



***In-vitro* Comparative Phytochemical Screening, Antimicrobial, Antioxidant and LC-MS/MS Analysis of *Ocimum basilicum* and *Origanum vulgare* extracts in Saudi Arabia**

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Medicinal plants have shown to be a bountiful wellspring of organically dynamic mixtures for the improvement of new lead synthetic substances for drugs. Screening of *Ocimum basilicum* and *Origanum vulgare* plant extricates affirms the presence of different phytochemicals like carbs, cardiovascular glycosides, coumarins, flavonoids, tannins, terpenoid, steroids and phenols in the chose plants. Different phytochemicals as anthraquinone, alkaloids, saponins, proteins and phytosterols were identified and shifted considering the plant constituents. The antioxidant and antimicrobial activities of the *Origanum vulgare* L. and *Ocimum basilicum* plant extracts were evaluated by DPPH and by the agar well-diffusion method, respectively. LC-MS/MS analysis of *O. basilicum* and *O. vulgare* extracts showed the presence of 14 different compounds. The results confirmed the role of these extracts as promising potent antioxidants and moderate antimicrobial agents. The aim of this investigation was to ascertain the phytochemical screening, the antimicrobial, antioxidant activities and LC-MS/MS analysis of *Origanum vulgare* and *Ocimum basilicum* ethanolic extract that obtained from AlBaha in Kingdom of Saudi Arabia.

Keywords: *Ocimum basilicum* and *Origanum vulgare*, phytochemical screening, antimicrobial, antioxidant activities and LC-MS/MS analysis

Introduction

Scientists have become increasingly interested in plant study during the last three decades, particularly in industrialized countries such as Europe and America. It is believed that currently, around 60% of the world's population in treatment relies on herbs and natural products, which are thus recognized as a significant source of medications [1].

The term of medicinal plants (MP) includes a various type of plants used in herbalism and some of these plants has a medicinal activity. MP is the “backbone” of traditional medicine (TM), which means more than 3.3 billion people in the developed countries utilize MP on a regular basis [2]. TM is used for health by almost 80% of people in poor nations. The sensible use of TM in primary health care should be based on the World Health Organization's (WHO) recommendations for the assessment of herbal medicines [3].

The natural products that got from MP have demonstrated to be a bountiful wellspring of organically dynamic mixtures, a significant number

of which have been the reason for the improvement of new tip synthetics for drugs [4]. The prescriptions have been utilized and archived in Roman, Greek, Egyptian, Chinese and Indian restorative frameworks for around 5000 years. MP used in large scale in many countries because its biological effects on microorganisms. The active role of MP lays in existence some active compounds such as: (phenol, flavones, Coumarins, alkaloids... etc.) that are considered sources of many drugs in pharmaceuticals [5]. It is misleading to describe Saudi Arabia “barren desert” when it is there many regions clothed with trees, herbs and flowers as those in the south and some of west area.

Phytochemicals

Phytochemistry studies, a huge variety of organic substances like alkaloids, flavonoids, phenolic compounds, saponins, steroids, tannins, and terpenoids which accumulated in the plants. Furthermore, phytochemical screening refers to the extraction, their biosynthesis, metabolism, natural distribution and biological activities, survey and identification of the medicinally active substances

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found in plants and subsequently may lead to drug discovery and development [6].

Alkaloids are the most important active compounds in herbs. They display antimicrobial, anti-parasitic properties, treating asthma and anticancer activities. Some alkaloid molecules can act as narcotics which is the analgesic action of morphine. Moreover, they play a very important role in the immune systems of organisms and plants [7].

Plants containing chemical constituents having a steroidal structure that is responsible for medicinal property as inflammatory disorders such as asthma, rheumatoid arthritis, rhinitis, conjunctivitis, and multiple sclerosis [8]. The most important use of the cardiac glycosides is its effects in the treatment of cardiac failure. On the other hand, some cardiac glycosides are used as an antitumor activity and an inhibitory activity against rhinovirus [9].

Most of MD plants have different effect from a medical point of view, but in this research, the aromatic plants of *Ocimum basilicum* and *Origanum vulgare* that are having biological importance and considered herbs present surrounding the Mediterranean Sea, in Saudi Arabia were investigated. The controlled use of plant substances of medication is believed to be less toxic compared to that of synthetic products [10]. The molecules extracted from plants may be safe and efficient against antibiotic-resistant bacteria, which is why natural product research is critical [11].

Ocimum basilicum

Ocimum basilicum (*O. basilicum*, Lamiaceae) is one of the most popular plants found in Kingdom of Saudi Arabia. It belongs to family Lamiaceae that considered as the most employed medicinal plants as a worldwide source of spices and it is also popular as a kitchen herb. The leaves of *O. basilicum* are ovate, tip acute, petiolate, and green, up to 5 cm long and finely serrated [12]. Essential oils are abundant in basil, the leaves, blossoms, and stems of the basil plant are used in a variety of treatments for illnesses like colds, fevers, skin conditions, coughs, vomiting, etc. There are further claims that basil has anti-inflammatory, anti-spasmodic, anti-cancer, antiviral, antiseptic, and antibacterial properties. Basil also has a lot of antioxidants [13]. Traditional uses for the leaves of *O. basilicum* include antispasmodic, aromatic, carminative, digestive, galactagogue, stomachic, and tonic properties. They have also been used as a folk cure to treat a variety of conditions, including fever, flu-like symptoms, poor digestion, nausea, cramps in the stomach, gastroenteritis, migraines, sleeplessness, depression, gonorrhoea, dysentery, and chronic diarrhoea/tiredness [14]. *O. basilicum* contains high amounts of flavonoids but its

taste and scent are mainly caused by monoterpenes and phenylpropanoids [15].

