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PHYSICOCHEMICAL PROPERTIES AND ITS APPLICATION OF DILL (*ANETHUM GRAVEOLENS*) AND PARSLEY (*PETROSELINUM SATIVUM*) AND THEIR ESSENTIAL OILS

[29]

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Keywords: Dill (*Anethum graveolens*), Parsley (*Petroselinum sativum*), Chemical composition, Essential oils, Application

ABSTRACT

The chemical composition for both dill and parsley herbs were studied. The highest content of crude protein, crude fiber, ash content, carotenoids, total chlorophyll and essential oil contents were found in dill herb were 27.25, 13.38, 16.02%, 174.32, 530.47 mg/100g and 0.879% (on dry weight basis), respectively, compared with that found in parsley. On the other hand, parsley was rich in both of ascorbic acid and β-carotene which were 682.77 and 34.95 mg/100g (on dry weight basis), respectively. Minerals content of dill and parsley herbs were analyzed and the results indicated that the dill contained higher amounts of Na, Ca, Cu and Zn than that found in parsley. Some physical properties of dill and parsley essential oils such as (color, specific gravity at 15°C, refractive index at 20°C, optical rotation and solubility in diluted ethyl alcohol) and some chemical properties (acid value, ester value and ester value after acetylation) were also studied. Composition of essential oils were identified, and contained (22.78%) phyllandrene and (23.85%) myristicin as the major terpene hydrocarbon compounds found in dill and parsley essential oils, respectively. The ability of extracted essential oils of dill and parsley as antimicrobial activity for (bacteria, yeast and mold) and as natural antioxidant agent applied in sunflower oil were also undertaken. The results indicated that the extracted essential oils of dill and parsley had antimicrobial activity which caused to inhibit the microorganisms growth and also, using these extracted essential oils as antioxidant which caused to improve the sunflower oil stability as natural extracts, achieved nutritional and economical gain.

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INTRODUCTION

Some leafy vegetables such as dill (*Anethum graveolens*) and parsley (*Petroselinum sativum*) as well as peppermint (*Mentha piperita*) are considered to be among the most famous, and important popular plants in Egypt and all over the world. These aforementioned crops are commercially cultivated for utilization of their consumed leaves, their characteristic flavor, nutritive value and medical effects. Many investigators reported that dill and parsley are called green drugs. This may be attributed to their medical effects on minimizing and curing stomach and bladder pains, besides headache and toothache (**Holden, 1965 and Francis, 1987**).

Dill (*Anethum graveolens L.*), a biennial or annual herb of the parsley family (*Apiaceae or Umbelliferae*), is native to south-west Asia or south-east Europe and cultivated since ancient times. The leafy tops can be clipped and used in cottage cheese, potato salad, cream cheese, tomato soup and salads (**Bailer *et al* 2001**). Also, **Mohamed *et al* (2001),** demonstrated that parsley leaves were rich in ascorbic acid content 676.10 mg/100g (on dry weight basis). Also, they found that parsley leaves contained a high amounts of total chlorophylls and total pheophytin, which were 1654.63 and 9.48 mg/100gm (on dry weight basis).

Faten (1998) reported that, the percentage of essential oil of dill and parsley were 1.2 and 0.8% (on dry weight basis), respectively. The physical properties (specific gravity at 15°C, refractive index at 20°C, optical rotation and solubility in alcohol for dill oil were 0.8950, 1.4822, +75° and soluble in 80% ethyl alcohol, while parsley oil were 0.8892, 14842, -3° and soluble in 75% ethyl alcohol. Also, the chemical properties (acid number, ester number and ester number after acetylation) of dill and parsley oils were, (0.8, 33.6 and 78.3) and (1.5, 10.2 and 20.8) for dill and parsley oil, respectively.

