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**PHENOLIC CONTENTS, *IN VITRO* ANTIOXIDANT AND ANTIMICROBIAL
ACTIVITIES OF *GENISTA MICROCEPHALA* COSS. & DUR.**

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Abstract

*This study was carried out to determine the phenolic contents as well as to evaluate the antioxidant and antimicrobial activities of *Genista microcephala*. Total phenolic and flavonoid compounds were quantified. Antioxidant activity was evaluated by various in vitro tests, including Ferric Ion Reducing Activity (FRP), Cuprac Ion Reducing Antioxidant Capacity (CUPRAC), and total antioxidant capacities (TAC). The antimicrobial activity of ethyl acetate and butanolic extracts of *G. microcephala* in*

*addition of fatty acids content of different parts of the studied plant was evaluated against human pathogenic bacteria, fish bacteria and two yeast. The used methods were agar diffusion method and the determination of minimal bactericidal and fungicidal concentrations (MBC and MFC). No significant difference was found between the two used solvents on total phenolic content (TPC). While, difference in total flavonoid content (TFC) were found to be significant. Ethylene acetate extract was the most active regarding antioxidant assay. The tested activity seems to be due to the high content of this extract on isoflavonoids. The tested extracts demonstrated the lowest inhibition concentration for *B.cereus* RSKK 863.*

Keywords: Antibacterial activity, Antioxidant activity, fatty acids, *Genista microcephala*, and Phenolics.

Introduction

The genus *Genista* represented by 150 species belongs to the family of *Leguminosae*, subfamily *Papilionaceae*. In the Mediterranean basin 16 species including 11 endemics are located [1]. Many *Genista* species possess medicinal properties related to flavonoids, especially isoflavonoids [2, 3].

Flavonoid compounds have attracted the attention of a number of researchers from different disciplines (biology, chemistry, pharmacy and medicine). They are an important family of antioxidants in plants, fruits and vegetables with no adverse effect on human health [4]. In addition they are cardioprotectors, vasodilators and demonstrate anticancer activities [5]. Some of them play the role of phytoalexins to fight against infections caused by fungi, or by bacteria [6]. Polyphenols are also used in the food industry as an additive, dye, aroma or preservative [7].

G. microcephala is used in traditional medicine as antimicrobial agent [8]. Chemical investigations on *G. microcephala* lead to the detection and isolation of several compounds with a wide diversity of structure such as alkaloids, isoflavonoids, terpenes and essential oil [9, 10, 11, 12, 13].

Lograda et al., [12] have reported a significant antimicrobial activity of the genus *Genista* on *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853 and *Staphylococcus aureus* ATCC 25923. This activity was correlated to alkaloids and essential oils components.

We aimed in the present study to investigate antimicrobial and antioxidant activities of *G. microcephala* extracts using different solvents. Moreover, fatty acid content of different parts, namely: flower, aerial part and a mixture of the two mentioned parts were also tested.

Methods

Plant material.

Genistami crocephala Coss et Dur. (1.7 Kg) was collected in 2011 (May-June) during the flowering stage from Chilia (35° 19' 05" north, 6° 38' 13" east), a mountain rising to 2,328 meters above sea level and situated in Algerian East on the border of the wilaya of Batna and Khenchela. The plant was authenticated by Prof. Mohamed Kaabache (Department of Biology, Faculty of Science, University Ferhat Abbass, Setif, Algeria). A Voucher specimen (RS / 71) was deposited at the laboratory of natural product of vegetable origin and organic synthesis. Department of Chemistry. University of Mentouri-Constantine 1. Algeria.

Chemicals and Reagents.

Ferric chlorides, potassium ferricyanide, Folin-Ciocalteu's phenol reagent, trichloroacetic acid (TCA), butylatedhydroxytoluene (BHT), were acquired from Merck (Darmstadt, Germany). Diethyl ether, dichloromethane, ethyl acetate, n-butanol, sulfinyl bis methane methyl sulfoxide (DMSO) and methanol, was acquired from (Sigma–Aldrich GmbH, Sternheim, Germany).

