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PHYTOCHEMICAL SCREENING, ANTIBACTERIAL AND ANTIOXIDANT ACTIVITIES OF *Ocimum Basilicum L.* CULTIVATED IN BISKRA, ALGERIA

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Abstract. Algerian flora contains a wide range of aromatic plants of great therapeutic interest thanks to their biologically active secondary metabolites which makes them a subject of scientific interest. In this work, we were interested in *Ocimum basilicum L.* of *Lamiaceae* family cultivated and harvested in Biskra city located at South-East of Algeria. Phytochemical screening has been performed to reveal the presence of flavonoids, tannins, coumarins, essential oil, and other phytochemicals. The contents of total polyphenols, total flavonoids and total tannins have been determined. The obtained essential oil and extracts have been the subject of antibacterial and antioxidant assays. Our outcomes confirmed that *Ocimum basilicum L.* has a multiplicity of potential phytochemicals with a considerable amount and can be used as an alternative natural source of antioxidant and antibacterial components.

Keywords: *Ocimum basilicum L.,* phytochemical screening, antibacterial, antioxidant, Biskra.

1. Introduction

Bacteria are microorganisms causing numerous life-threatening diseases such as tuberculosis, brucellosis, plague, diphtheria, typhoid, typhus, cholera, diphtheria, and pneumonia.[1] Recent statistics declare that the world population is suffering from various infections issue of these bacteria.[2]

Free radicals are highly reactive chemical species causing several diseases and disorders of different intensities. The reactive oxygen chemical species including hydroxyl radical $OH^{\cdot}$, hydrogen peroxide H_2O_2, and superoxide anion $O_2^{\cdot-}$ play an important role in the establishment of diseases such as arthritis diabetes aging, asthma, dementia, mongolism, carcinoma, immunosuppression, and Parkinson's disease.[3] These previously mentioned radicals oxidize and damage the DNA structure, proteins, and other biomolecules. In a healthy body, there is an equilibrium between free radicals and antioxidants. The antioxidants generated in the human body are not enough to face off the deleterious effects of free radicals which make an exogenous supply of antioxidants extremely necessary in this case.[4]

Aromatic plants constitute a very important natural wealth, the valuation of which requires a perfect knowledge of the properties to be valued. The properties of theses aromatic plants depend on the presence of various bioactive agents belonging to different chemical classes. This type of plants produces secondary metabolites having significant medicinal importance. The major phytochemical groups which impart biological activities against infections are alkaloids, phenols, polyphenols, quinones, flavonoids, tannins, terpenes, polypeptides, essential oil, and other compounds.[5,6]

The basilic "*Ocimum basilicum L.*" is a plant of the *Lamiaceae* family. This annual aromatic herb native to India, and widely cultivated in Algeria hardly exists in the wild, is morphologically very variable, perennial in a tropical climate and generally known under the name "Lahbeq" "Hamahim" and also "Hebeqelailaa".[7,8] Traditionally, basilic has been used as a medicinal plant in the treatment of nervous diseases, dizziness, colic, constipation, bloating, cough, whooping cough, migraine of nervous or gastric origin, and canker sores. Basilic essential oil is also known for its antimicrobial and insecticidal activity. It is also used in cooking for its sweet smell and very pleasant, minty flavor. Currently, basilic is therefore very widespread throughout the world. However, it remains deeply rooted in Mediterranean gastronomy.[9-14]

The different varieties of *Ocimum Basilicum L.* including *Ocimum basilicum var. album, Ocimum basilicum var. anisatum, Ocimum basilicum var. densiflorum, Ocimum basilicum var. misshapen, Ocimum basilicum var. glabratum, Ocimum basilicum var. majus, Ocimum basilicum var. minimum, Ocimum basilicum var. pilosum,*

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Ocimum basilicum var. purpurascens, and *Ocimum basilicum var. thyrsiflorum* are distinguished by their colors, shapes, sizes, and smells.[15,16]

In this work, we focus on the extraction of the *Ocimum Basilicum L.* plant. Phytochemical screening has been performed to characterize the presence of different secondary metabolites. The antibacterial and antioxidant activities of the phenolic extracts and essential oil have been evaluated using diffusion technique test and DPPH radical scavenging assay, respectively. The obtained results were well discussed.

2. Experimental

2.1. Plant Material Collection

Ocimum basilicum L. leaves were collected from Biskra city located in the South-East of Algeria (34° 50' 20'' N 5° 42' 52'' E) during May month. The obtained leaves have been dried in the shade using an open-air. The dried plant material was well grinded using an agate mortar to obtain a fine powder prior to the extraction process.