On the other hand, **Aly (1992)** analyzed the essential oils of parsley herb and identified the following constituents: myrcene, α- pinene, camphene, β-pinene, γ-terpinene, 1, 3, 8 – p – menthetriene, p – cymene, 1-methyl – 4 – isopropyl benzene, p-dimethyl stiarene, β-carbophyliene, α-hyomioline, β- cylrene, β-pharinene, α-pharinene, terpinene – 4 – ol, 2, 8 p- menthadaiene – 1 – ol, α- terpinol, cis-carviol, cumin aldhyde, carvacol and apiole.

Meanwhile, **Megahed (1980)** found that, the essential oil of dill contained α-pinene, β-pinene, α-phyllandrene, limonene, terpinene, p-cymene, methyl chavicol, anithol, myristicin and apiole.

Spices and herbs are used in food mainly for their flavors and aroma, in addition, certain spices prolong the storage life of foods due to their bacteriostatic or bactericidal activity, and for preventing rancidity using to their antioxidant activity (**Shelef *et al* 1980**). On the other hand, essential oils for many plants and spices were active against various food-borne bacteria and molds (**Gould, 1996).**

It is of interest to use the spices and their extracts as antibacterial agents in foods which can be attributed to use in foods because of: (1) their safety, as food additives being a trend toward the use of natural plant substances; (2) the reduction of salt ad sugar in dietary foods which caused to enhance flavor. Most essential oils of spices are classified as GRAS (generally regarded as safe) and considered to contain the antimicrobial activity of spices (**Ismaiel & Pierson, 1990**).

Most of the antimicrobial activity in essential oils derived from spices and culinary herbs appears to derive from phenolic compounds, while other constituents are believed to contribute little to the antimicrobial effect (**Delaquis *et al* 2002**).

This study was aimed to investigate the chemical composition of fresh dill and parsley herbs as well as studying the physicochemical properties of identified essential oils and its composition. The investigation was also undertaken the utilizing of their essential oils as natural antimicrobial and antioxidant agents.

MATERIALS AND METHODS

Materials

Fresh dill (*Anethum graveolens*) and parsley (*Petroselinum sativum*) were obtained from Aromatic and Medicinal Plants at El-Khairiya Barrage, Horticulture Research Institute, Agricultural Research Center, Giza, Egypt at season (2007). The root portions of the whole herbs were trimmed to obtain the herbs, then washed with current water, sorted and cut into small pieces, packed in polyethylene bags at refrigerator till uses.

Refined Sunflower oil free from antioxidant was purchased from Cairo Oils and Soap Company, El-Badrashin, Giza, Egypt. Butylated hydroxy toluene (BHT) as synthetic antioxidant was obtained from Gerbsaure Chemical Co. Ltd. Germany.

The strains were obtained from Cairo Mircen, Faculty of Agriculture, Ain Shams University, Egypt. The microorganisms tested were: Gram negatives (*Pseudomonas flourescens* and *Escherichia coli*; Gram positives (*Staphylococcus aureus and Bacillus subtilus)*; yeast *(Saccharomyces cerevisiae)*; and mold *(Aspergillus niger)*.

Analytical methods

Moisture content, total solids, ether extract (fixed and essential oils) , crude protein, total carbohydrates, crude fibers, ash, chlorophyll a, chlorophyll b, carotenoids, β-carotene and ascorbic acid were determined according to methods described in the **A.O.A.C. (2000).**

Minerals content for dill and parsley from (Na, Ca and K) were determined by using Emission Flame Photometer (Model Corning 410). Mean while, Zn, Fe, Mg, P and Cu were determined by using Atomic Absorption Spectrophotometer (Perkin-Elmer Instrument model 2380).

Essential oils from fresh dill and parsley herbs were obtained by steam distillation **(Guenther, 1961**), by using a Clevenger apparatus for more than 3 hours (**Egyptian Pharmacopeia, 1987**). The obtained essential oils were filtered and dried by anhydrous sodium sulphate before analysis.

