Original article

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ANTIOXIDANT ACTIVITY OF HYDROLATE OBTAINED FROM THE AERIAL PART OF SWEET BASIL *OCIMUM BASILICUM* L.

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Ocimum basilicum is a well-known aromatic, medicinal, and culinary plant. The essential oils obtained through steam distillation from the leaves and flowering tops of sweet basil have antiseptic, antimicrobial, antioxidant, antiviral, and anti-inflammatory properties. As a product of hydrodestination hydrolates can be obtained. The main objective of the study was the spectrophotometric quantification of the content of total phenols, tannins, nontannins and flavonoids in the hydrolate obtained from aerial part of *O. basilicum*, the evaluation of antioxidant activity and the correlation analysis of certain phenolic compounds and antioxidant activity. Quantitative analysis of the concentration of total phenolic, flavonoid, tannin and nontannin in the hydrolate determined 151.91 ± 23.491mg CE/L , 23.34 ± 3.978 mg CE/L, 119.75 ± 8.09 mg CE/L and 0.86 ± 0.07 mg Ru/L, respectively. The hydrolate showed antioxidant potential in three assays for study: scavenged 2,2-diphenyl-1 picrylhydrazyl (DPPH) radicals with IC_{50} 0.51 ± 1.07 %; total antioxidant potential, 392.15 ± 16.299 mmol of Fe^{2+}/L and prevention of lipid peroxidation in a way that depends on concentration. In addition, correlation between phenolic compounds contents and antioxidant activity in hydrolated was noted. The demonstrated antioxidant properties of *O. basilicum* hydrolate may be crucial to its future as a potential natural antioxidant.

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Key words: *antioxidant, ferrie reducing ability ofplasma, hydrosol, polyphenols, sweet basil*

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Introduction

The increasing appeal of using natural products has led to a growing interest in aromatic and medicinal plants. This is especially noticeable in the abundance of phytochemicals produced for use in various industries such as food, chemicals, pharmaceuticals, and cosmetics. A good example of this trend is the move towards the use of healthier and more organic substitutes for synthetic products, including aromatic chemicals derived from plants. Aside from the obvious benefits of using natural resources for human consumption, this approach offers a more commercially viable and environmentally sustainable method of production (1).

Hydrolates are obtained during the extraction of essential oils from aromatic plants. These are aqueous solutions that are a by-product of distillation and contain a certain amount of bioactive molecules that quantitatively and qualitatively might be different from the essential oils (2). As hydrolates are no longer classified as waste, their use is in line with the concepts of biorefinery and green extraction (3,4). Compared to essential oils, the production of hydrolate is easy and inexpensive. Furthermore, it appears to be less toxic to human health. Since hydrolates have such significant biological activity, it could be beneficial to turn them into valuable products. Furthermore, despite the published scientific articles and the various proposed practical applications, it is crucial to conduct further research on plant hydrolysis (2,5,6).

The Lamiaceae family is a world-renowned family of aromatic plants with strong antimicrobial and antioxidant properties. It is also the family of the well-known spices (7). Depending on the variety, *Ocimum* species vary greatly in form and chemical composition. These variations include phenolic profile and volatile

organic chemicals, as well as variations in leaf and flower size and color, plant height, flowering time and other characteristics. Of the 150 species of *Ocimum basilicum* L., so much is known about it, its cultivation, distribution and uses that it is referred to as sweet basil and the "*king of herbs*" as it is one of the most widely cultivated aromatic plants in the world. *O. basilicum* is also used in traditional medicine for a wide range of diseases and conditions of the respiratory and urinary tract as well as in the prevention of neurodegenerative and cardiovascular disorders (8−10). It also has antipyretic, hypoglycemic and antihypertensive effects. Additionally, it is even thought to be an anti-cancer agent. However, because of its relaxing effect on the nervous system, sweet basil essential oil is often used in aromatherapy. It can also help to improve memory, relieve migraines and combat mental fatigue and insomnia (11). Sweet basil essential oil has antiseptic, antimicrobial, antioxidant, antiviral and anti-inflammatory properties (10). In addition, both the plant itself and its essential oil have an insecticidal and fungicidal effect. When using essential oil as a biopesticide, it is important to pay attention to the dosage, as a higher concentration can be harmful to certain plants (12). Although a wide range of bioactive properties are attributed to *O. basilicum*, the antimicrobial and antioxidant properties are currently the most studied (1). These properties form the basis for many of the applications of *O. basilicum* mentioned above.