2.2. Extraction

2.2.1. Essential Oil Extraction

2.2.1.1. Hydrodistillation

It is a technique widely used for the extraction of essential oil. The advantage of this technique consists in the reduction of the distillation temperature. Volatile compounds are therefore entrained at temperatures much lower than their boiling point, which prevents their decomposition.

In this process, 50 g of the milled vegetable material was introduced into a 1-liter flask, adding a quantity of distilled water corresponding to 2/3 of the volume of the flask. The extraction operation is carried out for 6 hours from the start of boiling. The obtained distillate was subjected to liquid-liquid extraction three times with chloroform $CHCl_3$ and the organic phase is dried with anhydrous magnesium sulfate $MgSO_4$. After simple filtration, evaporation was carried out to completely remove the solvent. Finally, the obtained essential oil was stored in a smoked and well-sealed bottle at a temperature of 278 K.[17]

2.2.1.2. Clevenger Extraction Process

The extraction of the essential oil was carried out by hydrodistillation using a Clevenger apparatus. 100 g of the plant material was weighed and introduced into a 2-liter flask filled with water to 2/3 of its volume. The mixture was heated at 373 K for 6 hours. The vapors containing the volatile substances passed through the refrigerant and condensed. After condensation, both the light fraction rich in essential oil and the heavy fraction were separated thanks to their difference in density.[18]

2.2.2. Extraction of Polyphenols

To extract the polyphenols from the studied plant, 200 g of vegetable powder were macerated in a methanol/water solvent mixture (v/v as 70/30) three times, with stirring from time to time at room temperature and in the dark, a sufficient quantity of solvent was added every 24 hours. The hydro-alcoholic extracts were combined and evaporated under reduced pressure at a temperature below 323 K until obtaining a viscous solution. The resulted crude residue of dark green color was tromped in 150 mL of boiling distilled water overnight and then filtered. We proceed to the confrontation with organic solvents of increasing polarity. The residual phase is successively exhausted with dichloromethane, ethyl acetate and 1-butanol. Extractions are repeated three times. Thus, the obtained four organic phases were dried with anhydrous magnesium sulfate $MgSO_4$, filtered and evaporated under reduced pressure at a temperature below 323 K.[19]

2.3. Phytochemical Screening

Phytochemical screening was performed for essential oil, tannins, saponins, reducing sugars, flavonoids, alkaloids, terpenes, unsaturated sterols, coumarins and anthocyanins. The color intensity or the precipitate formation was used as analytical response to these tests.

2.3.1. Tannins

We introduce 10 g of the grinded plant material into a beaker containing 20 mL of ethyl alcohol C_2H_5OH 50 % to be macerated for about 15 min and then filtered. A few drops of ferric chloride $FeCl_3$ were added to the obtained filtrate. The appearance of a dark green or blue-green color indicates the presence of tannins. However, dark green color is dedicated to the presence of catechetical tannins while the appearance of a blue-green color is attributed to the presence of gallic tannins.[20]

2.3.2. Saponins

We dissolve 2 g of the powdered plant part in 80 mL of distilled water. The obtained aqueous mixture was heated and then filtered. The filtrate was cooled and then stirred vigorously. The appearance of persistent foam having more than 1 cm high indicates the presence of saponins.[20]

2.3.3. Sugars

The plant material was extracted in distilled water for 2 hours and then filtered. 2 to 3 drops of Fehling's liquor were added to the filtrate that was under heat in a water bath. The appearance of a brick red color indicates the presence of carbohydrates reducing Fehling's liquor.[20]

2.3.4. Flavonoids

Flavonoids screening has been made using the following tests:

Test 1

In a 100 mL Erlenmeyer flask, 3 g of the plant powder was introduced to macerate in 50 mL of hydrochloric acid HCl overnight. The obtained mixture was filtered and then 10 mL of the obtained filtrate was made basic by adding a concentrated ammonium hydroxide NH_4OH solution. The appearance of a light yellow color indicates the presence of flavonoids.[20]

Test 2

We macerated 3 g of the milled vegetable powder in 50 mL of distilled water for 30 min and then filtered. We added to the resulted filtrate 1 mL of the mixture hydrochloric acid/butanol (v/v as 80/20) and 1 mL of isoamyl alcohol together followed by a few magnesium. The red-orange coloration indicates the presence of flavones while the pink-purplish coloration indicates the presence of flavanones. The red coloration indicates the presence of flavonols and flavanonols.[20]

2.3.5. Alkaloids

The alkaloids can be screened using the following methods:[21]

Method 1

3 g of the powdered plant was macerated in 50 mL of distilled water for 24 hours and then filtered. The filtrate is divided into 3 tubes to be tested by two reagents:

– The 1st tube

A few drops of Dragendorf reagent were added to the first tube, the formation of orange-red precipitate confirms the presence of alkaloids.