Also, specific gravity at 15°C, refractive index at 20°C, optical rotation, solubility in diluted ethyl alcohol, acid value (as oleic acid) and ester value of the essential oils for tested samples were determined according to the methods described by the **A.O.A.C.(2000).**

The composition of essential oils for dill and parsley were identified with GC-MS analysis by using GC.5890A, Hewlett-Packard, Helium was used as the carrier gas under the following conditions: Oven temp. 80°C, oven equal time 2.00 min., inj. temp. 150°C, interface temp. 230°C, sampling time 3.0 min, column DB-1 (30.0 m. x 0.250 mm. x 0.25 micron), split less injection, column pressure 50 kpa, column flow rate 0.8 ml/min. Oven temp. progs: Initial temp. 80°C/2min. rate 5.0°C/min, 200°C/10min. mass range 40-350 M/8, solvent cut 3 min, GC prog. time 36 min. detector gain 1.8 kv.

Antimicrobial activity was determined using the method described by **Sleigh & Timburg (1981)**. The inhibition zones of microorganisms growth produced by dill and parsley essential oils were measured in(mm) and tabulated. Also, different concentrations 200, 300, 400 and 500 ppm of essential oils under the investigation were carried out, the filter discs were conducted to the above-mentioned concentrations and the used Petri dishes contained water agar media. The lowest quantity of the essential oils required to inhibit the growth of the microorganisms was designated as the minimum inhibitory concentration (MIC).

The oxidative stability of volatile oils were determined by *Rancimat* 679 (Metrom Ltd., CH 9100 Herisau, Switzerland) according to the method described by the **A.O.C.S. (1993)**.

RESULTS AND DISCUSSION

Chemical composition of fresh dill and parsley herbs

The chemical analysis of fresh dill and parsley herbs are shown in **Table (1)**.The results indicated that the moisture content of dill is higher than that of parsley, they were 89.76% and 83.22%, respectively.

The high percentage of ether extract related to the lower percentage of moisture in herb which may give an advantage to the plant as a good source of oil. While, a reverse trend was found for dry matter content, since parsley had a higher dry matter than that of dill. Parsley had a higher content of ether extract, total carbohydrates and ascorbic acid, when compared to dill herb. These values for dill herb were 4.49, 27.25, 38.87, 13.38 and 16.02 % (on dry weight basis) for ether extract, crude protein, total carbohydrates, crude fiber and ash. While, the corresponding values for parsley herb were 5.95, 21.72, 46.97, 11.10 and 14.26 % (on dry weight basis), respectively. However, parsley had higher contents of ascorbic acid either in fresh weight basis or dry weight basis.

The amount of different pigments in two herbs were 174.32, 385.35, and 145.12 mg/100g (on dry weight basis) for carotenoids, chlorophyll (a) and chlorophyll (b) for dill. While, the corresponding values were 83.01, 229.07 and 78.46 mg/100gm (on dry weight basis) for parsley.

Table 1. Chemical composition of fresh dill (*Anethum graveolens*) and parsley (*Petroselinum sativum*) herbs*

Constituents	Dill		Parsley	
	FWB**	DWB***	FWB	DWB
Moisture content (%)	89.76	-	83.52	-
Total solids (%)	10.24	100	16.48	100
Ether extract (fixed and essential oils) %	0.46	4.49	0.98	5.95
Crude protein (%)	2.79	27.25	3.58	21.72
Total carbohydrates (%)	3.98	38.87	7.74	46.97
Crude fiber (%)	1.37	13.38	1.83	11.10
Ash (%)	1.64	16.02	2.35	14.26
Ascorbic acid (mg/100g)	56.73	554.00	112.82	682.77
Carotenoids (mg/100g)	17.85	174.32	13.68	83.01
Chlorophyll (A) (mg/100g)	39.46	385.35	37.75	229.07
Chlorophyll (B) (mg/100g)	14.86	145.12	12.93	78.46
Total chlorophyll (mg/100g)	54.32	530.47	50.68	307.53
β-carotene (mg/100g)	3.47	33.89	5.76	34.95
Essential oil (% v/w)	0.09	0.879	0.07	0.425

*Fresh herb = whole herb (leaves + stem).
** FWB = Fresh weight basis. *** DWB =Dry weight basis.