The study of chemical compositions of hydrolates obtained from fresh and dried aerial parts of sweet basil *O. basilicum* was conducted. The main compounds in hydrolates obtained from fresh and dried aerial parts of basil were methyleugenol (51.0 %, 33.4 %), eugenol (26.0 %, 5.8%) and linalool (11.3 %, 10.2 %), retrospectively (13). In aid to creating safe, eco-friendly preservatives that prevent food spoilage by fungi the investigation of antifungal activity of hydrolate obtained from leaves and flowers of *O. basilicum* against *Rhizoctonia solani*, *Fusarium oxysporum* f. sp *tulipae*, *Botrytis cinerea* and *Alternaria citri* was done (14). Furthermore, research on basil hydrolate as a bioherbicide, which is now preferred as a natural compound in organic agriculture, has been done. Under laboratory conditions, the different concentrations of hydrolate obtained from *O. basilicum* leaves significantly decreased the germination of both *O. basilicum* and *Chenopodium quinoa* seeds (15).

Therefore, even as research about hydrolate obtained from aerial part of *O. basilicum* is still limited, there is a potential for use in pharmacy.

Aim

The aim of this study was: (i) spectrophotometric quantification of total phenolic, tannin, nontannin and flavonoid content in hydrolate obtained from aerial part of *O. basilicum*, (ii) *in vitro* evaluation of the antioxidant activity of the hydrolate obtained from aerial part of *O. basilicum* employing three different assays: 2,2-diphenyl-1 picrylhydrazyl assay, *β*-carotene bleaching assay and ferrie reducing ability ofplasma assay, (iii) correlation analysis of certain phenolic compounds and antioxidant activity.

Material and methods

Plant material and chemicals

The hydrolate of the aerial part of *O. basilicum* was obtained through the industrial steam distillation process by the company "PROMONTIS production", Vilandrica, Gadžin Han After the distillation process, double microbiological filtration was performed.

All chemicals used were obtained from Sigma Aldrich (USA), or Zorka pharma (Šabac, Serbia). All solvents and chemicals were of analytical grade.

Determination of total phenolic content

The total phenolic content of hydrolate obtained from the aerial part of *O. basilicum* was determined using the Folin−Ciocalteu method (16,17). The Folin-Ciocalteu reagent, which had previously been diluted 1:1 v/v with distilled water, and a 20% Na_2CO_3 solution were added to the test tubes into which the studied hydrolate had previously been added. The tubes were then shaken vigorously and allowed to stand for 40 minutes to develop a blue color, and the absorbance was measured spectrophotometrically at 725 nm in comparison to a blank containing the extraction solvent (water) instead of the sample. The experiment was done in triplicate. The total phenolic content of the sweet basil hydrolate was calculated using a (+)-catechin calibration curve (range 1−5 μg/mL) and expressed as mg catechin equivalents (CE) per L of hydrolate.

Determination of total tannin content

The total tannin content of hydrolate obtained from the aerial part of *O. basilicum* was determined using the same Folin−Ciocalteu method (18,19). Polyvinylpolypyrrolidone was added to the test tubes containing the sample and then shaken vigorously. The supernatant contains all phenolic compounds except the tannins. The test was performed on the clear supernatant and the results were expressed in mg catechin equivalents (CE) per L of hydrolate. The experiment was done in triplicate. The content of nontannin polyphenols is also expressed as mg catechin equivalents per L of hydrolate and results from the difference

between the total phenolic content and the total tannin content.

Determination of total flavonoid content

The total flavonoid content of hydrolate obtained from the aerial part of *O. basilicum* was estimated according to Lamaison and Carnat (18). Aluminum trichloride ($AlCl_3$) in ethanol was mixed with the same volume of the hydrolate. The blank sample consisted $AlCl_3$ with ethanol without extract solution. After incubation for 1 hour, the absorbance was measured spectrophotometrically at 430 nm. The experiment was done in triplicate. The total flavonoid content of sweet basil hydrolate was calculated and expressed as mg rutin (Ru) per L of hydrolate using a rutin calibration curve (range 0.5−5 µg/mL).