Preparation of plant extracts.

Aerial parts of *G. microcephala* Coss & Dur were extracted using the method described by Maanani et al., [10]. In brief, 70% MeOH was used for maceration. The obtained solution was then concentrated and the formed residue was dissolved in water and then successively extracted with methylene chloride, ethylene acetate and n-butanol affording 1.5g, 2.8g and 57g respectively. The extraction is repeated three times for each solvent.

Extraction of fatty acids.

Fatty acids were also extracted using the method described by IUPAC et al., [14]. Aerial parts (220.3g) flower (15g) and a mixture of both plant parts of *G. microcephala* Coss & Dur (101.3g) was extracted using diethyl ether in a soxhlet extractor during 6h at boiling point (34°C). The solvent was evaporated. The fatty acids is first saponified in the presence of alcoholic potash (0.5 N) forms methyl esters and then trans esterification in the presence of methanol with 14% BF₃ (v/v).

Determination of total phenolic content

The total phenolic content (TPC) was evaluated using Folin-Ciocalteu method involving the reduction of used reagent by phenolic compounds leading to formation of a blue complex. A volume of 0.5 ml of extract was mixed with 0.5 ml of Sodium carbonate (7.5%) and 0.1 ml Folin-Ciocalteu reagent. The tubes were kept 2 h at room temperature in the dark. The absorbance was then measured at 760 nm. Results were expressed as mg gallic acid equivalents per g of plant extract (mgGAE/ g plant extract) [15].

Determination of total flavonoid content

The total flavonoid content (TFC) based on the formation of flavonoids–aluminium complex was estimated by the method described by Quettier-Deleu et al. [16]. Equal volumes of extract and aluminium chloride solution (2%) were mixed. The absorbance

was measured at 415 nm after 15 min of incubation. Total flavonoid contents were expressed as mg quercetin equivalent per g of plant extract (mg QE/ g plant extract).

Antioxidant activity

The antioxidant activity of ethyl acetate and butanolic extracts were evaluated using three different methods: ferric reducing power (FRP), cupric ion reducing antioxidant capacity (CUPRAC) and total antioxidant capacity (TAC).

Reducing power assay (FRP).

The ferric reducing power was determined according to the method described by Oyaizu et al., [17]. The tested extracts were mixed with 0.5 ml of potassium ferric cyanide (1%) and 0.5 ml of 0.2 M phosphate buffer. After 20 min incubation at 50°C, 0.5 ml of ferric chloride (0.1%), 0.4 ml of trichloroacetic acid (10%) and 2 ml distilled water were added. The absorbance was then measured at 700 nm. The results were expressed as mg ascorbic acid equivalent (AAE) per g of plant extract (mg AAE/ g plant extract).

Cupric ion reducing antioxidant capacity (CUPRAC).

The cupric ion reducing antioxidant capacity was determined followed the method described by Apak et al., [18]. A volume of 0.5 ml of tested extracts, Ammonium acetate buffer (1M, pH 7.0), was added to 1 ml of Chloride dihydrate (10 mM) and 1 ml of neocuproine (7.5 mM), and 0.6 ml of deionized water. After 30 min incubation at room temperature, the absorbance was measured at 450 nm. Butylatedhydroxytoluene (BHT) was used as standard. Results were given as absorbance.

Total antioxidant capacity (TAC).

The total antioxidant capacity was evaluated by the phosphomolybdenum method as described by Prieto et al., [19]. A mixture of 3 ml of the phosphomolybdenum solution (sulfuric acid 0.6 M, sodiumphosphate 28mM, and ammonium molybdate 4mM) and 1 ml of the tested extract was incubated at 90°C. After 60 min, the absorbance was measured at 695 nm. The results were expressed as mg of ascorbic acid equivalent per g of plant extract (mg AAE/ g plant extract).