Results also indicated that, β–carotene content was 33.89 and 34.95 mg/100gm (on dry weight basis) for dill and parsley, respectively.

Consequently essential oils for both dill and parsley have an important constituent since it could be used as a flavor component in food industry. The results showed that the dill and parsley contained 0.879 and 0.425 % (v/w)) of essential oils (on dry weight basis), respectively.

These results are in agreement with those reported by **El-Nikeety *et al* (2000) and Mohamed *et al* (2001),** they reported that the parsley herb contained 23.53% crude protein, 1.69% ether extract, 14.23% fibers and 13.98% ash (on dry weight basis).

Minerals content of fresh dill and parsley herbs

The contents of different minerals in two herbs (dill and parsley) are presented in **Table (2)**. These results indicated that the contents of: Ca, Na, Zn and Cu were higher in dill herb than that of parsley herb, which were 1394.34, 222.17, 53.03 and 26.46 mg/100g (on dry weight basis), respectively for dill herb, while, the corresponding values were 1302.67, 207.22, 5.16 and 4.19 mg/100gm (on dry weight basis), respectively for parsley. On contrary, parsley contained higher contents of P, K and Mg than that of dill herb.

Also, from **(Table 2)** it could be showed that, calcium, phosphorus and potassium were the predominant minerals in both dill and parsley herbs. These results are similar with that obtained by **Slupski *et al* (2005).**

Conclusively, dill and parsley are an excellent source of mineral constituents whose importance in the human diet is undisputable. Some of them, such as potassium, sodium, phosphorus, calcium, magnesium, or iron, are indispensable in the sustainment of human health.

Physicochemical characteristics of essential oils for dill and parsley

Some physicochemical characteristics of dill and parsley essential oils were determined, and the obtained results are presented in **Table (3).**

These results indicated that the specific gravity at 15°C, refractive index at 20°C and acid value (as oleic acid) for essential oils of parsley were higher than that of essential oils for dill , which recorded 0.8974, 1.4892 and 1.02, respectively for essential oils of dill. The corresponding values for essential oils of parsley were 0.9463, 1.5124 and 1.45, respectively.

Whereas, optical rotation an important criteria for judging the purity of the essential oils, **Guenther (1961).** Which was +65° and -4° for essential oils of dill and parsley, respectively.

On the other hand, essential oils of dill had a higher value of both ester value and ester value after acetylation. Which, were (35.47 and 82.37) for dill compared with that (9.73 and 21.62) for parsley essential oils. Generally the physical and chemical properties values agreed with those mentioned by **Megahed (1980) and Faten (1998).**

Composition of essential oils for dill and parsley

Essential oils have an important values since it could be used as a flavor components. Using Gas Chromatography and identification by Mass Spectrometry were used to determine the chemical components of fresh dill and parsley essential oils. The results of chemical composition of these essential oils are shown in **Table (4) and Figures (1 and 2).**

These results represented that parsley oil had the highest concentration of β-pinene (19.47%) and myristicin (23.85%). Both considered the major components in fresh parsley essential oils. These results are in agreement with those reported by **Ashraf *et al* (1979).**

Whereas, the predominant major identified components in the fresh dill essential oils were phyllandrene (22.78 %) and Limonene (20.52%), also, it contains apiole (12.73 %) and carvone (13.22%). These results are in agreement with those obtained by **Faten (1998).**

Antimicrobial activity of essential oils for dill and parsley

Effect of extracted volatile oils for dill and parsley were tested on different microorganisms (gram negative and positive bacteria), yeast and mold. The inhibition zones (mm) and minimum inhibitory concentration (ppm) of different microorganisms by using dill and parsley essential oils were illustrated in **Table (5)**. The most striking feature of the chosen microorganism have been either pathogenic or food spoilage microorganisms.