Origanum vulgare

Origanum vulgare L., Lamiaceae family (*O. vulgare*) is a medium-sized, fragrant plant from the *Origanum* genus that grows every year. This species is considered to be one of the most extensively used aromatic plants within the Lamiaceae family [16]. *O. vulgare* is one of the most common herbs in Saudi Arabia compared with other regions. It grows in wet weather. *O. vulgare* is green herb have relatively long stem (30cm-60cm), and its leaves is small similar in shape to the tongue. When the herb is fully grown, the flowers appear that have similar color of cloves, and they are in dense and fusil form groups. *O. vulgare* is well known for its medicinal characteristics (diaphoretic, carminative, antispasmodic, antiseptic, tonic) and is utilized in many nations' traditional medicine systems. It has a fiery smell and has been broadly used in the farming, drug, and corrective businesses as a culinary spice, seasoning parts in food items, cocktails, and perfumery [17]. Previous studies have found that the essential oils of *O. vulgare* subsp. glandulos, which are high in carvacrol, have extremely good antibacterial action [18-19].

The aim of this study was to investigate the phytochemical screening, biological, and antioxidant activities, and the total phenols, flavonoids content as well as quantitative were analysis by LC-MS/MS for the air-dried leaves' extracts of two plants (*O. basilicum* and *O. vulgare*) that is cultivated in the kingdom of Saudi Arabia.

Material and Methods

Collection of the herb leaves. The two herbs (*Ocimum basilicum* and *Origanum vulgare*) were collected from Al-Baha City and the leaves of these herbs were taken and chopped softly.

Herbal Extraction The two plant leaves were collected and finely grinded after they are dried, then taken the weights of all the leaves. Plants were soaked two times with ethanol (75% concentration) for 48 h and filtered (**Table 1**).

The standard extraction scheme used during the study is shown below in figure (1): ***Aqueous Extracts of Plants for Primary***

Metabolic Analysis: plant was taken from each plant species 1:20 and afterward ground into a porcelain mortar utilizing refined water; the volume was finished in a standard cup, and the concentrate was kept at extremely low temperature (4°C) until play out the essential examination. The concentrates were sifted through a channel paper a few times and kept at 4°C in obscurity until use (20)

Ethanollic Extraction

Dried plant at flagon with around 1:15 of ethanol 75% after the principal filtration, the ethanolic remove was kept in the cooler. Similar stages multi day was rehashed. Then, at that point, tests were warming the concentrate utilizing water shower under condenser. The concentrates were sifted through a channel paper a few times following 24 hours and kept at 4°C in obscurity until utilize [21].

Phytochemical screening

Carbohydrates

Fehling and Benedict tests were determined according to Silva and Abeysundara [22] as well as the **Molisch's test** was determined using the method of Khayyat *et al.*, [21].

Proteins

Biuret and Xanthoproteic tests: The ethanolic layer pink color and th**Alkaloids**

Mayer's test and Wagner's test: The presence of alkaloid is indicated by a white creamy precipitate and a reddish brown precipitate, respectively [22].

Steroids (Liebermann-Burchard test):

Appearance of color development from violet to blue or bluish-green was an indication for the presence of steroids [21].

Cardiac glycosides (Keller-Kiliani test):

It was taken the formation of the brown ring as presented of cardiac glycosides [21].

Phytosterols

Liebermann-Burchard's test: The color change from violet to blue or green indicated the presence of phytosterols [23].

Salkowski's test: The formation of brown ring at the junction indicates the presence of phytosterols [22].

Saponins: The formation of stable thick foam (2cm) indicates the presence of saponins [22].

Terpenoids (Salkowski test): The red color appearance is an indication of terpenoids presence [21].

Phenols: The formation of bluish black color proves the presence of phenols [22].

Anthraquinone (Bontrager's test): The appearance of red, violet or pink color in the ammoniac layer (lower phase) was an indication for the presence of free anthraquinone [21].

Coumarins: The appearance of yellow color indicated the presence of coumarins [21].

e formation of yellow color indicates the presence of proteins respectively, while the purple color proves the presence of amino acids using the **Ninhydrin test** [22].

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Coumarins: The appearance of yellow color indicated the presence of coumarins [21]. **Flavonoids**

Alkaline reagent test: The intense yellow color solution becomes colorless on the addition of dilute acid proves the presence of flavonoids [22].

Shinoda test: The reddish color was an indication of flavonoids presence [21].

