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ORIGINAL RESEARCH PAPER

ANTIBACTERIAL, ANTIOXIDANT ACTIVITIES AND MINERAL CONTENT FROM THE ALGERIAN MEDICINAL PLANTS

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Abstract: The present study evaluated the antioxidant and antibacterial activities of six aromatic plants, indigenous to the flora of the Algerian. The plants studied were *Thymus vulgaris, Artemisia absinthium, scorzonena undulata*, *Malva Parviflora, Marrubium vulgare* and *Cotula cinerea.* Extracts of these plants were analyzed for their total phenol compounds and antioxidant activity. The antimicrobial screening of the extracts was evaluated by agar disc diffusion method, the liquid broth microdilution was used to determine the minimal inhibitory concentration (MIC) and minimal bactericidal concentration (MBC). Essential oil of *A. absinthium* possesses highest antioxidant activity among all extracts tested and phenolic extract of *T. Vulgaris* highest of DPPH radical. Among the plants screened, essential oil of *A. absinthium* and *T. Vulgaris* phenolic extract showed the best antibacterial activity. The mineral contents were determined by an atomic absorption spectrophotometer. Ca, K, Na, Mg, Fe and Cu were found to be the most abundant elements among the plants studied.

Keywords: *antibacterial activity, antioxidant activity, mineral elements*

INTRODUCTION

Formation of free radicals may play an important role in the origin of life and biological evolution. Oxidation is essential to many living organisms for the production of energy to fuel biological processes [1]. However, the uncontrolled production of oxygen-derived free radicals is involved in the onset of many diseases such as cancer, rheumatoid arthritis, cardiovascular problems, and diabetes as well as in degenerative processes associated with aging.

The past decades have shown the increasing interest of medicine and the pharmaceutical industry in the products of processing of plant materials, which contain rich complexes of biologically active substances [2, 3]. For some medicinal plant preparations, the therapeutic effect is correlated with their ability to inhibit free-radical oxidation processes in biological systems [4].

Medicinal plants are a very important natural resource whos evaluation requires a perfect knowledge of the properties to develop. The medicinal activities of herbal depend on the presence of various bioactive agents belonging to different chemical classes [5]. Algeria is one of the African countries with a diverse flora, where numerous species are believed to possess curative properties, presenting diverse interests and constitutes an axis of scientific research more particularly in the field of natural substances [6].

Infectious diseases are still a major problem of the animal kingdom, especially human being and their recent emergence of multidrug resistance to antibiotics by pathogenic microbial strains due to the genetic manipulation. Thus importance has created an urgent need for the rapid and continued development of novel antimicrobial agents to replace the current regimens [7]. The terrestrial plants are used traditionally as medicine and constitute a potentially useful source as new and safer drugs for the treatment of microbial infections and other diseases [8].

Minerals are of critical importance in the diet, even though they comprise only 4–6 % of the human body. Major minerals are those required in amounts greater than 100 mg per day and they represent 1 % or less of bodyweight. These include calcium, phosphorus, magnesium, sulfur, potassium, chloride and sodium. Trace minerals are essential in much smaller amounts, less than 100 mg per day, and make up less than 0.01 % of bodyweight. Essential trace elements are zinc, iron, silicon, manganese, copper, fluoride, iodine and chromium. The major minerals serve as structural components of tissues and function in cellular and basal metabolism and water and acid – base balance [9, 10].

Air pollution and agricultural events affect the soil media, because heavy metals are adsorbed via organic matters, minerals and carbonates that are in the soil [11]. The plants can be considered as an indicator for environmental conditions due to the use of water, soil, and air. The heavy metal accumulation in plants shows variability depending on the metal type, plant species and plant tissues. Some heavy metals such as Co, Cu, Fe, Mn, Ni and Zn are important for normal growth of plant at a certain level. Higher levels are toxic and they cause the plant to die.

The aim of the present study was to determine the total phenolic and flavonoid contents and to evaluate the antioxidant and antibacterial activities of six selected Algeria medicinal plants (phenolic extract and essential oils). In addition, this study also to

determine the content of both the various essential (Na, K, Ca and Mg) and macro (Fe, Zn, CU, Mn, Ni, Pb and Cd) elements.

MATERIAL AND METHODS

Chemicals and reagents

The chemicals DPPH (2,2-diphenyl-1-picrylhydrazyl radical), ABTS (2,2-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid radical cation), and Folin-Ciocalteu phenol reagent were purchased from Sigma Chemical Co. (St. Louis, MO, USA). BHT (2,6-di-*tert*-butyl-4-methylphenol), BHA (*tert*-butyl-4-hydroxyanisol), gallic acid, *β*-carotene and rutin were purchased from Aldrich Chemicals Co., U.S.A. All other chemicals were of analytical grade.

Plant material

The aerial parts of *Thymus vulgaris, Artemisia absinthium, Scorzonena undulata*, *Malva Parviflora, Marrubium vulgare* and *Cotula cinerea* were collected at flowering stage in April 2017 from Laghouat city (Algerian Saharan). The plant specimens were identified at the laboratory of Process Engineering, University of Laghouat. Vouchers of the specimens are deposited.

Extraction of essential oils

Essential oils were extracted by hydrodistillation of the dried plant material (100 g of sample in 500 mL of distilled water) for 4 h using a Clevenger-type apparatus. The oils were dried over anhydrous sodium sulfate and stored at 4 °C in amber glass vials until analysis [12].

Preparation of plant extracts

Antioxidant compounds were extracted and analyzed by using the method reported by Chong *et al.* [13]. Briefly, dried plant material (0.5 g) was mixed with of Ethanol/water (80 : 20 v/v) + 1 % HCl (10 mL), sonicated at 20 °C for 15 min, then left for 24 h at 4 °C. The extract was sonicated for 15 min, and then centrifuged at 15000 rpm for 10 min. The supernatant was collected and used to analyze total phenolic content and antioxidant capacity.

Total phenolic content

Total phenolic compounds content was assayed using the Folin – Ciocalteu reagent, following Gao *et al.* [14]. An aliquot (0.1 mL) of diluted sample extract was added to 0.2 mL of the Folin – Ciocalteu reagent and 2 mL of water. The mixture was shaken and allowed to stand for 3 min, before addition of 1 mL of Na_2CO_3 (20 %). After incubation in dark for 1 h, the absorbance at 760 nm was read versus the prepared blank. Total

phenolic content of plant parts was expressed as milligrams of gallic acid equivalents per gram of dry weight (mg GAE / g DW).

Total flavonoids

Flavonoid content was identified as described by Bakchiche *et al*. [15]. Briefly, 1 mL of 2 % $AlCl_3$ solution was added to 1 mL of sample or standard. After 1 h at room temperature, the absorbance was measured at 420 nm. Rutin was used as standard for the construction of calibration curve.

Antioxidant activity

DPPH radical-scavenging capacity

The DPPH radical-scavenging activity was determined using the method proposed by Brand-Williams *et al*. [16]. Methanol as negative control and extract without DPPH as blank. Results were expressed as percentage reduction of the initial DPPH absorption in relation to the control. The concentration of extract that reduced the DPPH color by 50 % (IC_{50}).

