THE INHIBITION ZONE (MM).

Org.	Essential oil Con.				
	100%	50%	25%	gentamicin	DMSO
<i>E. coli</i>	22.00 ± 1.52	13.33 ± 1.20	11.43 ± 0.99	4.3	–
<i>Staph. aureus</i>	6.65 ± 0.88	5.95 ± 0.61	4.31 ± 0.12	4.5	–

5 CONCLUSION

This is the first report of essential oil composition of *Urtica pilulifera* from Syria by the conventional hydro-distillation extraction (HD). 45 compounds representing 99.62% of the essential oil were characterized. By the domination of four compounds as majority of the components: (-)-Limonene (1.24%), 1,8-Cineole (8.20%), 3-Carene (3.76%), (+) Limonene (6.76%), Terpinene (2.41%), Vanillin (1.70%), Butyl acetate (3.23%), 1,2-Benzenedicarboxylic acid (13.50%), 7-Acetyl-6-ethyl-1,1,4,4-tetramethyltetralin (19.61%). The other compounds were mostly presented in low amounts. The *Urtica pilulifera* from Urticaceae family is a natural source of antimicrobial. The essential oil extracted this plant contains constituents with antimicrobial Activity. therefore, it can be used as a medical medicinal drug of high importance in the treatment of many diseases, and also described as anti-bacterial and inflammation.

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