Tannins: A bluish black or greenish coloration was observed. It was an indication of the presence of pyrogallol tannin [21]

Antimicrobial evaluation

The antibacterial activity of the plant extract (*O. basilicum* and *O. vulgare* fruits 5g crude extracts in 100 ml of d H₂O) was investigated by well diffusion method [23].

In Vitro Determination of Antioxidant Activity

Free-radical scavenging activity: DPPH assay

The capacity of the prepared extracts to scavenge the 'stable 'free radical 2, 2-diphenyl-1-picrylhydrazyl (DPPH) was monitored according to the method described by Mekni *et al.* [24] at 517 nm in a spectrophotometer (Lambda 265, Perkin Elmer).

Determination of total phenolic content

The total phenolic content of *O. basilicum* and *O. vulgare* extracts was determined using the Folin-

Ciocalteu method described by Mutahar *et al.* [25] and expressed as mg Gallic acid equivalents (GAE) per 100 g fruit dry weight.

LC/MS/MS

The analysis of the *O. basilicum* and *O. vulgare* samples was performed using liquid chromatography–electrospray ionization–tandem mass spectrometry (LC-ESI-MS/MS) with an Exion LC AC system for separation and SCIEX Triple, Quad 5500+ MS/MS system equipped with an electrospray ionization (ESI) for detection [26].

TABLE 1. Plants weight and the amount of solvent involved in the extraction process

No	Plants	Weight (gm)	Solvent (EHOH)	
			First time (ml)	Second time (ml)
1	<i>Ocimum basilicum</i>	405	825	850
2	<i>Origanum vulgare</i>	100	275	400

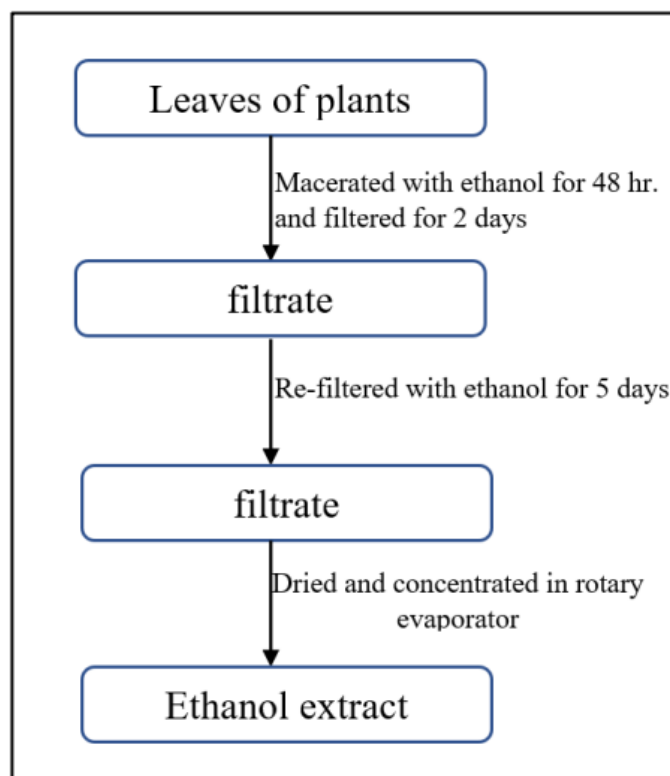


Fig. 1. Burn and treatment skin in rabbits.

Results and Discussions

Phytochemical Screening

The medicinal value of plants contains any chemical substances that have a clear physiological impact on the human body [26]. At the beginning of the extraction process, leaves of *O. basilicum* and *O. vulgare* herbs immersed in ethanol (EtOH) for two days, then were carried out

the filtration and preserve the extract. Then immersed the leaves again in EtOH for five days and then filtered to extract and obtain most of the phytochemicals contained within these leaves and to get the correct results during screening. According to the references, there are some screenings that have more than one method, such as screening of carbohydrate and proteins has

three ways, alkaloid, steroids, phytosterols and flavonoids were screened by two ways.

All Methods have been carried out in experiments to confirm the apparent results. The Preliminary phytochemical screening of ethanolic extracts (96%) of the plants was presented in Table (1). It showed that *O. basilicum* and *O. vulgare* contains tannins, Cardiac glycosides, Coumarins, Saponins, Steroid, flavonoids, phenols, terpenoids, alkaloids and carbohydrates but both are free from anthraquinone and proteins, while the *O. vulgare* only is free from Phytosterols only.

Qualitative screening using ethanolic extract indicated the presence of most the phytochemical constituents and the absence the others from of it in *O. basilicum* plant extract. As it is was seen from the Table (1), the constituents were revealed presenting in varying proportions in plants. Note that, the resulting colors may increase (>+), remain light (+) or absent (-) according to the results observed during the experiment. The results denoted that, phytochemical is the most abundant in all plants including cardiac glycosides, phenols, and saponins, as the very dark brown ring appeared in on all plants (+++ or ++), which can be considered the plants as having an active effect as antitumor activity and an inhibitory activity against pathogenic microbes.

The present results were in agreement with that obtained by Khairul-Bariyah *et al.* [26] and Pandey *et al.* [27], as they stated that the aqueous extract of *O. basilicum* revealed the following upon elemental

analysis and phytochemical screening: secondary metabolites, flavonoids, Phenolic acids, cardiac glycosides, tannins, saponins, calcium, sodium, potassium and magnesium. Also, the screening of the presence phytochemical constituents in *O. vulgare* were light positive (+).