$ABTS^{\bullet+}$ free-radical scavenging activity

ABTS radical scavenging activity was measured using the method of Dorman and Hiltunen [17] with slight modifications. ABTS was dissolved in water at a concentration of 7 mM. ABTS radical cations ($ABTS^{\bullet+}$) were produced by combining ABTS stock solution with 2.45 mM potassium persulfate. The mixture was left in the dark at room temperature for 12 – 16 h before use. To evaluate the antioxidant activity of each sample extract, the $ABTS^{\bullet+}$ solution was distilled with ethanol until an absorbance of 0.700 (± 0.020) at 734 nm was achieved. After adding 1.485 mL of the distilled $ABTS^{\bullet+}$ solution to 15 µL of each sample extract, the mixtures were left in the dark for 6 min at room temperature. Absorbances were then measured at 734 nm using a spectrophotometer (Uvikon XS, Bio-Tek Instruments, Saint Quentin Yvelines, France).

β-carotene bleaching assay

The *β*-carotene bleaching assay was determined using a Dapkevicius *et al.* [18] method by measuring the inhibition of hydroperoxides formed from linoleic acid oxidation. The linoleic acid solution was prepared by mixing 0.5 mg *β*-carotene in 1 mL $CHCl_3$, then 25 µL linoleic acid and 200 mg tween 40. $CHCl_3$ was totally evaporated using vacuum evaporator and 100 mL of distilled water was added, followed by shaking. Finally, 3 mL of the above solution was transferred to the test tube and 0.2 mL of each extract was added. Same procedure as above was followed for two blanks, one containing antioxidants (BHT or BHA) as a positive control and the other with the same volume of ethanol instead of the extracts. All samples were put into a water bath for 2 h at 50 °C. The absorbance value was measured at 470 nm. Measurement of observance was continued until the color of *β*-carotene disappeared; every 30 min up to 2 the percentage inhibition.

Total antioxidant capacity

The antioxidant activity was determined by the phosphomolybdenum method of Prieto *et al.* [19]. 0.3 mL of each extract was mixed with 3 mL of reagent solution (0.6 M sulfuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate). The tubes were capped and incubated in water bath at 95 °C for 90 min. After the samples had cooled to room temperature, the absorbance of the green phosphomolybdenum complex was measured at 695 nm. In the case of the blank, 0.3 mL of ethanol was used in place of sample. The antioxidant activity was determined using a standard curve with ascorbic acid solutions as the standard. The mean of three readings was used and the reducing capacity of the extracts was expressed as mg of vitamin C equivalents mmol of vitamin C / g dry weight).

Ferric-reducing antioxidant power assay (FRAP)

The reducing power was determined by ferric chloride method of Oyaizu [20]. Briefly, 1 mL of extract was mixed with 2.5 mL of 200 mM sodium phosphate buffer (*p*H 6.6) and 2.5 mL of 1 % potassium ferricyanide solution, followed by incubation at 50 °C for 20 min. 2.5 mL of 10 % trichloroacetic acid (TCA) was then added to it and the mixture centrifuged at 3000 x g for 10 min. 2.5 mL aliquot was carefully withdrawn from upper layer and mixed with 2.5 mL distilled water and 0.5 mL of 0.1 % $FeCl_3$ solution. Absorbance of the mixture was recorded at 700 nm against a reagent blank.

Antimicrobial activity

Tested microorganisms

Antimicrobial activity was tested against a panel of microorganisms: Gram-positive bacteria [*Staphylococcus aureus* ATCC 25923 (*S. aureus*), *Streptococcus pneumoniae* ATCC 29212 (*S. pneumoniae*), *Rhodococcus equi* ATC 6939 (*R. equi*)], Gram-negative bacteria [*Escherichia coli* ATCC 25922 (*E. coli*), *Pseudomonas aeruginosa* ATCC 27853 (*P. aeruginosa*), *Klebsiella pneumoniae* ATCC 700603 (*K. pneumoniae*)]. All microorganisms were graciously supplied from stock cultures of the Regional Veterinary Laboratory of Laghouat Algeria. Bacterial strains were cultured in Muller–Hinton agar. All microbial strains were incubated for 24 h at 37 °C.

Agar diffusion method

The antimicrobial activity of the extracts was determined by the disk diffusion method which is based on the spread of antimicrobial compound in solid medium [21]. The Mueller – Hinton agar was poured in sterile petri dishes (90 mm diameter). The paper discs (6 mm diameter) that were impregnated with 20 µL of each essential oil and phenolic extracts were placed on the inoculated agar surface. Petri dishes were allowed to stand for 30 min at room temperature before incubation at 37 C for 24 h. The effect of essential oils and phenolic extracts were reflected by the appearance around disc with a transparent circular zone corresponding to the absence of growth. The diameter of inhibition zone was measured in mm. The larger the diameter of the area the more susceptible the strain [22].

Determination of Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC)

MIC was determined as the lowest concentration of extracts that inhibited visible growth (turbidity) of the test organisms after 24 h [23]. MIC was investigated for the bacterial strains, being sensitive to the plant extracts in the disc diffusion assay. MIC was carried out using tube dilution method as described by the Clinical and Laboratory Standards Institute (NCCLS, 2000). 200 µL of each dilution of extract is added in test tubes containing a volume of 2.8 mL of nutrient broth inoculated with the bacterial suspension to obtain a final charge of 106 cells / mL. A tube without extract containing 200 µL of DMSO and inoculum was used as a control. Further, they were incubated at 37 °C for 15 to 24 h [24]. MBC is the lowest concentration of an antimicrobial agent needed to kill 99.9 % of the initial inoculums. MBC of the plant extract was determined following the method of Spencer and Spencer [25]. One milliliter of sample from the test tubes used in MIC, which did not show any visible growth, was streaked out on nutrient agar plates to determine the minimum concentration required to kill pathogens. The concentration of extract indicating a bactericidal effect after 24 h of incubation at 37 °C was regarded as MBC.

Mineral analysis

Dried samples (0.5 g) were digested for 3 h at a temperature below 130 °C in 65 % HNO_3 (5 mL) by using a multi-sample digestion block (Block Digest 20, Selecta, Barcelona, Spain) [26]. Samples were cooled to room temperature, transferred to volumetric flasks, and then diluted with ultra-high-purity deionized water. Samples were stored at 4 °C until analysis. Ca, K, Na, Mg, Fe, Ni, Cu, Co, Zn, Mn, Cd and Pb in previously mineralized samples were analyzed with an atomic absorption-emission spectrometer (Solaar 969, Unicam Ltd., Cambridge, UK). K and Na were analyzed by atomic emission and the other elements were analyzed by atomic absorption. The instrument was calibrated with certified standards. At least two reagent blanks were included in each batch of samples to assess the precision and accuracy of the chemical analysis. Calibration curves were used to quantify these elements and showed good linearity ($R^2 \geq 0.997$). Analyses were run in triplicate.