The results in **Table (5)** indicated that the gram positive bacteria (*Staphylococcus aureus* and *Bacillus subtilus*) were more sensitive to the effect of essential oils than gram-negative ones (*Pseudomonas flourecens* and *E. coli*). Also, it could be noticed that yeast (*Saccharmoyces cerevisae*) was more sensitive than mold (*Aspergillus niger*) by adding 200, 300,400 and 500 ppm for essential

Table 2. Mineral contents of fresh dill and parsley herbs* (mg/100g)

Mineral	Dill		Parsley	
	FWB**	DWB***	FWB	DWB
Sodium (Na)	22.75	222.17	34.15	207.22
Calcium (Ca)	142.78	1394.34	214.68	1302.67
Magnesium (Mg)	23.65	230.96	49.63	301.15
Phosphorus (P)	43.63	426.07	83.52	506.80
Iron (Fe)	4.21	41.11	6.78	41.14
Potassium (K)	547.82	5349.8	978.74	5938.96
Copper (Cu)	2.71	26.46	0.69	4.19
Zinc (Zn)	5.43	53.03	0.85	5.16

* Fresh herb = whole herb (leaves + stem).
** FWB = Fresh weight basis.
*** DWB = Dry weight basis.

Table 3. The physicochemical properties of essential oils for dill and parsley

Parameter	Essential oils of dill	Essential oils of parsley
Color	Greenish	Yellowish
Volatile oil content % (v/w)	0.879	0.425
Specific gravity at 15°C	0.8974	0.9463
Refractive index at 20°C	1.4892	1.5124
Optical rotation	+65°	-4°
Solubility	Soluble in 85% ethanol	Soluble in 90% ethanol
Acid value(% as oleic acid)	1.02	1.45
Ester value	35.47	9.73
Ester value after acetylation	82.37	21.62

oils of dill and parsley. These results were found to be in agreement with the results obtained by **El-Baroty (1988).**

Also, it could be noticed that, there is a relationship between the inhibitory act and chemical structures of the most abundant compounds in the essential oils under this investigation. However, both dill and parsley essential oils are characterized by containing a great amount of hydrocarbons and ketons such as (limonene 20.52%; phyllandrene 22.78% and carvone 13.22%) for dill essential oils and (β-pinene 19.47% and Myristicin 23.85%) for parsley essential oil **(Table 5)**.

The dill essential oil had the highest effect as antimicrobial against the microorganisms than parsley essential oil, these observations due to dill essential oil might be the presence of phenolic ether dill apiole (12.73%). These results are in agreement with data reported by **El-Baroty (1988),** who indicated that there is a relationship between the chemical composition of the oil and its antimicrobial effect.

Generally, from the previous data, It could be concluded that the dill and parsley essential oils may act as an antimicrobial activity against investigated microorganisms.

Table 4. Main components of essential oils for dill and parsley

Peak No.	Components	Aromatic herbs	
		Dill essential oil (%)	Parsley essential oil (%)
1	α- pinene	6.54	0.85
2	β- pinene	4.98	19.47
3	Camphene	-	4.20
4	limonene	20.52	Trace
5	P-Cymene	Trace	11.59
6	Myristicin	7.20	23.85
7	Phyllandrene	22.78	5.13
8	3-Carene	-	7.34
9	Terpinolene	Trace	0.37
10	Citronellol	-	0.93
11	Eugenol	0.78	-
12	Linalool	0.26	0.09
13	Linalyl acetate	5.14	0.12
14	Apiole	12.73	10.96
15	Carvone	13.22	-
16	Caryophyllene	0.15	-