Antioxidant activity examination

DPPH assay

The antioxidant activity of hydrolate obtained from the aerial part of *O. basilicum* was assessed using the DPPH (1,1-diphenyl-2 picrylhydrazyl) free radical scavenging assay. The color changed from violet to yellow when DPPH reduced to DPPHH, and an ELISA microplate reader was used to measure it at 540 nm (13). The assay was performed according to Pavlović et al. (19) by incubating different concentrations of hydrolate (10−70% v/v) with DPPH in 96% (v/v) ethanol solution for 30 minutes at room temperature and in the dark and then absorbance was measured on ELISA microplate reader at 540 nm (20). The distilled water is present in the blank sample. As a control, 96% (v/v) ethanol containing DPPH was used. Synthetic antioxidants BHT and BHA were used as the reference compounds. The experiment was done in triplicate. The following formula was used to determine the percentage of DPPH free radical inhibition:

$$\% \text{ DPPH} = (Ac – As) / Ac \times 100$$

where Ac is the absorbance of the control, and As is the absorbance of the sample.

β-carotene bleaching assay

The *β*-carotene bleaching method assesses the capacity of hydrolate obtained from the aerial part of *O. basilicum* to impede the process of lipid peroxidation. The microplate with freshly prepared *β*-carotene linoleic acid emulsion (200 mg of Tween-20 and 25 µl of linoleic acid were combined with 1 ml of *β*-carotene solution in chloroform 1 mg/5 ml) and different concentrations of hydrolate (1.11 – 11.1 % v/v) was read in a microplate reader immediately (t 0 min) and after 120 minutes of incubation at 55 °C (t = 120 min) on 450 nm (21). The percent inhibition of *β*-carotene bleaching by the samples was calculated according to formula (22):

$$\% \text{ inhibition} = (A_{120} / A_0) \times 100$$

where A_{120} is the absorbance of the sample at t = 120 min and A_0 is the absorbance of the sample at t = 0 min. The experiment was done in triplicate. The reference compounds were synthetic antioxidants BHT and BHA.

FRAP assay

The total reduction potential of hydrolate obtained from the aerial part of *O. basilicum* was measured using the FRAP (ferric reducing antioxidant power) assay. The method is based on reducing the ferric tripyrydiltriazine (Fe^{3+}-TPTZ) complex to the ferrous tripyrydiltriazine (Fe^{2+}-TPTZ) form in the presence of antioxidants at a low pH (23). After incubation at 37 °C for 30 min, a solution containing FRAP working reagent (10 mmol/L TPTZ in 40 mmol/L HCl, 300 mmol/L sodium acetate buffer, pH 3.6 and 20 mmol/L $FeCl_3 \times 6H_2O$ solution, each in a ratio of 10:1:1 (v/v/v)) was mixed with hydrolate and the absorbance was measured at 593 nm. The result of the FRAP assay of sweet basil hydrolate was calculated using an Fe^{2+} sulfate calibration curve (range 100−1000 mmol/L) and expressed as FRAP value, as mmol of Fe^{2+} per L of hydrolate. The experiment was done in triplicate.

Statistical analysis

All experimental measurements were performed in triplicate and expressed as the standard deviation of the average of the three measurements. The normality of the distribution of the continuous variants was determined using the Shapiro−Wilk test. The comparison of variable values between several groups was carried out using One-Way ANOVA followed by Tuckey's post hoc test. Pearson's linear correlation coefficient was used for the correlation analysis. A significance level of $p < 0.05$ is considered statistically significant. Statistical analyses were performed using SPSS v. 20.0 software. The heatmap was obtained by using OriginLab graphing and data analysis software.

Results

Determination of total phenolic content

The results of the spectrophotometric determination of the total phenolic content of the hydrolate obtained from the aerial part of *O. basilicum* are shown in Table 1. The total phenolic content of sweet basil hydrolate calculated using a (+)-catechin calibration curve (range 1−5 µg/mL) was 151.91 ± 23.491 mg CE/L.