– The 2nd tube

We add a drop of a concentrated hydrochloric acid HCl and 3 drops of Bouchardât reagent, the appearance of red-brown precipitate is referred to the presence of alkaloids.

Method 2

6 g of the dry powder was put in 30 mL of 10 % sulfuric acid for 30-60 min. A few drops of Mayer's reagent were added which gives a yellow color in the presence of alkaloids.

– The 1st tube

We added a few drops of Dragendorf's reagent which gives a red-orange precipitate.

– The 2nd tube

We added a few drops of Bouchardât's reagent which gives a red-brown precipitate.

– The 3rd tube

It was subjected to UV light to detect the presence of alkaloids that give an intense blue fluorescence at $\lambda = 365$ nm.

2.3.6. Terpenes and Unsaturated Sterols

In this test, 5 g of the studied powder was macerated in 20 mL of chloroform $CHCl_3$ for a few minutes and then filtered. 10 mL of the filtrate, 1 mL of 10% sulfuric acid H_2SO_4 on the walls of the tube, the meeting point between the two phases gives a green color which indicates the presence of unsaturated sterols and terpenes.[20]

2.3.7. Coumarins

Method 1

3 g of the dried plant powder was macerated in 50 mL of ethanol for 24 hours and then filtered. 2 mL of the filtrate was mixed with 0.5 mL of sodium hydroxide NaOH. The obtained mixture was heated to boiling and then cooled. 4 mL of distilled water and a few drops of concentrated hydrochloric acid HCl were added to the final mixture before get the analytical sample. The presence of coumarins was confirmed by the fluorescence of the tested sample under a UV lamp.[22]

Method 2

A weight of 2 g of crushed dried plant material was inserted in 10 mL of CH_2Cl_2. The mixture was heated for a few minutes then filtered. The migration of this solution was carried out on a thin layer in the solvent: toluene/ethyl acetate (v/v as 93/7). After drying in a ventilated hood, the development was made using NH_3 under UV at 365 nm.[22]

2.3.8. Anthocyanins

1 g of the studied vegetable powder was mixed with 10 mL of distilled water in Buchner that was placed in a water bath at boiling temperature for 15 min and then filtered. The filtrate was treated with a few drops of hydrochloric acid HCl and a quantity of concentrated ammonium hydroxide NH_4OH, successively. The change of the initial extract color indicates the presence of anthocyanins.[20]

2.4. Quantification of Phenolic Compounds

2.4.1. Determination of Total Polyphenol Content

The determination of total polyphenols was carried out using the Folin Ciocalteu reagent. The measurements have been performed using a UV spectrophotometer at $\lambda = 765$ nm. The concentrations were determined by referring to the gallic acid calibration curve and then expressed in mg EAG/g MS.[23,24]

2.4.2. Determination of Total Flavonoid Content

The determination of the flavonoids amount was carried out by aluminum trichloride $AlCl_3$ which gives a yellow color as a positive result. The calculation of the concentrations was made from the equation for a standard curve for quercitrin.[25,26]

2.4.3. Determination of Total Tannin Content

The determination of tannins content was done by vanillin and hydrochloric acid which gives red color as a positive result. The calculation of the concentrations was made from the equation of a tannic acid calibration curve.[27,28]

2.4.4. Antibacterial Test

2.4.4.1. Strains

The antimicrobial activity of the obtained essential oil was tested towards ten micro-organisms including *Escherichia coli* (ATCC25922), *Listeria monocytogenes* (ATCC35152), *Bacillus cereus* (ATCC14579), *Pseudomonas aeruginosa* (ATCC27853), *Staphylococcus aureus* (ATCC25923), *Klebsiella oxytoca* (ATCC13182), *Klebsiella pneumonia* (ATCC14352), *Salmonella* (ATCC14028), *Enterococcus faecalis* (ATCC10541), and *Vibriocholerae* (ATCC33090). These micro-organisms were provided by the CRSTRA research center.