Statistical analysis

Experimental results are presented as the mean ± standard error of three parallel measurements. Statistical analyses were performed by one-way ANOVA, followed by Tukey multiple comparisons test. A difference was considered to be statistically significant when $p < 0.05$.

RESULTS AND DISCUSSION

Determination of total phenolic content and total flavonoids

The total phenolic content as determined by Folin - Ciocalteu method were expressed in milligrams equivalent of gallic acid per gram of dry weight (mg GAE / g DW).

Flavonoids content was expressed as milligrams equivalent of rutin per gram of dry extract (mg RE / g DW). Total phenolic and flavonoid contents are shown in Tables 1.

Table 1. *Total phenol compounds and flavonoids content in the plant extracts*

Ethanolic extracts	Total phenolic content (mg GAE / g DW)	Total flavonoids content (mg ER / g DW)
T. Vulgaris	22.03 ± 0.87^{a}	11.18 ± 0.76^{a}
A. absinthium	15.01 ± 0.20^{f}	4.44 ± 0.42^{e}
S. undulata	19.73 ± 0.30^{b}	6.20 ± 1.18^{c}
M. Parviflora	17.58 ± 0.17^{d}	7.25 ± 1.70^{b}
M. vulgare	18.30 ± 0.24^{c}	3.06 ± 0.23^{f}
C. cinerea	16.42 ± 0.19^{e}	5.07 ± 0.30^{d}

Flavonoid content is expressed as mg of rutin-equivalents per g of dry extract. Polyphenol content is expressed as mg of gallic acid-equivalents per g of dry extract. Results are averages ± SE (n = 3). Different letters above the average bars denote significant differences at $p < 0.05$ - Tukey's test

From these results, the *T. Vulgaris* extract showed high phenolic and flavonoid compounds (22.03 ± 0.87 mg GAE/g and 11.18 ± 0.76 mg RE /g respectively) followed by *S. undulata* extract (19.73 ± 0.30 mg GAE/g of phenolics and 6.20 ± 1.18 mg RE/g of flavonoids), *M. vulgare* extract (18.30 ± 0.24 mg GAE/g and 3.06 ± 0.23 mg RE/g respectively) and *M. parviflora* extract (17.58 ± 0.17 mg GAE/g of phenolics and 7.25 ± 1.70 mg RE/g of flavonoids), whereas the lowest levels phenolic and flavonoid content in *C. cinerea* extract (16.42 ± 0.19 mg GAE/g and 5.07 ± 0.30 mg RE/g respectively) and *A. absinthium* extract (15.01 ± 0.20 mg GAE/g and 4.44 ± 0.42 mg RE/g respectively). Obtained results indicate an existence of some phenol rich species, characterized by the high presence of secondary metabolites of such class type.

Milella *et al*. [27] indicated that total phenolic content of methanol/water extract of *S. undulata* roots is about 80.7 mg GAE/g of extract, which seems to be more important than the results obtained in the present study. The phenolic content and flavonoid were observed in extracts from *T. vulgaris* (22.03 ± 0.87 mg GAE/g and 11.18 ± 0.76 mg CER/g respectively), which were higher than the studies of Zeghad and Merghem [28], that showed 9.07 ± 0.002 $\mu g \cdot mL^{-1}$ Tannic Acid Equivalent of dry material of the phenolic content and 8.56 ± 0.001 $mg \cdot g^{-1}$ Quercetin Equivalent of dry material of flavonoid, while Prabhakaran and Shwetha [29], indicating that phenolic content of total (91.42 ± 0.89 mg TAE/g extract) and the flavonoid (55.01 ± 0.42 mg RE/g extract) were higher than our studies. In previous studies [30], the reported phenolic content of *M. parviflora* (204.4 mg GAE/g DW) was lower compared to our results.

Variation in the amounts of phenolic compounds could be attributed to several reasons. The solubility of phenolic compounds is governed by the type of solvent used, the degree of polymerization of phenolics, as well as by the interaction of phenolics with other food constituents and formation of insoluble complex. Indeed, the phenolic contents of a plant depend on intrinsic (genetic) and extrinsic (environmental, handling and storage) factors [31, 32]. Plant materials rich in phenols are increasingly being used in the food industry because they retard oxidative degradation of lipids and improve the quality and nutritional value of food [33].

Antioxidant activity

DPPH radical-scavenging capacity

The DPPH method was introduced nearly 50 years ago by Blois [34] and it is widely used to test the ability of compounds to act as free radical scavengers or hydrogen donors, and to evaluate antioxidant capacity. The parameter IC_{50} (efficient concentration value), is used for the interpretation of the results from the DPPH method and is defined as the concentration of substrate that causes 50 % loss of the DPPH activity (color). IC_{50} values of phenolic extract of *T. vulgaris, M. parviflora, A. absinthium, S. undulata*, *M. vulgare,* essential oil of *A. absinthium,* phenolic extract of *C. cinerea* and essential oil *of C. cinerea* are (37.85 ± 8.51, 86.46 ± 16.06, 129.51 ± 4.40, 145.09 ± 13.08, 224.25 ± 45.71, 260.85 ± 11.41, 695.02 ± 147.28 and 1612.06 ± 163.64 $\mu g \cdot mL^{-1}$ respectively).
Though the antioxidant potential of extracts was found to be low ($P < 0.05$) than those of gallic acid, BHA, ascorbic acid and BHT (0.49 ± 0,04, 1.08 ± 0,006, 1.15 ± 0,06 and 1.27 ± 0.26 $\mu g \cdot mL^{-1}$ respectively). The study revealed that phenolic extract of *T. Vulgaris* have prominent antioxidant activity while essential oil of *A. absinthium* was established to have the lowest antioxidant capacity (Table 2). The results show that the scavenging effects of samples on DPPH radical and were in the following order: PhE of *T. Vulgaris* > PhE of *M. Parviflora* > PhE of *A. absinthium* > PhE of *S. undulata* > PhE of *M. vulgare* > EO of *A. absinthium* > PhE of *C. cinerea* > EO of *C. cinerea.* As the IC_{50} concentration and the antioxidant capacity have inversely proportional values.
Our results of *S. undulata* were within the range of the values found in the literature for the inhibition of DPPH with plant extracts [35] and were lower than those recently reported for EO of *A. absinthium* (IC_{50} = 5.85 $\mu g \cdot mL^{-1}$) [36]. The IC_{50} values found for PhE of *T. Vulgaris* were higher than those found for Köksal *et al.* [37] (IC_{50} = 12.1 $\mu g \cdot mL^{-1}$). The scavenging potency of phenolic extract of *M. Parviflora* these later was higher than found by Afolayan *et al.* [38] with an IC_{50}=1.48 $\mu g \cdot mL^{-1}$, as well as considerable antioxidant effect in DPPH assay (IC_{50} approx. at 9.3 ± 0.01 $\mu g \cdot mL^{-1}$) [30].