Antimicrobial activity

Microbial strains.

The screening of an eventual antibacterial activity was performed using pathogenic fish bacteria: *Lactococcus garvieae*, *Yersinia ruckeri* and *Vibrio anguillarum*. In addition, four gram-positive bacterias (*Listeria monocytogenes* ATCC 7644, *Staphylococcus aureus* ATCC 25923, *Bacillus cereus* RSKK 863, *Micrococcus luteus* NRRL B-4375), four Gram-negative bacteria (*Escherichia coli* ATCC 11229, *Escherichia coli* ATCC 35218, *Escherichia coli* O157:H7, *Yersinia enterocolitica* NCTC 11175) and two yeasts (*Candida albicans* ATCC 10231 and *Candida glabrata* RSKK 04019) were also tested. Nutrient agar (NA) and Tryptic Soy Agar (TSA) were used for the cultivation of bacteria while Yeast Peptone Dextrose (YPD) medium was used to culture the yeast. All bacterial

cultures were incubated at 37°C for 24 h whereas the yeast culture was incubated at 30°C for 48 h.

Inhibitory effect.

The inhibitory effect of ethyl acetate extract, butanolic extract and fatty acids were evaluated using disc diffusion method. All tested extracts were dissolved in sulfinylbismethane methyl sulfoxide (DMSO) to obtain a final concentration of 2 µg/ml and sterilized by filtration using 0.45 µm Millipore. The disc diffusion assay was performed using the method described by Murray et al., [20]. A volume of 100 µl of tested microorganisms previously cultured were adjusted to 0.5 McFarland and pooled in a petri plate. The different extracts were evaluated at a final concentration of 2000 µg/disc for ethylacetate and butanolic extracts and 1000 µg/disc for fatty acids. Ampicillin (Amp, 10µg/disc), Fluconazole (FCA, 25µg/disc), and Gentamycin (CN, 10µg/disc) were used as positive control.

Micro-well dilution assay.

Micro-well dilution assay of all tested extracts was performed. Minimal bactericidal and fungicidal concentrations (MBC/MFC) values were determined using the protocol described by Chandrasekaran et al., [21] with slight modification. The concentration of tested extracts ranged from 60.00 to 0.94 mg/ml. A standardized suspension of each tested microorganism adjusted to 0.5 McFarland standard turbidity was used. A positive and a negative control were also used. The MBC and MFC were determined as the lowest concentration of tested extracts with no growth after incubation on the appropriate agar medium.

Data processing and statistics.

Data were reported as mean \pm SD and subjected to statistical analysis using SPSS software (version 20.0) and origin 8. One-way and two-way analysis of variance (ANOVA) followed by Tukey post-hoc and Bonferroni's tests were used to analyse significant differences between treatments ($P < 0.05$). The correlation coefficients (R^2) for spectrophotometric assays were calculated using the Microsoft Office Excel 2011 software (Microsoft Corporation, Redmond, WA).

Results and Discussion

Total phenolic and flavonoid contents.

Total phenolic and flavonoid contents (TPC and TFC) of ethyl acetate and butanolic extracts of *Genista microcephala* Coss&Dur are illustrated in table 1.

Table 1. Total phenolic and flavonoid contents of *G. microcephala*.

| Extract | TPC (mg GAE/g) | TFC (mg RE/g) |
|------------------|--------------------|-------------------|
| Ethylene acetate | 690.32 \pm 69.10 | 159.94 \pm 0.11 |
| butanol | 661.91 \pm 17.21 | 66.83 \pm 5.01 |

Values are reported as means \pm S.D. of three measurements. TPC: Total phenolic content (mg GAE g⁻¹ extract). TFC: Total flavonoid content (mg RE g⁻¹ extract).