2.4.4.2. Diffusion Technique on Agar Medium

The sensitivity of strains to plant extracts has been carried out by *in vitro* the technique of diffusion in medium agar or disk method. This technique consists of introducing the germ under study at the surface of agar medium in Petri dishes of 4 mm thickness. Then we apply discs impregnated with the tested chemical substance. We can see the diffuse substances in the agar with a circular shape. After 18-24 hours, the discs appear surrounded by a zone of inhibition. In this study, Wattman paper was used in 6 mm disc. These must have a regular outline to give an easy measurable zone of inhibition. The discs, once prepared, are placed in a box of Petri dish containing 10 ml of distilled water and autoclaved for 20 min at 393 K.[29]

2.4.4.3. Preparation of the Inoculum

To prepare the inoculum, 4 to 5 colonies are scraped well isolated and perfectly identical from a culture of 6 p.m. on the isolation medium then discharged into water physiologically sterile. The bacterial suspension is then homogenized using a vortex and its turbidity is adjusted at 0.5 Mc Farland, *i.e.*, an optical density equal to 0.08 at 0.10; read at a wavelength of 625 nm corresponding to 108 CFU/mL.[29]

2.4.4.4. Seeding

This operation must be done within 15 min of the preparation of the inoculum. We dip a sterile swab in the bacterial suspension, and then float the swab over the entire agar surface, dry, of top to bottom, in tight streaks. The operation must be done two times by turning the Petri dish at an angle of 60° each time. After the immediate application of the discs, the boxes are left for 15 minutes at room temperature and then incubated at 310 K for 18 to 24 hours. It should be noted that each of the petri dishes contains four discs and using a micropipette, we put on each disc a different concentration. After incubation, the presence around the discs of a circular inhibition zone in which there is no growth of microorganisms denotes their sensitivity to this extract. The greater the inhibition, the more sensitive is the germ.[29]

2.5. DPPH Radical Scavenging Assay

The antioxidant activity was investigated using DPPH radical scavenging assay. 1 mL of different dilutions of the extracts was mixed with 250 µL of the methanol solution of DPPH in dry test tubes. After 30 min of incubation at room temperature and in the dark, the absorbance was measured at 517 nm using a UV-visible spectrophotometer.

– The negative control is composed of 1 mL of ethanol and 250 µL of the DPPH solution.

– The positive control is represented by a solution of a standard antioxidant (ascorbic acid). The absorbance of ascorbic acid was measured under the same conditions as that of the extract. The test was repeated three times. The anti-free radical activity was expressed by the reducing power of the methanol solution of DPPH• which was determined by applying the following formula:

$$PI = \frac{(AC - AE)}{AC} \times 100$$

where *PI* is the power of reduction in %; *AE* is the absorbance of the DPPH• solution in the presence of extract; *AC* is the absorbance of the DPPH• solution in the absence of extract.

The IC_{50} corresponds to the effective concentration of antioxidant necessary to remove 50% of the initial DPPH. The lower this value, the more the compound is a strong antioxidant.[30-32]

2.6. Statistical Analysis

All tests were performed in triplicate. Means and standard deviations are calculated with Microsoft Office 2013 Excel as well as histograms. The results of the dos-

age of phenolic compounds and those of biological activities have undergone a multi-factor analysis of variance (ANOVA) with SNK pair comparison using XLSTAT 2009 software.

3. Results and Discussion

3.1. Phytochemical Screening Results

Phytochemical screening represents an important preliminary analysis step allowing evaluating the chemical composition of the plant under investigation. Table 1 gives the results of the phytochemical screening of our studied plant leaves. As can be seen, the leaves of *Ocimum basilicum L.* contain different phytochemicals including essential oil, alkaloids, terpenes, unsaturated sterol, anthocyanins, gatechetical tannins, gallic tannins, sugars, flavones and unsaturated sterols. However, this plant material does not contain flavanones, flavanols, couramins and saponins.

Table 1. Phytochemical screening results of *Ocimum Basilicum L.* plant

Phytochemical compound	Result*
Essential oil	+
Catechetical tannins	+
Gallic tannins	+
Saponins	-
Sugars	+
Flavones	+
Flavanones	-
Flavanols	-
Alkaloids	+
Terpenes	+
Unsaturated sterols	+
Coumarins	-
Anthocyanins	+

* The qualitative results are expressed as + for the presence and – for the absence of phytochemicals.

3.2. Characteristics of the Obtained Essential Oil and the Phenolic Extracts

The characteristics of the phenolic extracts obtained by maceration and the essential oil obtained by hydrodistillation of *Ocimum basilicum L.* plant are summarized in Tables 2 and 3. The yields were determined from the weight of the extracts obtained, after evaporation at dry compared to the initial weight. The yields were calculated by the average of the three trials for each extraction. Our results revealed that the plant under probe is rich in essential oil and polyphenols and the best yield is that of the butanol extract.