$ABTS^{\bullet+}$ cation radical scavenging activity

$ABTS^{\bullet+}$ assay is an excellent tool to determine the antioxidant activity of hydrogen-donating antioxidants (scavenging aqueous phase radicals) and of chain breaking antioxidants (scavenging lipid peroxyl radicals). ABTS radical scavenging assay involves a method that generates a blue/green $ABTS^{+}$ chromophore via the reaction of oxidation the ABTS and potassium persulfate, its reduction in the presence of hydrogen-donating antioxidants is measured spectrophotometrically at 745 nm. In $ABTS^{\bullet+}$ cation radical scavenging method. The results of IC_{50} the six phenolic extracts and oil essential of *A. absinthium* and *C. cinerea* were presented in Table 2. IC_{50} is a parameter widely used to measure antioxidant activity. As the IC_{50} value of the extract decreases, the free radical scavenging activity increases. The antioxidant activity of the samp extracts ranges from 3.96 ± 0.85 to 1326.02 ± 178.19 $\mu g \cdot mL^{-1}$ and the values are significantly ($p < 0.05$) different. Among them, the essential oil of *A. absinthium* exhibited higher (3.96 ± 0.85 $\mu g \cdot mL^{-1}$) and comparable activity on a par with the standard antioxidants BHA and BHA, followed by phenolic extract of *T. vulgaris* (37.38 ± 11.64 $\mu g \cdot mL^{-1}$), then phenolic extract of *M. parviflora* (305.04 ± 108.14 $\mu g \cdot mL^{-1}$), *A. absinthium* (308.07 ± 183.22 $\mu g \cdot mL^{-1}$), phenolic extract of *S. undulata* (457.87 ± 15.37 $\mu g \cdot mL^{-1}$),

finally phenolic extract of *M. vulgare* (1165.04 ± 21.32 µg·mL^{-1}) and essential oil of *C. cinerea* (1326.02 ± 178.19 µg·mL^{-1}).
In comparison with another study, the ABTS activity of *Artemisia capillaris* is about 0.27 ± 0.02 mM TE/g DW, *Artemisia argyi* (0.24 ± 0.007 mMTE/g DW) and *Artemisia apiacea* (0.05 ± 0.002 mMTE/g DW) [39], which is much lower than the results obtained in the present study.
In one study by Baykan et al. 2012 [40], it has been revealed that TEAC value the Essential oils *A. absinthium* from western Anatolia was found as 2.87 ± 0.17 mM Trolox, Altunkaya *et al.* [41], also reported a TEAC value as 3.4 mM Trolox activity.
In an earlier study, the IC_{50} value of the ABTS radical scavenging of ethanol extract of *T. vulgaris* was 54.08 µg·mL^{-1} [37] and in another study, the IC_{50} value of the ABTS activity of methanol extract the *M. Parviflora* obtained was found to be 94.3 ± 0.03 µg·mL^{-1} [38]. Essential oil of *A. absinthium* possessed strong ABTS scavenging activity an observation that is supported by other researchers [42].

β-carotene bleaching assay

The linoleic acid-free radical attacks *β*-carotene bleaching assay and hence it loses chromophore and orange color. The *β*-carotene principle is based on the loss of the orange color of *β*-carotene due to its reaction with radicals, which are formed due to oxidation of linoleic acid in an emulsion. The rate of *β*-carotene bleaching can be decreased in the presence of antioxidants [43]. This principle is used in the determination of antioxidant activity of six plant extracts in comparison with BHT and BHA. The IC_{50} values of different extracts of six plants are shown in Table 2. The extracts with the lowest value, have the most powerful anti-radical activity. According to Table 2, all phenolic extracts and essential oil of *A. absinthium*, *C. cinerea* are endowed with antioxidant activity, which varies from one extracted another. Essential oil of *C. cinerea* exhibit highest activity (IC_{50} = 61.77 ± 13.23 µg·mL^{-1}) but considerably lower than BHT and BHA activity ($p < 0.01$). Present results showed that the β-carotene bleaching assay of samples can be ranked as BHT > BHA > EO of *C. cinerea* > PhE of *C. cinerea* > EO. *A. absinthium* > Ph E of *M. parviflora* > PhE of *T. vulgaris* > PhE of *A. absinthium* > PhE of *M. vulgare* > PhE of *S. undulata.*
According to Taherkhani *et al.* [36], in *β*-carotene-linoleic acid test system, oxidation of linoleic acid was effectively inhibited by *A. absinthium* oil (58.56 ± 2.5 %, 0.625 µg·mL^{1} of essential oil). Differences in antioxidant activity of plants extracts reflect their phenolic extracts and essential oil content (Table 2). It is important to add that *β*-carotene bleaching assay only provides an indication of the level of lipophilic compounds because the *β*-carotene bleaching test is similar to an oil-in-water emulsion system [44].
The *β*-carotene-linoleate bleaching (BCB) method employs an emulsified lipid and therefore applicable specially to investigate lipophilic antioxidants such as the antioxidant activity of essential oils. If polar compounds such as ascorbic acid, rosmarinic acid, etc., are tested by the BCB method, they would be considered weak antioxidants [45].
Antioxidant activity in the *β*-carotene assay, the oxidation of linoleic acid generates peroxyl free radicals due to the abstraction of a hydrogen atom from diallylic methylene groups of linoleic acid [46]. The free radical will then oxidize the highly unsaturated

β-carotene. The presence of antioxidants in the plant extracts will minimize the oxidation of β-carotene by hydroperoxides.

Table 2. *Antioxidant activity of the Phenolic extracts for six plants and essential oils of A. absinthium and C. cinerea, expressed in IC_{50} (µg/ mL) for DPPH and β-Carotene bleaching assay and ABTS assay, VCEAC (vitamin C equivalents mmol of vitamin C/ g dry weight) for Phosphomolybdate Assay*

Plants	Total antioxidant capacity	DPPH radical	ABTS radical	*β*-carotene bleaching assay
Ph. extract of *C. cinerea*	0.03 ± 0.06^{d}	695.02 ± 147.28^{g}	923.30 ± 17.84^{f}	88.80 ± 17.73^{c}
E. oil of *C. cinerea*	0.23 ± 0.02^{a}	1612.06 ± 163.64^{h}	1326.02 ± 178.19^{g}	61.77 ± 13.23^{c}
Ph. extract of *S. undulata*	0.02 ± 0.005^{e}	145.09 ± 13.08^{e}	457.87 ± 15.37^{e}	374.32 ± 50.43^{f}
Ph. extract of *M. vulgare*	0.05 ± 0.004^{c}	224.25 ± 45.71^{f}	1165.04 ± 21.32^{g}	370.96 ± 54.92^{f}
Ph. extract of *A. absinthium*	0.04 ± 0.006^{c}	129.51 ± 4.40^{e}	308.07 ± 183.22^{d}	327.94 ± 26.35^{f}
Ph. extract of *T. vulgaris*	0.10 ± 0.007^{b}	37.85 ± 8.51^{c}	37.38 ± 11.64^{c}	$286.39 \pm 44.84^{e,f}$
Ph. extract of *M. parviflora*	0.03 ± 0.003^{d}	86.46 ± 16.06^{d}	305.04 ± 108.14^{d}	178.19 ± 110.69^{d}
E. oil of *A. absinthium*	0.25 ± 0.05^{a}	260.85 ± 11.41^{f}	3.96 ± 0.85^{b}	123.11 ± 5.95^{d}
BHT	–	1.27 ± 0.26^{b}	1.89 ± 0.40^{a}	0.49 ± 0.02^{a}
BHA	–	1.08 ± 0.006^{b}	1.03 ± 0.06^{a}	7.32 ± 0.96^{b}
Ascorbic acid	–	1.15 ± 0.06^{b}	–	–
Gallic acid	–	0.49 ± 0.04^{a}	–	–