Determination of total tannin and nontannin content

The results of the spectrophotometric determination of the total tannin and nontannin content of the hydrolate obtained from the aerial part of *O. basilicum* are shown in Table 1. The total tannin content of sweet basil hydrolate calculated using a (+)-catechin calibration curve (range 1–5 μg/mL) was 23.34 ± 3.978 mg CE/L. The total nontannin content of hydrolate obtained from the aerial part of *O. basilicum* was 119.75 ± 8.09 mg CE/ L.

Determination of total flavonoid content

The results of the spectrophotometric determination of the total flavonoid content of the hydrolate obtained from the aerial part of *O. basilicum* using aluminum trichloride complexing agent are shown in Table 1. The total flavonoid content of sweet basil hydrolate was calculated using a rutin calibration curve (range 0.5–5 μg/mL) was 0.86 ± 0.07 mg Ru/L.

Antioxidant activity examination

DPPH assay

The results of the ability of different concentrations (10–70% v/v) of the hydrolate obtained from the aerial part of *O. basilicum* to inhibit the DPPH radical are presented in Figure 1. According to DPPH assay all tested concetracions of sweet basil hydrolate possess anti-radical activity, IC_{50} value 0.51 ± 1.07 % (v/v). A statistically significant difference ($p < 0.05$) in the percent of inhibition of DPPH radical was recorded between the second and third concentrations of the hydrolate, 20% *vs* 40%. Under the same conditions IC_{50} values for BHT and BHA, commercial synthetic antioxidants, were 22.82 ± 2.07 μg/ml and 2.44 ± 0.09 μg/ml, respectively.

β-carotene bleaching assay

The results of the determination percent of *β*-carotene bleaching as a function of concentration of the hydrolate obtained from the aerial part of *O. basilicum* expressed in percent inhibition are presented in Figure 2. In the current study sweer basil hydrolate showed mean inhibition of bleaching from 5.023 ± 0.809 to 21.37 ± 3.207, for concentracion range 1.11–11.1% (v/v), respectively. A statistically significant difference ($p < 0.05$) in the percent of *β*-carotene bleaching was recorded between the second and third concentration (2.78% *vs* 5.56%) and between the third and fourth concentration (8.3% *vs* 11.1%). Under the same conditions conditions IC_{50} values for BHT and BHA, commercial synthetic antioxidants, were 0.03 ± 0.00 μg/ml and 0.04 ± 0.01 μg/ml, respectively.

FRAP assay

The results of *in vitro* determination of the total reduction potential of the hydrolate obtained from the aerial part of *O. basilicum* using the FRAP assay are shown in Table 1. The total reduction potential of basil hydrolate was measured using the standard curve of known concentrations of ferrous sulfate solutions (range 100–1000 mmol/l) was 392.15 ± 16.299 expressed as mmol of Fe^{2+} per L of hydrolate.

Correlation between phenolic compounds contents and antioxidant activity

Pearson correlation coefficients analysis was worked out among total phenolic, flavonoid, tannin, nontannin content and antioxidant activity of hydrolate obtained from the aerial part of *O. basilicum*. The total phenolic, tannin and nontannin content showed a weak correlation with the percentage of DPPH free radical inhibition, while total flavonoids content showed a strong negative correlation (r = -0.961) (Figure 3). Furthermore, the examined phenolic compounds showed a weak negative correlation with the percent of *β*-carotene discoloration. Similar results were noted between phenolic compounds and total antioxidant potential, except for total flavonoid content where we observed a significant positive correlation (r = 0.987)(Figure 3).