Table 2. The properties of *Ocimum Basilicum L.* essential oil

Yield (%)	2.4
Aspect	Liquid
Color	Pale yellow
Flavor	Spicy
Odor	Gentle
Density	0.95
Refractive index	1.477
Rotating power	-0.46 °

Table 3. The properties of *Ocimum Basilicum L.* extracts

Dichloromethane extract	
Yield (%)	6.06
Aspect	Pasty
Color	Dark green
Ethyl acetate extract	
Yield (%)	7.34
Aspect	Solid
Color	Honey
n-butanol extract	
Yield (%)	10.06
Aspect	Crystals
Color	Brown

3.3. Total Polyphenol, Flavonoid and Tannin Contents

It can be seen that the *Ocimum basilicum L.* extracts contain phenolic compounds because they gave a blue color with the Follin Ciocalteu reagent. The total phenolic content is low compared to the dry residue because of the decomposition or the loss of these compounds during the drying process as reported in many papers. However, the dichloromethane extract is the richest extract in polyphenol (49.947 ± 3.907 mg GAE/g) followed by the ethyl acetate extract (36.902 ± 0.300 mg GAE/g), the lowest concentration of polyphenol (08.580 ± 0.610 mg GAE/g) was dedicated to the butanolic extract. The high level of total phenol should be recorded in the extract of ethyl acetate or dichloromethane; this could be due to the high content of the plants studied in compounds non-polar phenolic compounds such as phenolic acids and so on which are extractable by moderately polar solvents than polar ones such as tannins which are extractable by n-butanol. The dichloromethane extract is the richest (3.570 ± 0.261 mg TAE/g) followed by n-butanol extract (0.708 ± 0.009 mg TAE/g) and ethyl acetate extract is shown the poorest with a concentration of 0.468 ± 0.046 mg TAE/g. The flavonoid content varied from 1.239 ± 0.094 to 1.806 ± 0.192 mg QE/g for the dichloromethane and ethyl acetate extracts, respectively. The data are summarized in Table 4.

3.4. Antibacterial Activity Results

The evaluation of the antibacterial power of ten pathogenic bacterial strains as mentioned above was made *in vitro* by agar disk-diffusion method according to Clinical and Laboratory Standards Institute (CLSI) for bacteria testing in Mueller–Hinton growth medium. The diameters of the inhibition zones of the strains inoculated with the obtained essential oil and phenolic extracts are noted and expressed in mm as resumed in Tables 5 and 6.

The tested microbial strains were almost moderately sensitive to the effect of essential oil of the plant under investigation. According to reports, an extract is considered effective when the diameter of the inhibition zone is greater than 8 mm. In the current research, it is noticeable that the inhibition zone varies from 8.33 ± 0.44 to 16.33 ± 0.44 mm. However, the most sensitive strains are *Staphylococcus aureus* and *Klebsiella oxytoca* with a fairly large inhibition zone up to 16.33 ± 0.44 mm followed by *Listeria monocytogenes* (16.33 ± 0.44 mm), *Bacillus cereus* (14.33 ± ± 0.44 mm), *Enterococcus faecalis* (11 ± 0.44 mm), *Salmonella* (10.33 ± 0.44 mm) and even the strain *Vibriocholerae* showed low sensitivity (10 mm) as exhibited by Fig. 1. Similar results have been reported by Ngom *et al.* who demonstrated the sensitivity of the strains *Salmonella, Bacillus cereus,* and *Escherichia coli* against this essential oil, this effectiveness is due to its high amount in alcohols and phenolic compounds about 69 % of the total oil.[33]

Table 4. Contents of total polyphenol, flavonoids, and condensed tannin in *Ocimum Basilicum L.* extracts

Extracts	Total polyphenol content (mg GAE/g)	Total flavonoid content (mg QE/g)	Total tannin content (mg TAE/g)
dichloromethane	49.947 ± 3.907	1.239 ± 0.094	3.570 ± 0.261
ethyl acetate	36.902 ± 0.300	1.239 ± 0.094	0.468 ± 0.046
1-butanol	08.580 ± 0.610	1.631 ± 0.037	0.708 ± 0.009

Table 5. Antibacterial activity of *Ocimum Basilicum L.* essential oil against human pathogenic bacteria

Bacterial strain	Growth medium	Essential oil	T+ (p< 0.0001)
Bacillus cereus	Mueller–Hinton Agar	14.33±0.44	32.33 ± 2.22
Staphylococcus aureus	Mueller–Hinton Agar	16.33±0.44	35.00 ± 2.00
Listeria monocytogenes	Mueller–Hinton Agar	16.00±0.66	34.33 ± 3.11
Escherichia coli	Mueller–Hinton Agar	9,66±0.44	23.00 ± 2.00
Vibriocholerae	Mueller–Hinton Agar	10	24.00 ± 0.66
Salmonella	Mueller–Hinton Agar	10.33±0.44	25.33 ± 1.11
Pseudomonas aeruginosa	Mueller–Hinton Agar	6	23.33 ± 0.44
Klebsiella pneumonia	Mueller–Hinton Agar	7.00±0.44	22.00 ± 2.00
Klebsiella oxytoca	Mueller–Hinton Agar	16.33±0.44	25.66 ± 0.44
Enterococcus faecalis	Mueller–Hinton Agar	11.00±0.44	24.66 ± 0.88

T+: Gentamicin; an antibiotic with a broad spectrum of activity used as a positive reference.
p< 0.0001: significance level value.
Filter paper discs are 6 mm in diameter.