Values are presented as mean ± SE (n = 3). Different letters above the average bars denote significant differences at $p < 0.05$ - Tukey's test

Total antioxidant capacity

The antioxidant capacity of the plant extracts was measured spectrophotometrically through phosphomolybdenum method, based on the reduction of Mo (VI) to Mo (V) by the test sample and the subsequent formation of green phosphate/Mo (V) compounds with maximum absorption at 695 nm. The results indicated that the VCEAC (vitamin C equivalents mmol of vitamin C/ g dry weight) values six phenolic extracts and the oil essential of *A. absinthium* and *C. cinerea* ranged from 0.25 ± 0.05 to 0.02 ± 0.005 $mmol \cdot g^{-1}$ (Table 2). Here again, essential oil of *A. absinthium* and *C. cinerea* displayed maximum total antioxidant activity (TAA) of 0.25 ± 0.02, 0.23 ± 0.05 $mmol \cdot g^{-1}$ respectively, followed by the phenolic extract of *T. vulgaris* (0.10 ± 0.007 $mmol \cdot g^{-1}$), the phenolic extract of *M. vulgare* (0.05 ± 0.004 $mmol \cdot g^{-1}$) and phenolic extract of *A. absinthium* (0.04 ± 0.006 $mmol \cdot g^{-1}$) and lower total antioxidant activity (TAA) of phenolic extracts of *M. Parviflora, C. cinerea* are 0.03 ± 0.003, 0.03 ± 006 $mmol \cdot g^{-1}$ respectively and finally, phenolic extracts of *S. undulata* is 0.02 ± 0.005 $mmol \cdot g^{-1}$.

The present study demonstrated that essential oil of *A. absinthium* exhibited the highest antioxidant capacity due to presence of bioactive molecules for phosphomolybdate reduction.

Phosphomolybdate is another important in vitro antioxidant assay to access the total antioxidant capacity of the plant extract. The assay principal follows the conversion of Mo (VI) to Mo (V) by extract or the compound which possess antioxidant potential resulting in green phosphate Mo (V). The electron/hydrogen donating pattern of antioxidants depends upon its structure and series of redox reactions occurring in the activity [47] .
In an earlier study, Köksal *et al.* [37] reported that the total antioxidant activity of ethanol extract of *T. vulgaris* was 64.4 %.

Reducing Capacity
A strong reducing power was noted for essential oil of *A. absinthium* (A = 2 at C = 46.57 µg·mL^{-1}, A = 1.60 at C = 11.64 µg·mL^{-1}, A = 1.33 at C = 2.91 µg·mL^{-1}, A = 0.41 at C = 0.36 µg·mL^{-1}) and phenolic extract of *T. vulgaris* (A = 1.53 at C =46.57 µg·mL^{-1}, A = 0.65 at C = 11.64 µg·mL^{-1}, A = 0.37 at C = 2.91 µg·mL^{-1}, A = 0.19 at C = 0.36 µg·mL^{-1}) the values are comparable with the standard antioxidant ascorbic acid, BHA and BHT. Much lower reducing power was assisted to essential oil of *C. cinerea* (A = 0.15 at C = 46.57 µg·mL^{-1}, A = 0.13 at C = 11.64 µg·mL^{-1}, A = 0.10 at C= 2.91 µg·mL^{-1}, A = 0.07 at C = 0.36 µg·mL^{-1}). The reducing power of the samples was in the following order: BHA > BHT > As. A > EO. *A. absinthium* > Ph E of *T. vulgaris* > PhE of *S. undulata* > PhE of *A. absinthium* > PhE of *M. parviflora* > PhE of *M. vulgare* > PhE of *C. cinerea* > EO of *C. cinerea* (Figure 1). The observed trend in the reducing power might be as a result of that obtained for total phenolic content in which *T. vulgaris* contained more phenolic compounds than other plants, as the phenolic content increases the ability to reduce the ferricyanide complex of Fe^{3+} to the ferrous (Fe^{2+}) form.

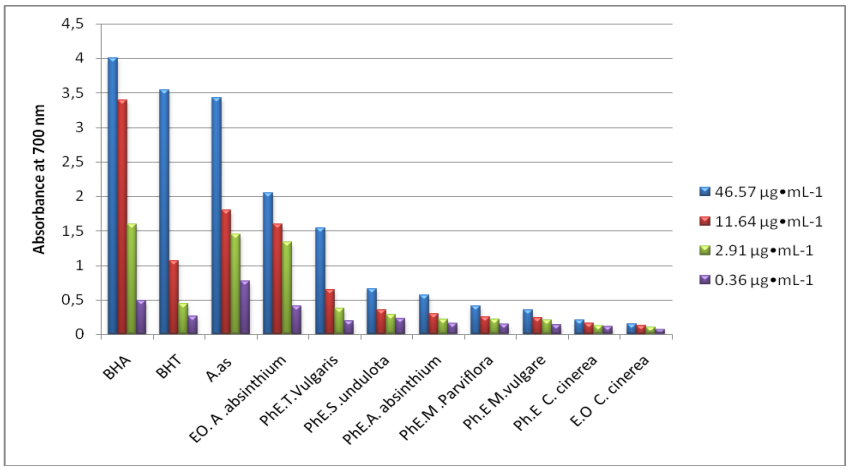

Figure 1. *Reducing power of different extracts from the six medicinal plants*

Taherkhani *et al.* [36] reported that the ferric reducing power of the essential oil of *A. absinthium* was determined 10.67 ± 0.45 gallic acid equivalent (mg·g^{-1}). Moreover, the absorbance of reducing powers of 10 µg·mL^{-1} was 1.88 displayed by ethanol extract of *T. vulgaris* is the highest to our results [36].

The antioxidants present in the extracts of essential oil of *A. absinthium* and phenolic extract of six plants studies caused their reduction of Fe^{3+}/ ferricyanide complex to the ferrous form, and thus proved the reducing power.

Antibacterial activity of the plant extracts

Diameter of zone of inhibition

In the present study, the antimicrobial activity of six medicinal plants was evaluated against *E. coli, P. aeruginosa* and *K. pneumoniae* (Gram-negative), *S. aureus, S. pneumoniae* and *R. equi* (Gram-positive). Most of the ethanolic extracts exhibited low antimicrobial activity against these pathogenic microorganisms, and the results were summarized in Table 3.