Table 1. Phenolic compounds contents and *in vitro* antioxidant assay of *O. basilicum* hydrolate

Hydrolate	Phenolic compounds content				Antioxidant activity
	Total phenolic content	Total tannin content	Total nontannin content	Total flavonoid content	FRAP assay
	(mgCE/L)	(mg CE/L)	(mg CE/L)	(mg Ru/L)	(mmol Fe^{2+}/L)
Ocimum basilicum L.	151.91 ± 23.491	23.34 ± 3.978	119.75 ± 8.09	0.86 ± 0.07	392 ± 16.299

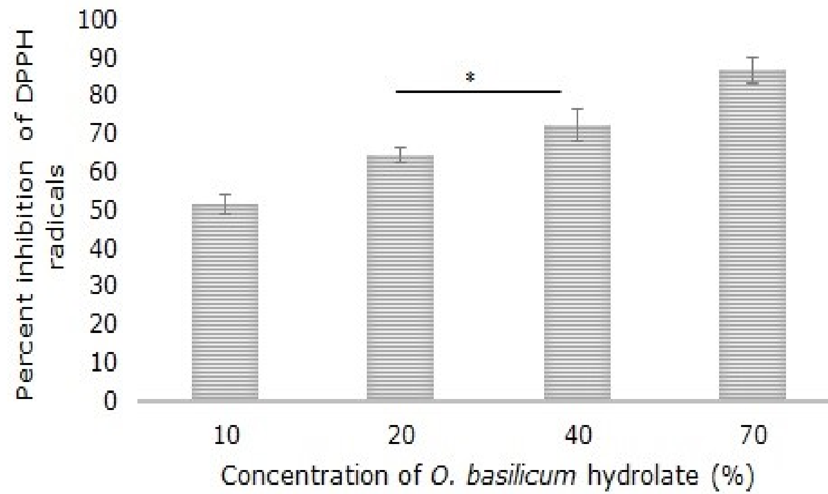

Figure 1 Results of antioxidant testing: inhibition of DPPH radicals. Data are presented as mean ± SD and further compared using One-Way ANOVA followed by Tuckey's post hoc test. * $p < 0.05$

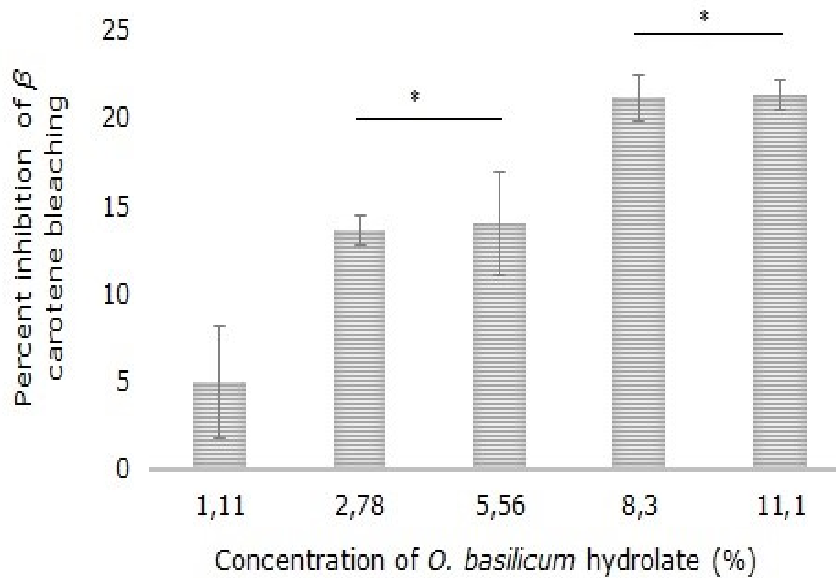

Figure 2 Results of antioxidant testing: inhibition of β-carotene bleaching. Data are presented as mean ± SD and further compared using One-Way ANOVA followed by Tuckey's post hoc test. * $p < 0.05$