The difference in the proportion of the presence phytochemicals and absence it in plants depends on the difference area, weather and soil. So, these constituents may be absent and appear on the same plant based on these reasons.

As well as, the obtained results were in accordance of several authors who denoted that extracts from the leaves of *O. basilicum* and *O. vulgare* were investigated the existence of many phytochemical constituent which of them have antioxidant activity [27]. Tests for tannins, steroids, terpenoids, flavonoids and cardiac glycosides were positive in both methanolic and aqueous extracts. The presence of all phytochemicals, especially (cardiac glycosides and phenol) made the plant, promising source of antimicrobial, antioxidant and cytotoxicity properties [28,29]. These plants can be a source of useful drugs, but further phytochemical analysis of these plants confirms the studies are required to isolate the active component from the crude plant extract for proper drug development.

Table (2), Fig (1) illustrated the commendable antibacterial effects of ethanolic extracts of *O. vulgare* and *O. basilicum* on the development of different Gram positive and negative bacteria, and fungi using the agar well diffusion method

TABLE 2. Phytochemical screening of ethanol extract of *O. basilicum* and *O. vulgare*

Plants	<i>Ocimum basilicum</i>	<i>Origanum vulgare</i>
	Phytoconstituents	
Anthraquinone	-	-
Alkaloid	+	+
Carbohydrate	++	++
Cardiac glycosides	+++	++
Coumarins	+	+
Flavonoid	++	+
Tannins	++	+
Saponins	++	+
Terpenoid	++	+
Steroid	+	+
Proteins	-	-
Phytosterols	+	-
Phenols	+++	+

Antimicrobial activity of *O. basilicum* and *O. vulgare*

The results indicated that the *O. basilicum* extract showed a moderate antibacterial activity against all

tested pathogens with inhibition zone ranged from 12-16 mm and there is no activity was detected against *S. aureus*. As well as, the *O. vulgare* extract showed a strong antibacterial activity against *S.*

aureus (IZ 22 mm) as a gram-positive bacterium with IZ of 18 mm against *E. coli* as a gram-negative bacterium, while it characterized by a moderate antibacterial activity with the others pathogens. On the other hand, the weak antifungal activity was noticed against *C. albicans* and *A. niger*. About the antifungal activity, the extracts inhibit the growth of *C. albicans* with inhibitory activity of 16 mm from *O. basilicum* extract and 12 mm from *O. vulgare* extract. While, the current plant extracts exhibited a weak antifungal activity (12 mm) against *A. niger*.

The results indicated that whereas *E. coli* and *P. aeruginosa*, which are Gram negative bacteria, were found to be more resistant to both plant extracts than *C. albicans* and *A. niger*, *Staphylococcus aureus* was shown to be more sensitive (with the broadest zones of inhibition). On the other hand, compared to Gram positive bacteria, Gram negative bacteria were more robust (a moderate zones of inhibition).

The outcomes were consistent with Stanislava *et al.* [30], who noted that *O. basilicum* has an antibacterial action since the plant is rich in essential oils and phenolic components (flavonoids, phenolic acids), which are known to have an antimicrobial activity. Additionally, our findings supported those of Martins *et al.* [31], who found that the ethanol extract of the oregano plant at a concentration of 20 mg/mL had a modest antibacterial effect against Gram-negative bacteria. The antibacterial properties of basil and oregano essential oils have been the subject of earlier research. The presence of phenolic chemicals such as carvacrol, thymol, and linalool has been linked to high antibacterial action [11; 32]. There have been fewer studies on the antibacterial properties of oregano and basil extracts, because of the poisonous effects of carvacrol, thymol (oregano), and estragol (basil), which are concentrated in essential oils, testing of these extracts may be useful [33].

Basil and oregano are both widely used spices that may be useful in the treatment and prevention of a variety of illnesses, including bacterial infections. As a plentiful source of secondary metabolites with antibacterial action and phenolic compounds were denoted by Stanislava *et al.* [30]. *O. vulgare* has high cytotoxicity, antioxidant, and antibacterial properties and is traditionally used to treat respiratory conditions, dyspepsia, and rheumatoid arthritis. These properties are mostly due to the presence of isomeric phenolic components [34] and monoterpenoids [35] as major chemical classes of secondary metabolite.

In Oregano and basil extracts, phytochemical analysis revealed the presence of phenolic, flavonoids, steroids, triterpenoids, and saponins (fig 2). These chemicals may be responsible for the antibacterial activity and function as antibacterial agents of the herbs such as steroids, flavonoids, and phenols. Numerous investigations have noted the antibacterial properties of phenolic and flavonoids, steroidal saponins and triterpenoids, saponins (36), and steroidal glycosides [37].