Table 3. *Diameter of zone of inhibition of different phenolic extracts, against pathogenic bacteria*
Different superscript letters denote significant differences at p <0.05 - Tukey's test

Plants extracts		Zone of inhibition (mm)						
		Ph. extract Of *T. vulgaris*	**Ph. extract of *A. absinthium***	**Ph. extract of *S. aundulota***	**Ph. extract of *M. parviflora***	**Ph.extract of *M. vulgare***	**Ph.extract of *C. cinerea***
Gram -	*E.coli* ATCC 25922	8.71±0.25 ^{b,c}	11.28±0.55^{a}	9.28±0.23^{b}	7.42±0.29^{c}	7.99±0.19^{c}	6.00^{d} (NA)
	P.aeruginosa ATCC 27853	8.79±0.42^{a}	7.26±0.22^{b}	8.51± .54^{a}	7.77±0.15^{b}	7.19±0.17^{b}	7.07±0.10^{b}
	K. pneumoniae ATCC 700603	7.16±0.05^{b}	11.23±0.04^{a}	7.12±0.17^{b}	7.17±0.05^{b}	7.09±0.16^{b}	6.54±0.46^{c}
Gram +	*S. aureus* ATCC 25923	7.45±0.23 ^{b,c}	9.96±1.10^{a}	7.8±0.14^{b}	8.23±0.54^{b}	9.35±0.83^{a}	6.95±0.10^{c}
	S. pneumoniae ATCC 29212	10.02±0.46^{a}	9.43±0.44^{b}	9.76 ±0.38^{b}	6.73±0.01^{c,d}	7.37±0.09^{c}	6.00^{d} (NA)
	R.equi ATCC 6939	6.00^{c} (NA)	11.02 ± 0.25^{a}	6.00^{c} (NA)	6.00^{c} (NA)	8.98±0.28^{b}	6.00^{c} (NA)

Table 4 demonstrates the results of antibacterial activity the different dilution of *A. absinthium* essential oil. The maximum antibacterial activity was exhibited against *E. coli* at 1/4 dilution of concentration (25.16 ± 0.61 mm inhibition zone), minimal antibacterial activity was identified against *K. pneumoniae* at *1/8* dilution of essential oil (7.90 ± 0.95 mm inhibition zone) and didn't show any inhibitory activity against *R. equi* at *1/8* dilution of *A. absinthium* essential oil. The activity was concentration-dependent. Moreover, *A. absinthium* essential oil were the most effective extracts and showed strong antibacterial activity against these pathogenic microorganisms.

Table 4. *Diameter of zone of inhibition of different dilution A. absinthium essential oil, against pathogenic bacteria*

Essential oil of *A. absinthium*				
		1/4	1/6	1/8
Zone of inhibition (mm)				
Gram (-)	*E.coli* ATCC 25922	25.16±0.61^{a}	17.82±0.66^{a}	9.9 ±0.57^{a}
	P.aeruginosa ATCC 27853	11.31±0.13^{c}	10.14±0.17^{b}	8.43±0.76^{b}
	K. pneumoniae ATCC 700603	9.61±0.59^{e}	9.16±0.49^{c}	7.90±0.95^{b,c}
Gram (+)	*S. aureus* ATCC 25923	12.02±0.13^{c}	10.81±0.79^{b}	7.97±0.49^{b,c}
	S. pneumoniae ATCC 29212	10.49±0.54^{d}	9.63±0.44^{c}	8.27±0.07^{b}
	R.equi ATCC 6939	16.10±0.40^{b}	10.89±0.13^{b}	6.00^{d} (NA)

Different superscript letters denote significant differences at $p < 0.05$ - Tukey's test

Results of antimicrobial activity of the plant extracts can suggest that *R. equi* was the most resistant strain to plant extracts followed by *S. pneumoniae*, *P. aeruginosa* and *K. pneumonia* while *S. aureus* and *E. coli* were the susceptible strains to the extracted plants respectively.

In general, the zones of inhibition obtained in the present study are different from results in the previous reports. This is due to several variables which influence the bioactive plant constituents against tested bacteria such as the environmental and climatic conditions under which the plant grows, choice of plant extracts, choice of extraction methods and antimicrobial test method as well [48].

They also found that *A. absinthium* essential oil had higher activity against pathogenic microorganisms than other ethanolic extracts of these plants study.

Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC)

The in vitro antibacterial activity of plants was evaluated against both Gram-positive and negative strains using the microdilution method to determine their MIC and MBC values. The MIC and MBC values of the extracts from various plant species against test bacteria are listed in Table 5.

***Table 5**. MICs and MBCs in mg·mL^{-1} of extracts from the studied plants*

Plants extracts			Ph. extract of *T. vulgaris*		Ph. extract of *A.absinthium*		Ph. extract of *S.aundulota*		Ph. extract of *M.Parviflora*		Ph. extract of *M.vulgare*		Ph.extract of *C.cinerea*		E. oil *of A. absinthium*	
			CMI	CMB	CMI	CMB	CMI	CMB	CMI	CMB	CMI	CMB	CMI	CMB	CMI	CMB
bacterial strains																
Gram (-)	*E.coli* ATCC 25922		0.625	–	1.25	5	1.25	2.5	2.5	–	2.5	–	1.25	–	0.059	1.904
	P.aeruginosa ATCC 27853		1.25	–	1.25	5	1.25	5	2.5	–	2.5	–	1.25	5	0.059	0.476
	K. pneumoniae ATCC 700603		0.625	5	1.25	5	1.25	5	2.5	–	2.5	–	1.25	5	0.014	1.904
Gram (+)	*S. aureus* ATCC 25923		0.625	5	1.25	2..5	1.25	5	2.5	5	2.5	5	1.25	–	0.0002	0.476
	S. pneumoniae ATCC 29212		0.625	–	1.25	–	1.25	–	2.5	–	2.5	–	1.25	–	0.059	0.476
	R.equi ATCC 6939		0.625	–	1.25	5	1.25	–	2.5	–	2.5	–	1.25	–	0.0002	15.233

According to Kuete *et al*. [49], the antibacterial activity of a plant extract is considered significant when the MICs are below 100 µg·mL^{-1}. The activity is considered moderate when 100 ≤ MIC ≤ 625 µg·mL^{-1} and weak when MIC is above 625 µg·mL^{-1}.

Among the different extracts, the extract of *T. vulgaris* showed significantly high antimicrobial activity against both Gram-positive and Gram-negative bacteria than the other extracts.

The essential oil of *A. absinthium* tested displayed impressive antibacterial efficacies against Gram-positive et gram-negative bacteria with the MIC values ≤ 0.059 mg·mL^{-1}. In particular, the extract of *T. vulgaris* showed a potent antibacterial activity against *K. pneumoniae* and *S. aureus* tested with the MIC and MBC values of (0.62 mg·mL^{-1} and 5 mg·mL^{-1}), high activity was also observed for the extracts of *A. absinthium* and *S. undulota* (MIC 1.25 mg·mL^{-1} and MBC 2.5 mg·mL^{-1}, respectively), followed by the extracts of *C. cinerea* (MIC, 1.25 mg·mL^{-1} and MBC value of 5 mg·mL^{-1}), finally the extracts of *M. parviflora* and *M. vulgare* (MIC, 2.5 mg·mL^{-1}and MBC value of 5 mg·mL^{-1}).