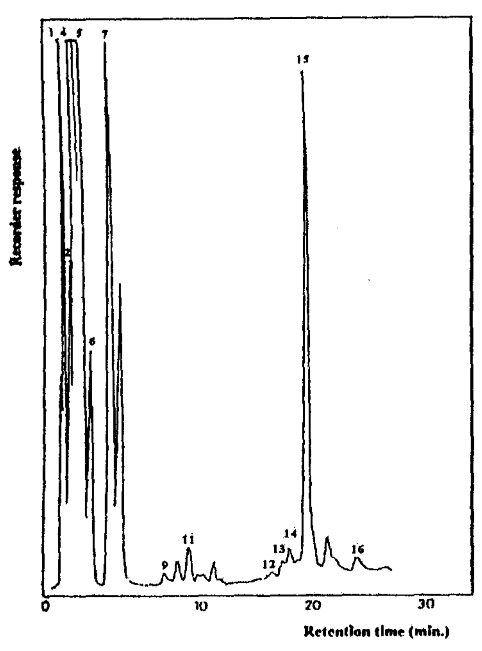

Figure 1. Gas chromatogram of the volatiles in dill

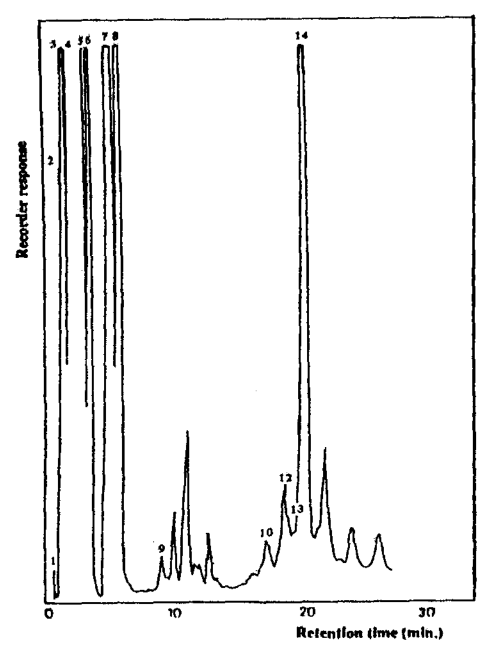

Figure 2. Gas chromatogram of the volatiles in parsley

Table 5. Effect of using dill and parsley essential oils for inhibition zones (mm) and minimum inhibitory concentration (ppm) on different microorganisms

Microorganism	Dill essential oil		Parsley essential oil	
	Zone*	MIC**	Zone	MIC
Staphylococcus aureus	30	300	17	300
Bacillus subtilus	32	300	16	300
Pseudomonas flourescens	14	400	12	400
Escherichia coli	18	400	14	300
Saccharomyces cerevisiae	20	300	15	400
Aspergillus niger	17	400	11	400

* The zone of inhibition (mm).
** MIC = Minimum inhibitory concentration (ppm).

Antioxidant activity

The essential oils extracted from dill and parsley herbs were added to sunflower oil with the concentrations of 200, 400, 800, 1200 and 1400 ppm. Stability of sunflower oil was measured by *Rancimate* method.

From results in **Table (6)**, it is clear that the addition of essential oils extracted from dill and parsley caused to increase the stability of sunflower oil at all used concentrations up to induction 8 hours in sunflower oil without addition of essential oils compared with that induction for 10 hours in sunflower oil with 200 ppm butylated hydroxy toluene (BHT) to 11.30, 10.30; 13.50, 12.00; 19.30, 16.50; 23.50, 19.30 and 24.30 and 20.50 hours for sunflower oil with 200, 400, 800, 1200 and 1400 ppm essential oil from dill and parsley herbs, respectively **(Table 6)**.

Also, the addition of dill and parsley essential oils may act as antioxidant and caused to increase the shelf life of sunflower oil during storage at ambient temperature. Therefore, it could be used dill and parsley essential oils instead of using synthetic antioxidants.