Meanwhile, antibacterial efficacy will depend on the species of oregano and basil extracts, the effectiveness of the extraction process, and the location of their active compounds. Our findings, however, demonstrated that all the test organisms were inhibited by the ethanol extract of oregano and basil, locational or seasonal differences may be to blame for this variation. In addition to being used as spices in daily life, the two aromatic species *O. vulgare* L. and *Basilicum* L. (Lamiaceae) also have medicinal uses that include the treatment of digestive and respiratory issues [18]. Antioxidant activity is known to be influenced by genetic and environmental factors, including the quality of the original plant, its origin and climate, the date of harvest, storage, and processing, as well as the extraction process and its chosen parameters [38].

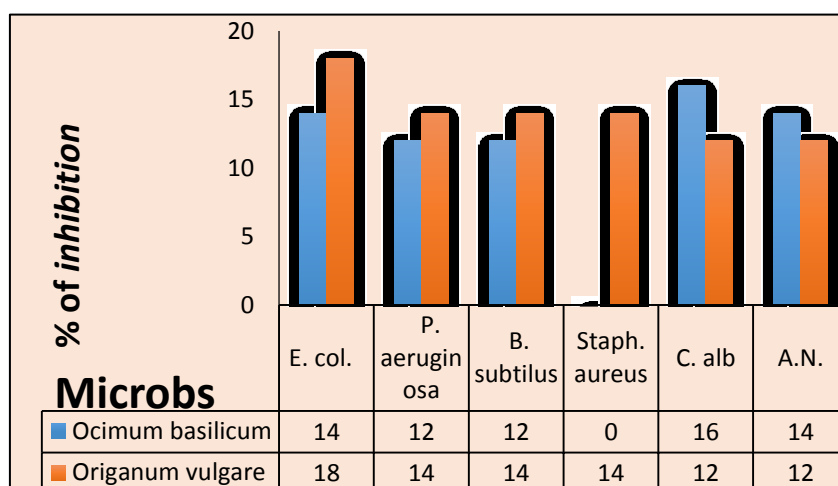


Fig. 2. Antimicrobial activity of ethanol extract of *O. basilicum* and *O. vulgare*.

Free radical scavenging DPPH assay

The DPPH technique was used to assess the antioxidant activity of *O. vulgare* and *O. basilicum* extracts in an *in vitro* assay. According to Kaurinovic *et al.* [39], the comparison of the antioxidant activities of the analyzed extracts revealed variable results that varied according to the tested extracts and the model system used for the experiment. Due to its stability (in radical form) and straightforward testing, the DPPH radical is one of the most often employed substrates for quick evaluation of antioxidant activity. In the DPPH assay, the standard Mekni *et al.* [24] technique was used to examine the studied extracts' capacity to serve as donors of hydrogen atoms or electrons in the transformation of DPPH into its reduced form, DPPH-H. In order to assess the antioxidant activity of different plant extracts, Gallic acid has been utilized as a reference.

The result obtained showed that the antioxidant activity of the *O. basilicum* and *O. vulgare* extracts was 87.05 % and 90.43 % respectively, by DPPH assay (fig 3). It showed that the antioxidant activity of both plant extracts was more than 50%, they consider having strong antioxidant activity. These results were in accordance to Kaurinovic *et al.* [39], who mentioned that *O. basilicum* and *O. vulgare* have a strong antioxidant activity that has been evaluated using DPPH assay. It is possible to deduce that extracts of *O. basilicum* and *O. vulgare* have some link between DPPH scavenger activity and flavonoids concentration. This demonstrated once more that the antioxidant ability of extracts is determined not only by the quantity but also by the type of flavonoids present. The presence of caffeic acid derivatives in these extracts, which have two hydroxyl groups in the ortho position, could explain the high activity of the H₂O extracts of *O. basilicum* and *O. vulgare*.

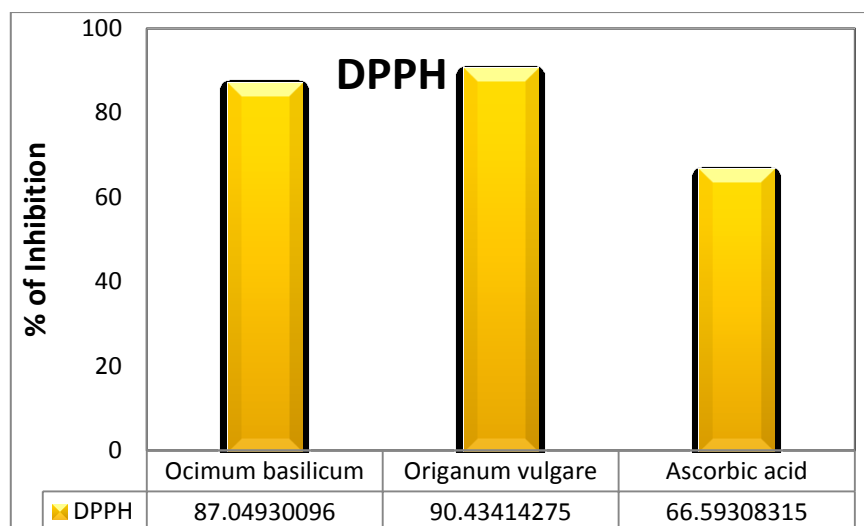


Fig. 3. Antioxidant Activity DPPH% of ethanol extract of *O. basilicum* and *O. vulgare*