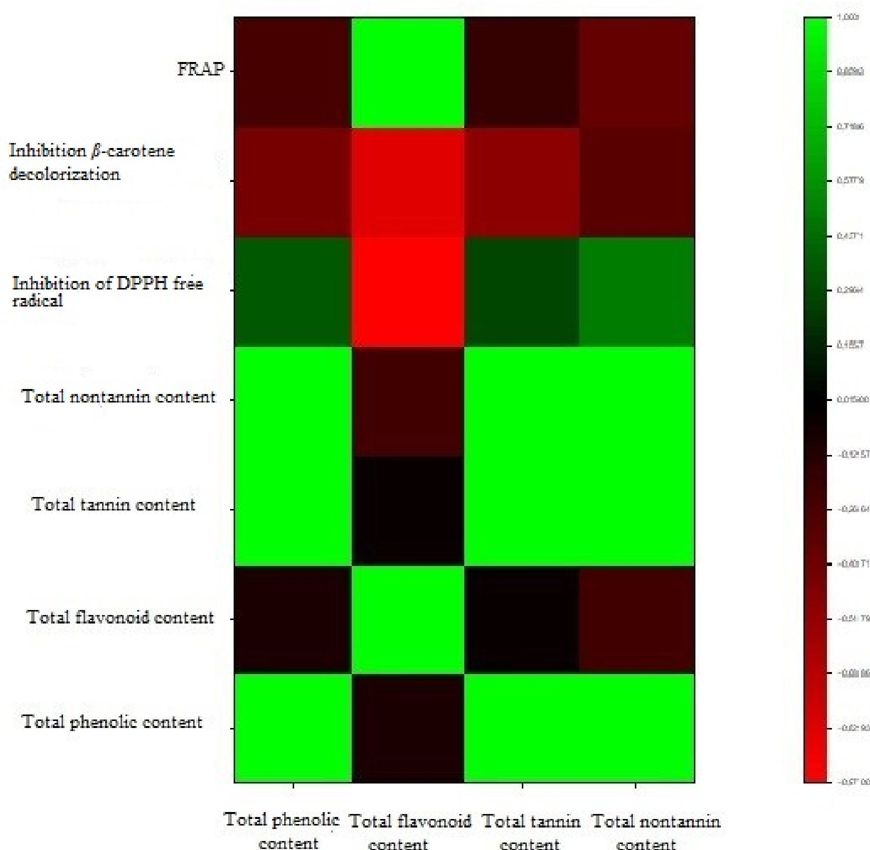

Figure 3. Heatmap of the correlation matrix generated by the Pearson r correlation coefficient for the phenolic compounds and antioxidant activity (inhibition of DPPH free radical, inhibition of β-carotene decolorization and FRAP) for *O. basilicum* hydrolate. The scale ranges from -0.961 (red) to +1 (green)

Discussion

Many diseases of the 21st century, including malignancies, cardiovascular disease and inflammation, are thought to be caused by oxidative stress. An imbalance between the antioxidant systems and the production of oxidant species leads to oxidative stress.

These diseases are caused by oxidative damage to vital elements such as nucleic acids, proteins, carbohydrates and lipids. Studies have shown that the consumption of antioxidants, especially those found in foods, can be beneficial in combating these effects. Plants are rich sources of antioxidants such as ascorbic acid, carotenoids, various phenolic compounds, alcohols, stilbenes, tocopherols and tocotrienols (24).

In recent decades, the food industry has focused on finding natural antioxidants from plants that are effective for human use and have minimal side effects. This search has become a major industry and research problem in the field of food technology. Despite extensive research, the potential of plant antioxidants has not yet been fully understood, and their benefits have not been fully realized yet. Therefore, there is a need for reliable *in vitro* methods to screen for antioxidant activity (25).

Hydrolates have historically been employed in food and functional drink items. For instance, in Turkey, sage hydrolate is consumed as a natural antibiotic to treat digestive issues and a variety of bacterial illnesses (26). Also, hydrolates are utilized as drinks in traditional and folk Persian medicine to cure a variety of ailments (27,28). These facts are highlighted only that hydrolates are valuable, innovative natural products that have great potential for use in the food industry as functional beverages.

Phenols are one of the main compounds with antioxidant activity, because they have an aromatic ring in their structure that allows the stabilization and relocation of unpaired electrons. Further, the aromatic ring also makes it easier for their hydroxyl groups to donate hydrogen atoms and electrons (29). The species, tissue, and developmental stage of the plant affect the