Table 6. Antibacterial activity of *Ocimum Basilicum L.* extracts against human pathogenic bacteria

Bacterial strain	Growth medium	Dich (p < 0.0001)	Acet (p< 0.0001)	n-but (p< 0.0001)	T+ (p< 0.0001)
Bacillus cereus	Mueller–Hinton Agar	9	8	6	32.33 ± 2.22
Staphylococcus aureus	Mueller–Hinton Agar	6	6	6	35.00 ± 2.00
Listeria monocytogenes	Mueller–Hinton Agar	7 ±0.66	6	6	34.33 ± 3.11
Escherichia coli	Mueller–Hinton Agar	6	6	6	23.00 ± 2.00
Vibriocholerae	Mueller–Hinton Agar	6	6	6	24.00 ± 0.66
Salmonella	Mueller–Hinton Agar	6	6	6.68 ± 0.44	25.33 ± 1.11
Pseudomonas aeruginosa	Mueller–Hinton Agar	7.33 ± 0.88	7.33 ± 0.44	7	23.33 ± 0.44
Klebsiella pneumonia	Mueller–Hinton Agar	6	6	6	22.00 ± 2.00
Klebsiella oxytoca	Mueller–Hinton Agar	6	6	6	25.66 ± 0.44
Enterococcus faecalis	Mueller–Hinton Agar	6	6	6	24.66 ± 0.88

T+: Gentamicin; an antibiotic with a broad spectrum of activity used as a positive reference.
p< 0.0001: significance level value.
Filter paper discs are 6 mm in diameter.

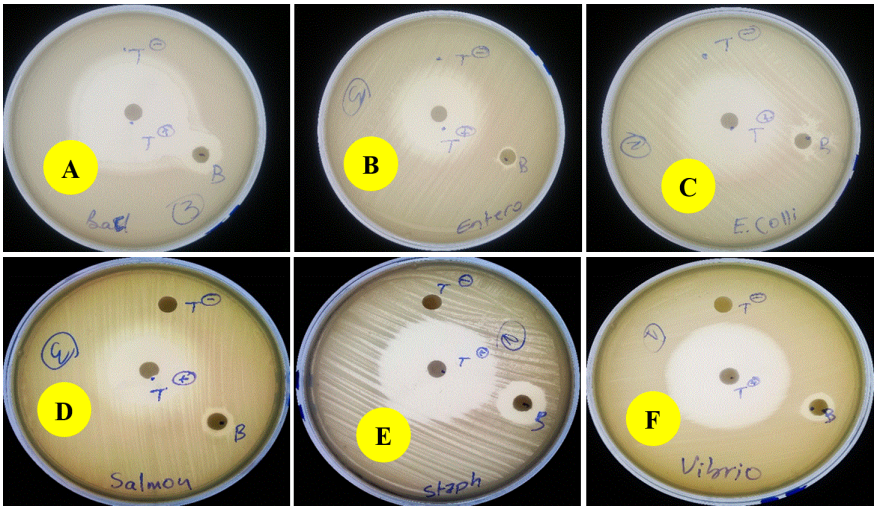

Fig. 1. Antibacterial activity of *Ocimum Basilicum L.* essential oil against the most sensitive bacterial species

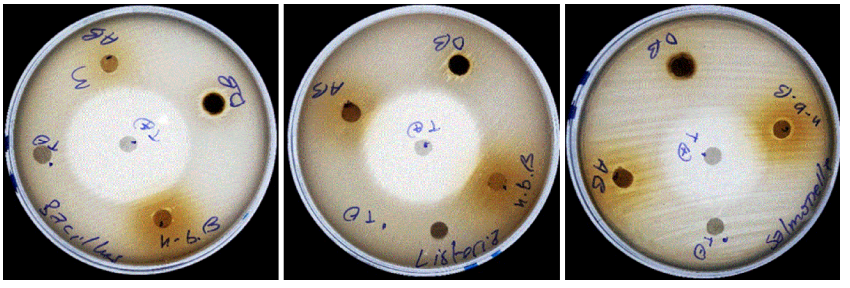

Fig. 2. Antibacterial activity of *Ocimum Basilicum L.* extracts against the most sensitive bacterial species