Total Phenolic content

The concentration of phenolic in the two plant extracts was presented in (Figure 4) was dependent on the solvent and the experimental conditions. The total phenolic content was expressed as Gallic acid equivalents (GAE) in milligrams per gram of dry extract. The amount of phenolic compounds in the *O. basilicum* plant ethanol extract was 15.80 mg GAE/1g of extract). On the other hand, the amount of phenolic compounds in the *O. vulgare* plant ethanol extract (7.80 mg GAE/1g of extract). These results were in agreement with Kaurinovic *et al.* [39], who mentioned that the total phenolic content of *O. basilicum* and *O. vulgare* extracts ranged from 4.21 ± 0.01 mg GAE/g dry extract (d.e.) (from *O. basilicum*)

to 14.13 ± 0.05 mg GAE/g d.e. (extract of *O. vulgare*).

As well as, Hamdan *et al.* [40] reported that *O. basilicum* is one of the most common plants used traditionally all over the world and in Saudi Arabia to treat many diseases [41]. The results obtained in (Table 4) were in accordance with Dörr *et al.* [15] who reported that *O. basilicum* contains high amounts of flavonoids contents increases antioxidant activity and there was a linear correlation between phenolic or flavonoids content and antioxidant activity.

Kaurinovic *et al.* [39], conducted another study and found that *O. basilicum*'s total phenolic content and

flavonoids content were higher in the H₂O extract than they were in the other solvent extracts. According to Bozin *et al.* [35], phenols contain antioxidant qualities and the capacity to put an end to free radical processes. By lowering membrane fluidity, phenols like flavonoids stabilize membranes by limiting the transport of free radicals and lowering the peroxidation of membrane lipids. The ability of phenolic compounds, especially flavonoids, to bind to certain of the necessary phospholipids and membrane proteins is what causes membrane stabilization [42].

Previous research has investigated the links between total phenolic content and antioxidant

properties in a variety of plants [43; 44]. Some researchers found good positive linear correlations, whereas others found poor linear correlations or couldn't explain the relationship between total antioxidant activity and phenolic content, as shown by Mata *et al.* [45] and observed in our analyses. Plant extracts and phyto-products are being used to relieve a variety of stresses such as diverse factors such as species, soil conditions, climatic, harvest season, geographical location, growth conditions and extraction technique which emphasize the need to standardize quality control studies in the production of *O. vulgare* preparations due to their accessibility, cost effectiveness, and biodegradability [27; 46].

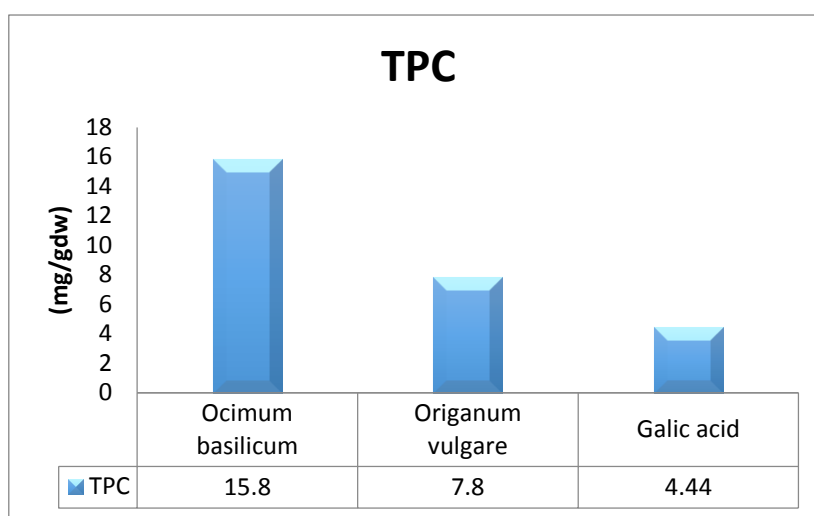


Fig. 4. Total phenolic compounds of ethanol extract for *O. basilicum* and *O. vulgare*

LC/MS/MS Analyses

The *O. basilicum* and *O. vulgare* extracts were tested by LC-MS/MS, and their composition measured by $\mu\text{g/g}$ dry weight plant extract was shown in Table 5 and Figures 5 and 6. LC-MS/MS analysis of *O. basilicum* and *O. vulgare* extracts

showed the presence of 14 different compounds. The most abundant in *O. basilicum* and *O. vulgare* were Naringenin (68.50 and 168.30 $\mu\text{g/1gm}$ plant extract) respectively, and Caffeic acid 3.17 and 3.22 $\mu\text{g/1gm}$ plant extract, Luteolin 2.20 and 1.40 $\mu\text{g/1gm}$ plant extract in *O. basilicum* and *O. vulgare*, respectively.