The ethyl acetate and butanolic extracts showed TPC of 690.32 ± 69.10 and 661.91 ± 17.21 mg GAE/g respectively with no significant difference between the two used solvents. While, difference in TFC were found to be significant with the highest value for ethyl acetate extract 159.94 ± 0.11 mg QE/g of plant extract. The finding values are higher than those reported by Hanganu et al., [22] for *G. tinctoria* and *G. Sagittalisare*. While, Bouchouka et al., [23] reported values of 459.28 mg GAE/g and 242 mgQE/g for *G. Sahara*. Our results are in accordance with previous investigation on *Genista* species. TPC and TFC of different extracts of *G. ulcina Spash* were investigated by Chebbah et al., [24]. Ethylene acetate extract exhibited the highest values compared to butanolic extract.

Antioxidant activity.

Oxidation is considered as a complex process using different mechanisms. The reductive capabilities of *G.microcephala* extracts was measured using FRP, CUPRAC and TAC assays.

Results of reducing power activity (FRP) are summarized in figure 1. Similar to TPC, no significant difference was found between ethyl acetate and butanolic extracts. However, no correlation was detected between FRP and TPC. In comparison with previous report on *Genista* sp, *G. microcephala* seems to show similar results as *G. vuralii* and *G. sandrasica* reported a values of 1.005 ± 0.018 and 0.860 ± 0.046 respectively for a testing concentration of 1000 μ g/ml[3].

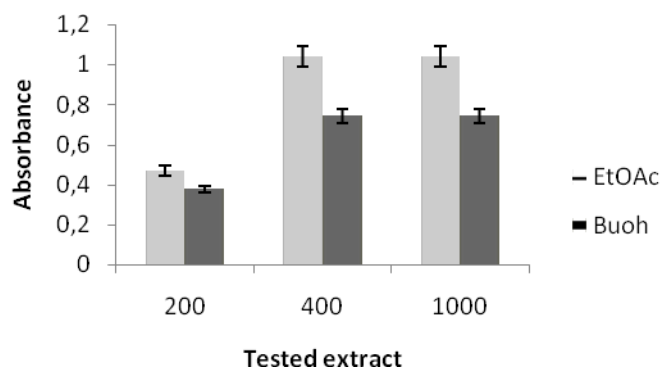


Figure 1. Ferric Ion Reducing Power Activity (FRP) activity of *G. microcephala*

Values are reported as means \pm S.D. Extract were evaluated in a concentration range from 200 to 1000 μ l/ml.

Cupric ion reducing antioxidant capacity method was also used. The cited method is reported as an inexpensive and a simple assay to study plant antioxidant activity [18]. In agreement with TFC, CUPRAC assay demonstrated the higher antioxidant activity for ethyl acetate extract (Figure 2). Difference was found to be significant. Our results are

different from those reported for methanolic extract of *Centaurea urvillei* sub sp. *Hayekiana* (0.64 mg/ml) by Zengin et al., [25].

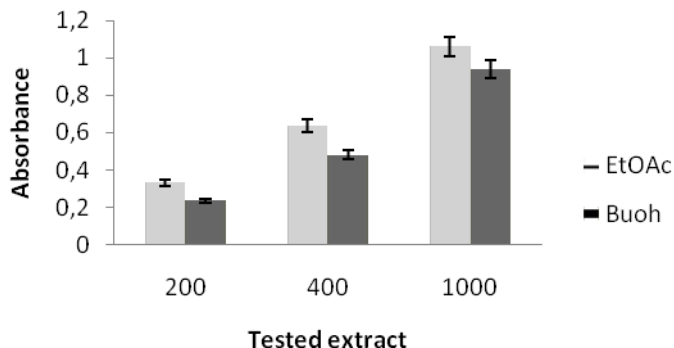


Figure 2. Cupricion reducing power activity (CUPRAC) activity of *G. microcephala*. Values are reported as means \pm S.D. Extract were evaluated in a concentration range from 200 to 1000 μ l/ml.