Table 6. Effect of adding different concentrations of dill and parsley essential oils on the oxidative stability of sunflower oil

Items	Oxidative stability (Induction periods = hours)
Sunflower oil (Control)	8
Sunflower oil + 200 ppm (BHT)	10
Dill essential oil added in ppm	
Sunflower oil + 200 ppm	11.30
Sunflower oil + 400 ppm	13.50
Sunflower oil + 800 ppm	19.30
Sunflower oil +1200 ppm	23.50
Sunflower oil +1400 ppm	24.30
Parsley essential oil added in ppm	
Sunflower oil + 200 ppm	10.30
Sunflower oil + 400 ppm	12.00
Sunflower oil + 800 ppm	16.50
Sunflower oil +1200 ppm	19.30
Sunflower oil +1400 ppm	20.50

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مجلة اتحاد الجامعات العربية
للعلـــــوم الزراعيـــــة
جامعة عين شمس ، القاهــرة
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الخواص الفيزيوكيميائية والتطبيقات لكلاً من الشبت والبقدونس وزيوتهما الطيارة

[٢٩]

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الملخــص العربـــي

تم دراسة التركيب الكيماوي لكــل مــن الـشبت والبقدونس: وكان محتوي الشبت عالياً من البـروتين الخام والألياف الخـام والرمـاد والكاروتينويـدات والكلوروفيل الكلي والزيوت الطيـارة حيـث بلغـت ٢٧,٢٥، ١٣,٣٨، ١٦,٠٢%، ١٧٤,٣٢، ٥٣٠,٤٧ ملجم/١٠٠ جرام و ٠,٨٧٩% (علي أساس الـوزن الجاف) علي التوالي، مقارنة بتلك المكونات الموجودة في البقدونس، ومن ناحية أخرى فلقد كان البقـدونس غنيـا في كل من حمض الاسكوربيـك والبيتـا- كاروتين والتي بلغت نـسبتـهما ٦٨٢,٧٧، ٣٤,٩٥ ملجم/ ١٠٠جـرام (علي أساس الـوزن الجاف) علي التــوالي.

هذا ولقد تم أيضاً دراسـة وتحليـــل العناصـر المعدنية لكلاً من الشبت والبقدونس ودلـت النتـائج المتحصل عليـها علي أن الشبت احتـــوي علـى نسب مرتفعة من الصوديـوم، الكالسيـوم والنحـاس والزنك مقارنة بتلك الكمية الموجودة في البقدونس.

ومن ناحية أخـرى فلقـد تـم دراسـة وتقـدير الخصائــص الطبيعية للزيوت الطيارة المستخلصـة من كل من الشبت والبقدونس والتـي شملت (اللون، الوزن النوعي علي درجة ١٥°م، معامـل الانكـسار عند ٢٠°م، الدوران الضوئي ودرجة الـذوبان فـي كحول الإثيل المخفـف) مـع دراسـة الخـصائص الكيميائية التي شملت (رقم الحامض، رقـم الاسـتر ورقم الاستر بعد الاستلة).

وبدراسة المركبات المكونة للزيوت الطيارة بجهاز الـــ GC-MS Spectrometry ووجـد أن المكونـات الأساسية هى الفيلاندرين (٢٢,٧٨%) والميرستـسين (٢٣,٨٥%) في الزيوت الطيـارة لكل مـن الـشبت والبقدونس على التوالي.

ولقد اشتملت الدراسة على اختبار قدرة الزيـوت الطيـارة المستخلصة كمواد مضادة لنمـو الكائنـات الحية الدقيقة (بكتريـا وخميـرة وفطر) وأيضا كمواد مضادة للأكـسدة بالنـسبة لزيـت عبـاد الـشمس. وأوضحت النتائج المتحـصـل عليهـا أن الزيـوت المستخلصة من الشبت والبقدونس ذات قـدرة عاليـة كمواد مضادة لنمو الكائنات الحية الدقيقة وكمضادات أكسدة لزيت عباد الشمس كمستخلصات طبيعية بـدلاً من استخدام مضادات الأكسدة الصناعية.

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