TABLE 5. LC/MS/MS of *O. basilicum* and *O. vulgare* ethanolic extracts

No	Name	Precursor m/z	Rt	Conc. $\mu\text{g/1gm}$ plant extract	
				<i>Ocimum basilicum</i>	<i>Origanum vulgare</i>
1.	Chlorogenic acid	355.1/163	7.34	0.06	ND
2.	Daidzein	255.1/199	12.93	ND	ND
3.	Gallic acid	168.9/124.9	3.85	0.06	0.13
4.	Caffeic acid	178/135	8.04	3.17	3.22
5.	Rutin	609/299.9	9.72	0.00	0.00
6.	Coumaric acid	162.9/119	9.53	4.11	0.86
7.	Vanillin	151/136	9.57	0.16	0.31
8.	Naringenin	271/119	15.05	68.50	168.30
9.	Quercetin	301/151	13.59	0.02	0.46
10.	Ellagic acid	301/145	9.92	ND	0.01
11.	3,4-Dihydroxybenzoic acid	152.9/109	5.72	1.27	0.80
12.	Hesperetin	301/136	15.64	5.33	ND
13.	Myricetin	317/137	11.72	ND	ND
14.	Cinnamic acid	146.9/102.6	14.20	ND	ND
15.	Methyl gallate	183/124	7.45	ND	ND
16.	Kaempferol	284.7/93	15.36	ND	0.02
17.	Ferulic acid	192.8/133.9	10.25	1.04	0.25
18.	Syringic acid	196.8/181.9	8.41	0.64	2.56
19.	Apigenin	269/151	15.05	0.89	1.08
20.	Catechin	288.8/244.9	7.34	ND	ND
21.	Luteolin	284.7/132.9	13.52	2.20	1.40

The results detected the other compounds different in each plant extracts as, Hesperetin and Coumaric acid in *O. basilicum* with value of (5.33 and 4.11 $\mu\text{g/1gm}$ plant extract) while detected the Syringic acid of 2.56 $\mu\text{g/1gm}$ plant extract) in *O. vulgare* extract. These results concur with earlier studies, which demonstrated the presence of carvacrol and thymol in *O. vulgare* ethanol extract (Eos) made from variously processed plant materials.

Additionally, 67 compounds that made up 99% of the peaks in the GC-MS chromatograms were used to characterize Basil Eos, while, the most common class of volatile chemicals (54.32%) was monoterpenes. On the other hand, the obtained results were consisted with previous reports that mentioned, Quercetin, luteolin, apigenin, flavonoids and kaempferol were detected as abundant constituents in the *Origanum* genus [47; 48 and 41].