Total antioxidant capacity was evaluated using phosphomolybdenum assay. Results are expressed as ascorbic acid equivalent and represented in figure 3. The lower value was observed for butanolic extract with 421.08 ± 31.55 mg AAE/ g of plant extract. In accordance to previous assays, ethyl acetate extract showed the highest value. *G. microcephala* investigated in the present study demonstrated a notable TAC compared to *Centaurea urvillei* DC sub sp. *Hayekiana* which showed a TAC of 39.70 mg AE/ g of plant extract [25]. A high correlation was observed between TAC assay and TPC and TFC for ethyl acetate extract with a correlation coefficient $R^2 = 0.99$ and 0.97 respectively. This observation is in accordance with previous research on plant extract of *Leguminosae* in particular *Vigna sub terranea* seeds [26].

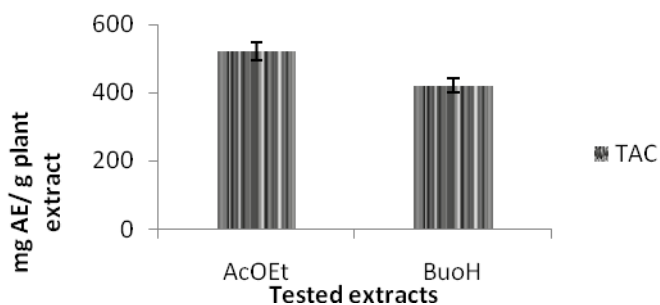


Figure 3. Total antioxidant capacity of *G. microcephala*.

Values are reported as means \pm S.D.

Our previous investigation of Ethyl acetate extract of *G. microcephala* Coss. & Dur lead to the isolation of four isoflavonoids namely: genistein, daidzein, isopuntine (5-methoxy daidzein), genistin (7-O- β -D-glucopyranosylgenistein) and daidzin (7-O- β -D-glucopyranosyldaidzein)[10]. The isolated flavonoids may be responsible of the observed antioxidant activity. In most of cases the genistein and daidzein and glucose forms are considered as phytoalexins [27,28]. Many studies supported that flavonoids and isoflavonoids possess antioxidant activity [23]. The cited activity is deeply influenced by the presence of free or glycosylated flavonoids [29]. In addition, Foti et al., 2005 demonstrated that genistein and daidzein exhibited a strong antioxidant activity [30]. In comparison with glycosylated forms such as genistein 7-O-glucoside heterosides and 4', 7-di-O-glucoside isoprunetin, free isoflavonoids or the aglycon ones were found to be more active [29]. Other researchers have shown that genistein have low antioxidant ability, suggesting that it may not have such a significant effect as antioxidant [31]. In a recent study reported that genistein and isoprunetin do not show antioxidant activity[32]. Further investigations on isoflavonoids are in need and may help a better comprehension of their role in antioxidant activity of natural product such as plant extract.

Antimicrobial activity

Antimicrobial activity of extracts

The antimicrobial activity of ethyl acetate and butanolic extracts of *G. microcephala* against human and fish pathogenic bacteria in addition to yeast was evaluated using agar diffusion method and the determination of MBC and MFC. Results are presented in table 2.

Table 2. Antimicrobial activity of *G. microcephala* extracts.