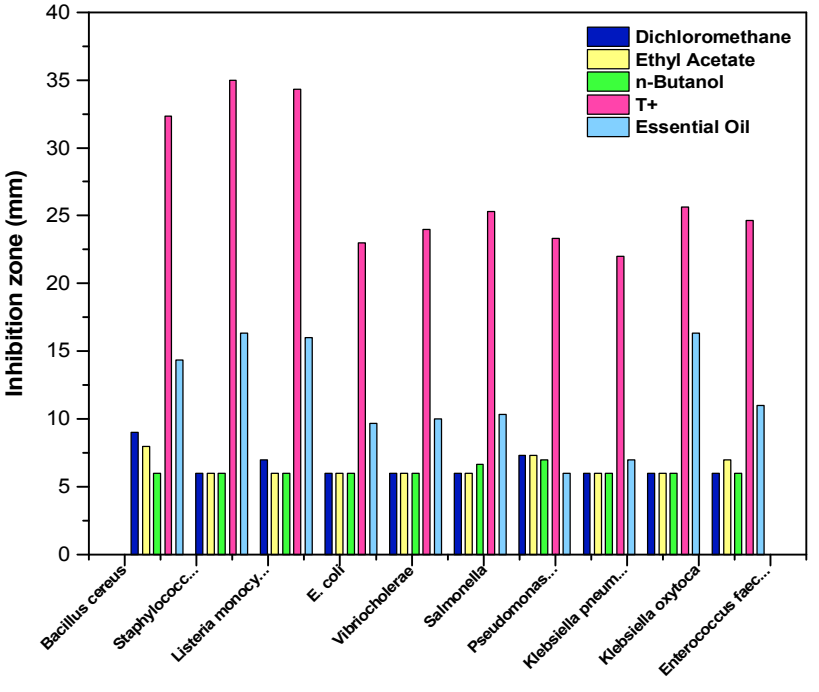

Fig. 3. Antibacterial activity of *Ocimum Basilicum L.* extracts against the studied bacterial strains

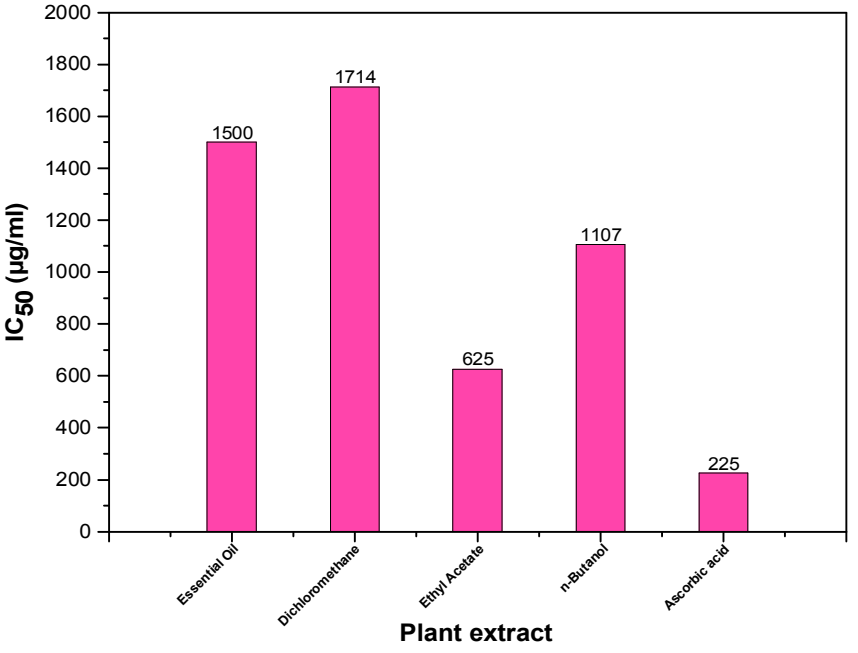

Fig. 4. Scavenging activity in the DPPH test with *Ocimum Basilicum L.* extracts

Regarding the results displayed in Table 6 and Figs. 2 and 3, it can be seen that the phenolic extracts of *Ocimum basilicum L.* have no effect on almost all of the selected bacterial strains except *Bacillus cereus* strain that showed a slight sensitivity towards the dichloromethane extract with a zone of inhibition of 9 mm, these results are similar for those obtained by Metali and Kerras[34] for an aqueous extract. Maidi *et al.* found similar results for his butanolic extract while the ethyl acetate extract exhibited significant inhibiting power against *Bacillus cereus*, *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas aeruginosa* species at a concentration of 3 mg/mL per disk. Plants contain many compounds with significant antimicrobial action. These constituents include phenolic compounds, flavonoids, coumarins, and triterpenoids,[35] the antimicrobial power of plant extracts is dependent on their concentration as well as their chemical composition. So, we conclude that this difference is due to the content of our plant in polyphenols, flavonoids and tannins, which is very far from the levels obtained in plants from other regions, in particular.