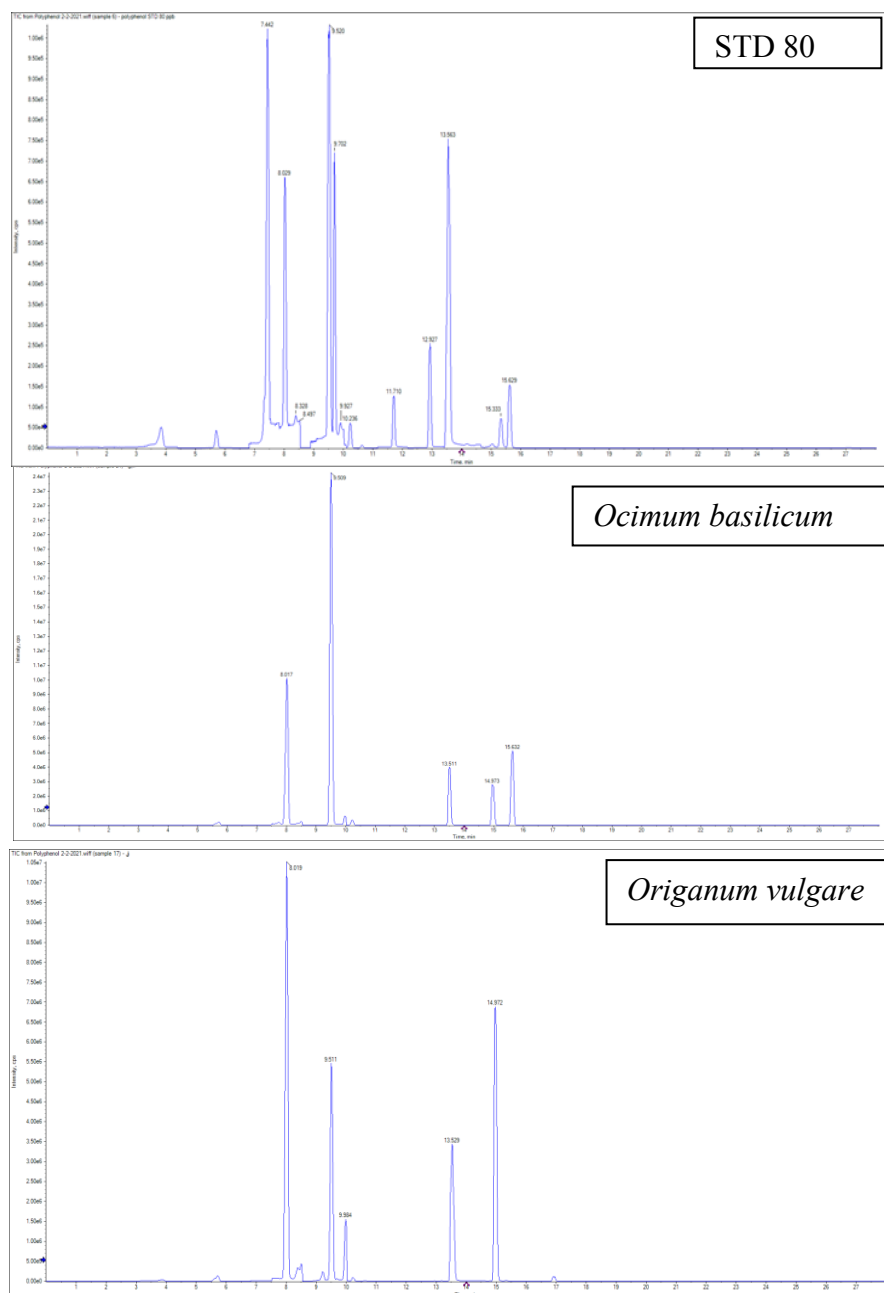


Fig. 5. LC/MS/MS of *Ocimum basilicum* and *Origanum vulgare* ethanolic extract

As should be visible from the above conversation, various metabolites were at the same time tracked down close by *Lamiaceae* species. It ought to be noted that critical varieties in the synthetic piece of the two current plants separates have been accounted for. A few variables were found to impact the structure of concentrates, including the geographic beginning, a piece of the plant, time of collecting, consequently the phenological phase of the plant, and furthermore the concentrate segregation technique, natural contrasts inside species and test extraction time [49].

The discussion above showed that two *Lamiaceae* species concurrently contained many specialized metabolites. It should be highlighted that there are considerable differences in the chemical composition of the two present plant extracts. According to Teixeira *et al.* [49], a number of variables were discovered to have an impact on the composition of extracts, including the geographical origin, part of the plant, season of harvesting, and consequently the phenological stage of the plant.

Due to their abundance in phenolic compounds and other secondary metabolites with antimicrobial activity, Origano and basil, which are relatively prevalent spices in daily diet, may have a significant role in the prevention and treatment of many diseases, including bacterial infection. The study also suggests that more research be done on medicine formulations that are efficient, safe, affordable, and non-toxic. This would not only add value to our resources but also produce a logical strategy for utilizing them.

Conclusion

Various Phytochemical screening revealed the presence of carbohydrates, cardiac glycosides, coumarin, flavonoids, tannins, terpenoid, steroids and phenols in all two herbs. On the other hand, the biological activities of two herbs were tested against microbial strain (gram-positive and gram-negative). However, the DPPH radical scavenging method was used to examine the antioxidant activity of the two plant extracts, and the results revealed average inhibition. The total phenolic and flavonoids content were investigated. LC-MS/MS analysis of *O. basilicum* and *O. vulgare* extracts showed the presence of 14 different compounds. The most abundant in *O. basilicum* and *O. vulgare* were Naringenin (68.50 and 168.30 µg/1gm plant extract) respectively.

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Conflict of interest

All Authors declare that there is no conflict of interest.

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مقارنة للفحص الكيميائي النباتي في المختبر وتحليل لمضادات الميكروبات ومضادات الأكسدة وتحليل
الكروماتوجرافي LC-MS/MS لمستخلصات *Ocimum basilicum* و *Origanum vulgare*
في المملكة العربية السعودية

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في هذا العمل أثبتت النباتات الطبية (MP) أنها مصدر وفير للمركبات النشطة بيولوجيًا لتطوير مواد كيميائية جديدة تستخدم في تحضير للأدوية. يؤكد فحص مستخلصات نبات *Ocimum basilicum* و *Origanum vulgare* وجود مواد كيميائية مختلفة مثل الكربوهيدرات، جليكوسيدات القلب، الكومارين، الفلافونويدات، التربينات، التيربينويد، الستيريديتات والفينولات في النباتات المختارة. كما تم الكشف عن مواد كيميائية أخرى مثل الأنثراكينون والقلويدات والصابونين والبروتينات والفيتوستيرول وتنوعت كميتها بناءً على مكونات النبات. تم تقييم الأنشطة المضادة للأكسدة والمضادة للميكروبات لمستخلصات نبات *Origanum vulgare* و *Ocimum basilicum* بواسطة DPPH وبواسطة طريقة انتشار الأجار، على التوالي. وقد أظهرت نتائج تحليل LC-MS/MS لمستخلصات *O. basilicum* و *O. vulgare* وجود 14 مركبًا مختلفًا. وأكدت النتائج دور هذه المستخلصات كمضادات أكسدة قوية ومضادات ميكروبات معتدلة.

حيث كان الهدف من هذا البحث هو التأكد من الفحص الكيميائي النباتي والأنشطة المضادة للميكروبات ومضادات الأكسدة وتحليل LC-MS/MS بواسطة المستخلص الإيثانولي *Origanum vulgare* و *Ocimum basilicum* الذي تم الحصول عليه من منطقة الباحة في المملكة العربية السعودية.

الكلمات المفتاحية: الريحان والأوريغانوم الشائع، الفحص الكيميائي النباتي، أنشطة مضادات الميكروبات ومضادات الأكسدة وتحليل الكروماتوجرافي LC-MS/MS