| Microorg anismsstr ains | MBC ^[a] /MF C ^[b] (mg/ml) | | Inhibitionzonediamet er ^[c] (mm) | | Antibiotics Inhibitionzonediameter ^[c] (mm) | | |
|-------------------------------|---|------|--|------------------------------|---|-----------------|-----|
| | AE E | BE | AEE (1000 μ g/disc) | BE (1000 μ g/disc) | Amp | CN | FCA |
| | | | | | | | |
| <i>B.cereus</i> R | 3.7 | 15.0 | 11.32 \pm 0. | 10.52 \pm 0. | 37.23 \pm 0. | 13.89 \pm 0.6 | - |
| SKK 863 | 5 | 0 | 36 | 11 | 21 | 8 | - |
| <i>E.</i> | 7.5 | 15.0 | 10.10 \pm 0. | 8.39 \pm 0.6 | 26.54 \pm 0. | 12.27 \pm 1.2 | - |
| <i>coli</i> O157: | 0 | 0 | 43 | 2 | 89 | 5 | - |
| H7 | | | | | | | |
| <i>M.</i> | 15. | 15.0 | 9.62 \pm 0.2 | 9.05 \pm 0.3 | 26.25 \pm 0. | 13.85 \pm 0.2 | - |
| <i>luteus</i> NRR | 00 | 0 | 0 | 2 | 26 | 1 | - |
| L B-4375 | | | | | | | |
| <i>Y.</i> | 15. | 30.0 | 10.08 \pm 0. | 7.42 \pm 0.3 | 25.25 \pm 0. | 22.13 \pm 0.0 | - |
| <i>enterocolit</i> | 00 | 0 | 24 | 5 | 29 | 7 | - |
| <i>ica</i> NCTC | | | | | | | |
| 11175 | | | | | | | |
| <i>E.</i> | 15. | 15.0 | 10.67 \pm 0. | 9.09 \pm 0.1 | 24.72 \pm 0. | 12.32 \pm 0.6 | - |

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| | | | | | | | |
|-----------------------------------|-----|----------------|------------|------------|------------|------------|------------|
| <i>coli</i> ATCC 11229 | 00 | 0 | 37 | 6 | 15 | 1 | |
| <i>S. aureus</i> ATCC 25923 | 7.5 | 7.50 | 12.49±0.81 | 9.43±0.74 | 24.93±1.34 | 14.65±0.19 | - |
| <i>E.coli</i> ATC C 35218 | 15. | 15.0 | 9.44±0.5 | 8.93±0.1 | 24.03±0.42 | 13.73±0.49 | - |
| <i>L. monocytogenes</i> ATCC 7644 | 15. | - ^d | 9.11±0.2 | - | 26.75±0.66 | 14.77±0.05 | - |
| <i>C. albicans</i> ATCC 10231 | 15. | 30.0 | 11.61±0.27 | 9.49±0.8 | - | - | 21.85±1.76 |
| <i>C. glabrata</i> RSKK 04019 | 7.5 | 30.0 | 13.78±0.92 | 10.33±0.66 | - | - | 25.06±1.92 |
| <i>L. garvieae</i> | 30. | 30.0 | 14.33±1.09 | 9.24±0.1 | 30.17±2.39 | 11.77±1.48 | - |
| <i>V. anguillarum</i> A4 | 15. | 30.0 | 16.08±0.18 | 9.83±0.1 | 29.72±1.51 | 10.76±0.6 | - |
| <i>Y. ruckeri</i> | 30. | 30.0 | 13.17±0.63 | 9.31±0.1 | 29.50±0.71 | 17.16±0.6 | - |

AEE: Ethyl acetate extract. BE: butanolic extract. [a]: Minimal Bactericidal Concentration (MBC). [b]: Minimal Fungicidal Concentration (MFC). [c]: Diameter of the inhibition zone including disc diameter.[d]: Indicates no antimicrobial activity.Values are reported as means \pm SD of three separate experiments.

Disc diffusion method produced clear zone with diameters ranging from 7.42±0.35 to 16.08±0.18 mm. The obtained diameters are very weak compared to ampicillin (24.03±0.42 to 37.23±0.21mm). MBC and MFC range was 3.75-30 mg/ml. No significant difference was observed between ethyl acetate and butanolic extracts. The tested extracts demonstrated the lowest inhibition concentration for *B.cereus* RSKK 863 indicating that this strain is the most sensitive. No effect was detected for butanolic extract on *L. monocytogenes* ATCC 7644. Among the three classes of microorganisms *G. microcephalase* seem to have more effect on Gram positive bacteria. While, for Gram negative bacteria, the two extracts seem to have the same effect on *E. coli* O157:H7, ATCC 11229 and ATCC 35218. The highest MBC values were obtained for fish bacteria (*L. garvieae*, *V. anguillarum*A4, *Y. ruckeri*) indicating a less pronounced effect. The MFC of butanolic extract are two and four fold higher than ethyl acetate extract for *C. Albicans* ATCC 10231 and *C. Glabrata* RSKK 04019, respectively. The results suggest that ethyl acetate solvent might be the best solvent in term of testing antifungal activity.