3.5. Antioxidant Activity Results

The antioxidant activity of the obtained extracts from the studied plant is determined by the DPPH radical scavenging method, this method is generally used to assess the ability of plant extracts to scavenge free radicals generated from the DPPH reagent. The antioxidant activity was evaluated for the different extracts as well as the standards control. This concentration of extract neutralizes 50 % of the free radical DPPH, the lower the IC_{50}, the more powerful the antioxidant potential of the extract. After 30 min of incubation of the DPPH-extract solution at different concentrations, the purple color (2,2 diphenyl-1picrylhydrazyl) turned to a yellow color in the extracts, this color change is due to the reduction of DPPH, which shows that the samples have a scavenger effect of the DPPH radical. The results of the antioxidant activity of the various extracts shown in Fig. 4 allowing observing that: (i) all the results obtained for the different phenolic extracts as well as the essential are higher than that recorded for ascorbic acid of 225 µg/mL and (ii) the most active fraction is that of ethyl acetate extract with an IC_{50} of 625 µg/mL thanks to its high amount of flavonoids and polyphenols that are responsible for the antioxidant activity followed by the extract of 1-butanol with a value of 1107 µg/mL; the extract of dichloromethane is the least active one having an IC_{50} of 1714 µg/mL.

4. Conclusions

1. The phytochemical screening results revealed the presence of various secondary metabolites such as essential oil, polyphenols, flavonoids, tannins, sterols, terpenes, saponins, *etc.*

2. The total polyphenol content obtained by Folin Ciocalteu method ranging from 49.97 ± 3.907 to 8.580 ± 0.61 mg GAE/g refers to dichloromethane and 1-butanol extracts.

3. The determination of flavonoids content revealed that it varies from 1.239 ± 0.094 to $1.806 \pm \pm 0.192$ mg QE/g for the dichloromethane and ethyl acetate extract, respectively.

4. The highest content of condensed tannins is dedicated to the dichloromethane extract (3.578 mg TAE/g) while the lowest content of these compounds is referred to the butanolic extract (0.708 mg TAE/g).

5. The phenolic extracts and the essential oil were tested for their capacity to scavenge DPPH radical. The obtained results showed that the majority of the phenolic extracts of the studied plant are active. The most active fraction is that of the ethyl acetate extract with an IC_{50} equals 625 µg/ml.

6. For essential oil, the zones of inhibition vary from 8.33 ± 0.44 to 16.33 ± 0.44 mm. The most sensitive strain is *Staphylococcus aureus* and *Klebsiella oxytoca* with a fairly large inhibition zone of up to 16.33 ± 0.44 mm followed by *Listeria monocytogenes, Bacillus cereus, Enterococcus faecalis, Salmonella* and even the strain *Vibriocholerae* exhibited low sensitivity.

7. Almost of extracts have no effect on the selected bacteria except that the *Bacillus cereus* strain showing a slight sensitivity towards the dichloromethane extract with an inhibition zone of 9 mm.

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ФІТОХІМІЧНИЙ СКРИНІНГ, АНТИБАКТЕРІАЛЬНА Й АНТИОКСИДАНТНА АКТИВНІСТЬ *Ocimum basilicum L.*, КУЛЬТИВОВАНОГО В БІСКРІ, АЛЖИР

Анотація. *Алжирська флора містить багато ароматичних рослин, які становлять великий терапевтичний інтерес завдяки їхнім біологічно активним вторинним метаболітам, що робить їх предметом наукового зацікавлення. У цій роботі досліджено Ocimum basilicum L. сім'ї Lamiaceae, культивований і зібраний у місті Біскра, розташованому на південному сході Алжиру. Фітохімічний скринінг виконанодля виявлення наявності флавоноїдів, танінів, кумаринів, ефірної олії й інших фітохімічних речовин. Визначено загальний вміст поліфенолів, флавоноїдів і танінів. Отримані ефірна олія й екстракти були предметом антибактеріальних та антиоксидантних досліджень. Наші результати підтвердили, що Ocimum basilicum L. містить велику кількість різноманітних потенційних фітохімічних речовин і що її можна використовувати як альтернативне природне джерело антиоксидантних і антибактеріальних компонентів.*

Ключові слова: *Ocimum basilicum L., Фітохімічний скринінг, антибактеріальний, антиоксидантний, Біскра.*