Results of antimicrobial activity of the six plant extracts can suggeste that *R. equi* was the most resistant strain to plant extracts followed by *K. pneumoniae* and S. *pneumoniae,* while *S. aureus* and *P. aeruginosa* were the most susceptible strains to the extracted plants respectively.

The MBC was confirmed by absence of bacterial growth of the tested strains streaked form inhibition zone corresponding to their lowest MIC's. *A. absinthium* essential oil showed potentially bactericidal activity against the tested pathogenic bacteria (*S. aureus, S. pneumoniae* and *P. aeruginosa*) with MBC of 0.47 mg·mL^{-1} while MBC of *S. aundulota* and *A. absinthium* phenolic extract reached to 2.5 mg·mL^{-1} against *E. coli* and *S. aureus* respectively. *K. pneumoniae* which was less sensitive and its minimal bactericidal concentration reached 5 mg·mL^{-1}

Correlation coefficients between phenolic compounds and antibacterial activity

A correlation analysis was carried out between phenolic compounds and antibacterial capacity (the MIC values of the extracts from various plant species against test bacteria, which is mg·mL^{-1}). In the present study, total phenolic compound was correlated with the MIC values of six plants against *E. coli* ATCC 25922, *P. aeruginosa* ATCC 27853, *K. pneumoniae* ATCC 700603, *S. aureus* ATCC 25923, *S. pneumoniae* ATCC 29212 and *R. equi* ATCC 6939 with correlation coefficient $R^2 = 0.1$, $R^2 = 0.005$, $R^2 = 0.01$, $R^2 = 0.10$, $R^2 = 0.01$ and $R^2 = 0.10$ respectivly (Figure 2).

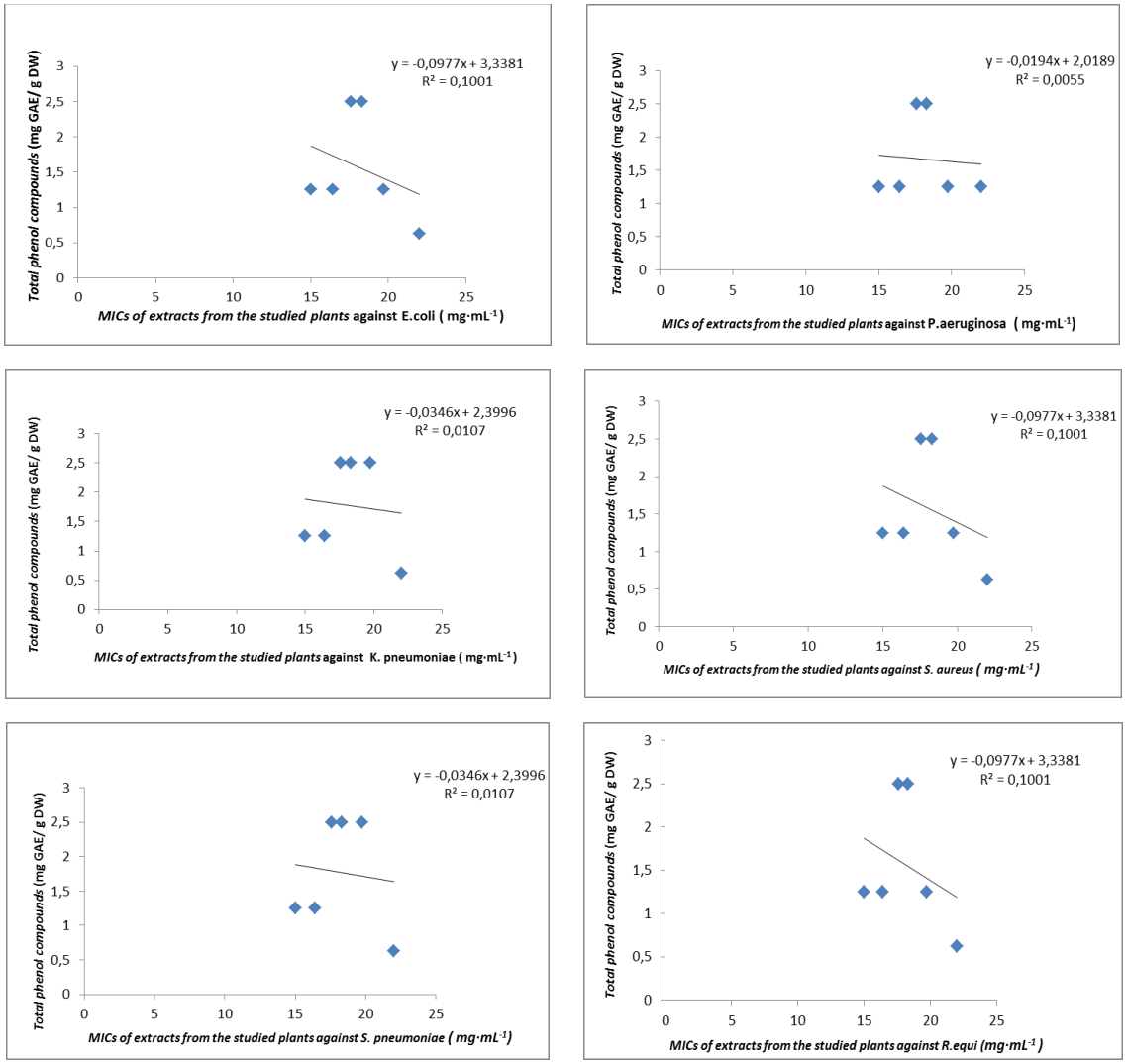

Figure 2. *Correlation between phenolic compounds and MIC values of the extracts plants*

As can be seen, none of the phenolic compounds was correlated with any MIC values of six plants. It indicates other phytochemical compounds which contribute to the antibacterial activity of these plants.