Our results are in agreement with previous reports on the genus *Genista*. Erdemoglu et al., [2] investigated antibacterial and antifungal activities of alkaloid extract of *G. vuralii* and reported an effect on *Staphylococcus aureus* and *Candida krusei*. The alkaloid extract of *G. microcephala* was also found to inhibit the growth *Escherichia coli* [9], in other study found that the aqueous and ethanolic extracts of *G. microcephala* did not inhibit the growth of *Escherichia coli* but had an effect on *Salmonella*, *Pseudomonas aeruginosa*, *Staphylococcus saprophyticus* and *Staphylococcus epidermis* [13]. In addition, Bouchouka et al., [23] reported a good activity of *G.sahara* on *S.aureus*.

Antimicrobial activity of fatty acids

The antimicrobial activity of fatty acids components of flower, aerial parts and a mixture of both parts of *G. microcephala* was also studied and presented in table 3. According to the results of disc diffusion method, all tested parts presents a weak activity on yeast, Gram negative, Gram positive and fish bacteria with a diameter varying from 7.16 ± 0.17 to 10.61 ± 0.11 . The antibiotic and antifungal used as a control present a remarkably higher values with a range of 24.03 ± 0.42 - 37.23 ± 0.21 and 21.85 ± 1.76 - 25.06 ± 1.92 for gentamicin and fuconazol, respectively.

Regarding MBC and MFC, the lowest value was obtained for inhibition of *B.cereus* RSKK 863 by aerial part (3.75 mg/ml). Flowers seem to act in the same way on *Y. Enterocolitica* NCTC 11175, *M. luteus*NRRL B-4375, *Y. enterocolitica*NCTC 11175, *E. coli* O157:H7, ATCC 11229 and ATCC 35218. While aerial part and a mixture of both parts of *G. microcephala* did not affect the growth of all cited bacteria. The highest MBC values were obtained for fish bacteria. No significant difference was found between antimicrobial activities of fatty acids components of flower, aerial parts and a mixture of both parts of *G. microcephala* on fish bacteria.

Table 3. Antimicrobial activity of *G. microcephala* fatty acids.

| Microorganisms strains | MBC ^[a] /MFC ^[b] (mg/ml) | | | Inhibition zone diameter ^[c] (mm) | | | Antibiotics Inhibition zone diameter ^[c] (mm) | | |
|------------------------------|--|----------------|-------|--|-------------------------|----------------------|--|------------------|-----|
| | Flower-E | Aerial-E | Mix-E | Flower-E (1000 µg/disc) | Aerial-E (1000 µg/disc) | Mix-E (1000 µg/disc) | Amp | CN | FCA |
| <i>B.cereus</i> RSKK 863 | 7.50 | 3.75 | 7.50 | 9.65 ± 0.44 | 10.01 ± 0.70 | 9.54 ± 0.39 | 37.23 ± 0.21 | 13.89 ± 0.68 | - |
| <i>E. coli</i> O157:H7 | 15.00 | - ^d | - | 8.66 ± 0.41 | - | - | 26.54 ± 0.89 | 12.27 ± 1.25 | - |
| <i>M. luteus</i> NRRL B-4375 | 15.00 | - | - | 10.36 ± 0.24 | - | - | 26.25 ± 0.26 | 13.85 ± 0.21 | - |
| <i>Y. enterocol</i> | 15.00 | - | - | 8.10 ± 0.33 | - | - | 25.25 ± 0.29 | 22.13 ± 0.07 | - |