Mineral analysis

Mineral elements are inorganic elements which are regarded as essential to all of life's processes. Mineral requirement of the body has placed minerals in two groups; the macro minerals (macroelements or macronutrients) which are required in large quantities; they include calcium, magnesium, potassium, sodium and the micro minerals (microelements or micronutrients or trace elements), those elements which are required in small quantities; consisting of iron, manganese, zinc, cobalt, copper, cadmium, lead, nickel, lead [50]. The concentration reported in the analysis of the minerals of these selected plants was in ($mg \cdot kg^{-1}$) on dry weight. Mean values for mineral content of nutritional importance are presented in Table 6. The species analyzed in this study contained remarkably high amounts of K, Ca, Na et Mg. The lowest content of Na that is 7168.46 ± 216.03 $mg \cdot kg^{-1}$ was in *M. vulgare* and maximum concentration was estimated as 24395.16 ± 34.41 $mg \cdot kg^{-1}$ in *M. parviflora.*
The calcium contents varied between 3362.38 ± 442.00 – 110354.91 ± 3446.46 $mg \cdot kg^{-1}$, the highest calcium content was obtained for *M. parviflora* (110354.91 ± 3446.46 $mg \cdot kg^{-1}$), followed by *A. absinthium* (50200.38 ± 2529.01 $mg \cdot kg^{-1}$) and *S. undulata* (49648.51 ± 3311.26 $mg \cdot kg^{-1}$). The smaller calcium concentration was determinate for *M. vulgare* (3362.38 ± 442.00 $mg \cdot kg^{-1}$).
The values obtained for magnesium in analyzed medicinal plants ranged between 1605.97 ± 33.70 – 3861.42 ± 184.00 $mg \cdot kg^{-1}$. The highest content was measured in *M. parviflora* (3861.42 ± 184.00 $mg \cdot kg^{-1}$) and the smallest in *T. vulgaris* (1605.97 ± 33.70 $mg \cdot kg^{-1}$).
The range of potassium varied between 3375.34 ± 234.54 $mg \cdot kg^{-1}$ in *M. vulgare* and 273019.90 ± 238.3 $mg \cdot kg^{-1}$ in *S. undulata*.
The concentration of iron in the studied plants was high with a minimum of 265.50 ± 34.62 $mg \cdot kg^{-1}$ in *S. undulata* and maximum of 687.75 ± 0.90 $mg \cdot kg^{-1}$ in *M. vulgare.*
The Table 6 shows that the concentration of copper ranged between 448.08 ± 38.60 $mg \cdot kg^{-1}$ in *T. vulgaris* and 613.91 ± 5.12 $mg \cdot kg^{-1}$ in *C.cinerea* .
Zinc, manganese, lead and nickel were found in very low quantities in all analyzed medicinal plants, values below the detection limit of the equipment. Cadmium and Cobalt were not detected; this indicate that these minerals are not present in a detectable amount in the plants. This is beneficial to consumers, since it has been reported that some of these minerals like lead, cobalt and cadmium are highly toxic even at low concentrations [51]. All the plant parts have nutritional qualities which when used in the right proportions could be of tremendous benefit to the body.
Mineral analysis conducted by Imelouane *et al*. [52] showed that the *Thymus vulgaris* contained Ca (313044 $mg \cdot kg^{-1}$), Mg (53873 $mg \cdot kg^{-1}$), K (279491 $mg \cdot kg^{-1}$), Fe (27095 $mg \cdot kg^{-1}$), Zn (415.33 $mg \cdot kg^{-1}$) and Mn (731.41 $mg \cdot kg^{-1}$). And low level of Pb (88.88 $mg \cdot kg^{-1}$) and Cd (1.04 $mg \cdot kg^{-1}$). This values had higher then our results.

Table 6. *Mineral content of six plant extracts*

	T. vulgaris	*A. absinthium*	*S. undulata*	*M. Parviflora*	*M.vulgare*	*C. cinerea*
	Macro-elements (mg·kg^{-1}dw)					
Na	7840.50 ± 216.03 ^{b,c}	7728.46 ± 134.40 ^{c,d}	8445.34 ± 169.12 ^{c}	24395.16 ± 134.41 ^{a}	7168.46 ± 216.03 ^{d}	16241.04 ± 216.03 ^{b}
Ca	37507.23±4166.57 ^{c}	50200.38 ±2529.01 ^{b}	49648.51 ± 3311.26 ^{b,c}	110354.91±3446.46 ^{a}	3362.38 ± 442.00 ^{e}	5510.44 ± 437.20 ^{d}
Mg	1605.97 ± 33.70 ^{c}	2018.56 ± 38.39 ^{b}	2850.43 ± 250.67 ^{b}	3861.42 ± 184.00 ^{a}	1838.09 ± 145.22 ^{c}	2375.57 ± 217. ^{c,b}
K	47419.25 ±569.19 ^{c}	6659.47 ± 175.80 ^{b}	273019.90 ± 238.3 ^{a}	272946.97±221.01 ^{a}	3375.34 ± 234.54 ^{d}	4535.79 ± 264. 63 ^{c,d}
	Micro-elements (mg·kg^{-1}dw)					
Fe	568.00 ± 32.27 ^{b}	379.00 ± 22.76 ^{b}	265.50 ± 34.62 ^{d}	302.24 ± 24.04 ^{c,d}	687.75 ± 0.90 ^{a}	500.33 ± 1.42 ^{b}
Zn	32.88 ± 1.80 ^{d}	38.99 ± 2.17 ^{c}	39.80 ± 1.68 ^{c}	56.46 ± 0.70 ^{a}	35.70 ± 4.80 ^{c}	43.94 ± 4.32 ^{b,c}
Cu	448.08 ± 38.60 ^{c}	570.41 ± 46.27 ^{b}	610.66 ± 58.84 ^{a}	534.62 ± 41.33 ^{b}	612.58 ± 4.06 ^{a}	613.91 ± 5.12 ^{a}
Mn	10.95 ± 0.30 ^{e}	38.27 ± 2.34 ^{a}	21.49 ± 0.98 ^{b}	32.84 ± 0.61 ^{a}	13.31 ± 1.00 ^{e}	25.31 ± 1.06 ^{b}
Pb	27.80 ± 0.04 ^{b}	35.05 ± 1.08 ^{a}	28.61 ± 0.16 ^{b}	27.80 ± 0.38 ^{b}	28.06 ± 0.18 ^{b}	30.28 ± 0.15 ^{a,b}
Ni	33.91 ± 2.51 ^{c}	35.86 ± 5.15 ^{b}	40.11 ± 3.23 ^{a}	33.65 ± 9.71 ^{c}	37.17 ± 7.66 ^{a,b}	38.36 ± 10.40 ^{a,b}
Cd	ND	ND	ND	ND	ND	ND
Co	ND	ND	ND	ND	ND	ND

Values are presented as mean ± SE (n = 3). Different letters above the average bars denote significant differences at $p < 0.05$ - Tukey's test

The values obtained from this study in comparison to those available in the published literature showed disagreement with those reported by Erden et al. [53] of of some Scorzonera Species. However, there was some agreement in relation to mineral content in M. parviflora L. as observed by El-Tantawy [54]

The content of Pb, Ni, Cu, Mn, Zn and Fe for *Thymus vulgaris* was different than those reported by Abu-Darwish (The values obtained for Pb is 32.03 ± 0.04 ppm, Ni is 23.85

± 0.03 ppm, Cu is 13.23 ± 0.13 ppm, Mn is 15.52 ± 0.16 ppm, Zn is 16.18 ± 0.24 ppm and Fe is 141.3 ± 0.67 ppm [55].

CONCLUSION

From the present study, it can be concluded that the antioxidant activity observed is in a very good correlation with the selected medicinal plant extracts. Also, it was evident that the essential oil of *A. absinthium* showed potent antimicrobial activity. These medicinal plants are rich in some essential minerals, especially Ca, Na, K, Fe and Mg and very small, insignificant concentrations of toxic elements: Pb and Ni. Results presented here clearly show that the examined medicinal plants play a meaningful role in human nutrition as micro-nutrients sources. Thus, the study ascertains the value of plants used in ethanobotany by the locals, which could be of considerable interest in the development of new drugs.

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