| | | | | | | | | | |
|--|-------|-------|-------|------------|------------|------------|------------|----------------|------------|
| <i>itica</i> NCT C 11175 | 15.00 | - | - | 10.61±0.20 | - | - | 24.72±0.15 | 12.32±0.6 1 | - |
| <i>E. coli</i> ATC C 11229 | 7.50 | - | 15.00 | 10.61±0.11 | - | 11.65±0.09 | 24.93±1.34 | 14.65±0.1 9 | - |
| <i>S. aureus</i> ATCC 25923 | 15.00 | - | - | 9.86±0.93 | - | - | 24.03±0.42 | 13.73±0.4 9 | - |
| <i>E. coli</i> AT CC 35218 | 15.00 | 30.00 | 30.00 | 9.44±0.54 | 7.16±0.17 | 7.32±0.36 | 26.75±0.66 | 14.77±0.0 5 | - |
| <i>monocyt ogenes</i> ATCC 7644 | 7.50 | 30.00 | 7.50 | 10.19±0.56 | 10.15±0.74 | 11.32±0.88 | - | - | - |
| <i>albicans</i> ATCC 10231 | 7.50 | 15.00 | 15.00 | 10.33±0.12 | 9.36±0.41 | 10.16±0.45 | - | - | 21.85±1.76 |
| <i>glabrata</i> RSKK 04019 | 30.00 | 30.00 | 30.00 | 9.53±0.14 | 9.43±0.52 | 7.01±0.36 | 30.17±2.39 | 11.77±1.4 8 | - |
| <i>garvieae</i> <i>V. anguill</i> <i>arum</i> A4 | 30.00 | 30.00 | 30.00 | 9.76±0.70 | 9.73±0.43 | 11.97±0.22 | 29.72±1.51 | 10.76±0.6 6 | - |
| <i>Y. ruckeri</i> | 30.00 | 30.00 | 30.00 | 10.30±0.38 | 9.52±0.11 | 8.74±0.32 | 29.50±0.71 | 17.16±0.6 2 | - |

Aerial-E: Aerial part of plant extract fatty acid. Flower-E: flower extract of fatty acid. Mix-E: mixture of the Aerial part extract and flower extract of fatty acids. [a]: Minimal Bactericidal Concentration (MBC). [b]: Minimal Fungicidal Concentration (MFC). [c]: Diameter of the inhibition zone including disc diameter. [d]: Indicates no antimicrobial activity. Values are reported as means ± SD of three separate experiments.

Recent research on distilled oil composition of *G. microcephala* showed a fatty acid content of 64.4% with 0.8% caprylic acid, 21.5% lauric acid, 8.7% myristic acid, 29.6% palmitic acid, 1.1% octadécan-9.12-dienoate methyle, 2.5% linoleic and 0.2% stearic acid. Interesting activity on *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853 and *Staphylococcus aureus* ATCC 25923. Witch mean that the nature of this oil is fatty acids [12]. The cited study concerned three bacteria strains. We reported here our results on the investigation of antimicrobial activity of *G. microcephala* on twelve bacterias, two fish bacterias and two yeast pathogens. Our study might complete previous investigation on *G. microcephala* performed by Lograda et al., [12].

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Conclusion

This study provides information on the antioxidant and antimicrobial capacities of *Genista microcephala* extracts and their maybe use in ethno medicine. Our results demonstrate a good antioxidant activity of ethyl acetate extract in comparison to butanolic extract. In addition, the MFC of butanolic extract are two and four fold higher than ethyl acetate extract for *C. Albicans* ATCC 10231 and *C. Glabrata* RSKK 04019, respectively. The finding results suggest that ethyl acetate solvent might be the best solvent in term of testing antifungal activity.

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Conflicts of Interests

No potential conflict of interest was reported by the authors.

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