overall phenolic content. Also, environmental elements such as water stress, light conditions and temperature have an impact on phenolic content (30). As shown in Table 1. the total phenolic content of the hydrolate obtained from the aerial part of *O. basilicum* was 151.91 ± 23.491 mg CE/L determined by Folin−Ciocalteu method. Thanks to the presence of free OH groups, particularly 3-OH groups flavonoids, secondary metabolites from plants have the potential to be antioxidants (31). According to the results of our study, hydrolate obtained from the aerial part of *O. basilicum* contained 0.86 ± 0.07 mg Ru/L, as shown in Table 1. Also, phenolic compounds include tannins, which, like other phenolic compounds, among other activities, have proven antioxidant activity (31). The total tannin content was also examined using the Folin−Ciocalteu method. We found that the hydrolate obtained from the aerial part of *O. basilicum* contained 23.34 ± 3.978 CE/L as shown in Table 1. As the obtained results indicate, the studied hydrolate obtained from the aerial part of *O. basilicum* contains phenolic, tannic and flavonoid compounds. These phytochemical compounds may be responsible for many of the health benefits of this hydrolate.

Plant extracts can be assessed for their antioxidant activity using various methods. It is suggested that a minimum of two different methods be used for accurate measurement. In our study, we utilized three assays, each with a different mechanism of reaction (32). One of the assays was based on hydrogen atom transfer, while the other two were based on electron transfer.

The synthetic free radical generator DPPH is used to test the scavenging power of certain antioxidants in DPPH assay. When the H^+-donating antioxidant reacts with DPPH, it results in a reduction in DPHH to hydrazine, which causes a decrease in the absorbance of the reaction (33). Additionally, the response causes a changed color from purple to yellow. The degree of discolouration and absorbance drop is correlated with the sample's antioxidant activity, concentration, and capacity to donate H^+ (34). Proteins and nucleic acids, two important macromolecules, can be harmed by hydroxyl radicals, which are highly reactive and have a short lifespan when produced by the Haber–Weiss/Fenton process (35). Because of their high sensitivity, hydroxyl radicals seriously damage both individual cells and the components that make them up, as well as entire organisms (36). Further, by measuring the process by which antioxidants prevent lipid oxidation, DPPH free-radical scavenging is used to evaluate antioxidant activity and, consequently, free-radical scavenging capacity. As a result, a moderate DPPH scavenging action of hydrolate could be related to the existence of phenols, as shown in the current work. To assess the antioxidant capacities of the hydrolate obtained from the aerial part of *O. basilicum in vitro*, we used the FRAP assayand compared it with two other assays. The molecular mechanisam of FRAP assay is performed on electron-transfer processes. The reduction of iron(III)-2,4,6-tripyridyl-S-triazine at low pH to an intense blue colored iron (II) tripyridyltriazine complex is the reaction mechanism (37). According to the result of the assay sweet basil hydrolate might be consider possessing moderate antioxidant capacities. In the *β*-carotene bleaching assay, the yellow color of *β*-carotene is lost due to its reactivity with radicals formed in an emulsion when linoleic acid oxidizes. Antioxidants are able to slow down the bleaching of *β*-carotene (24). In the current study, all tested concentracions of sweer basil hydrolate showed mean inhibition of *β*-carotene bleaching. The obtained results might be considered significant due to the inhibition of lipid peroxidation using the *in vitro β*-carotene-linoleic acid system mimics *in vivo* oxidation/protection of valuable biological fatty acids present in cell membranes.

This is the first study to determine the phenolic compound content and antioxidant activity of the hydrolate from the aerial part of *O. basilicum*. The results show that the sweet basil hydrolate contains certain phenolic, flavonoid and tannic compounds. The overall phenolic, tannic and flavonoid properties of basil hydrolate are associated with its antioxidant activity. However, a comprehensive phytochemical analysis is required before using hydrolates to treat diseases associated with oxidative stress.

Conclusion

Spectrophotometric quantification of hydrolate obtained from the aerial part of *O. basilicum* a certain concentration of total tannin, flavonoid, tannin and nontannin was observed 151.91 ± 23.491mg CE/L , 23.34 ± 3.978 mg CE/L, 119.75 ± 8.09 mg CE/L and 0.86 ± 0.07 mg Ru/L, respectively. The tested concentration of hydrolate affected the neutralization of DPPH radicals from 51.57 ± 2.592% to 86.87 ± 3.304% and *β*-caroten bleaching from 5.023 ± 0.809% to 21.37 ± 3.207%. The total reducing power in the FRAP assay was 392.15 ± 16.299 mmol Fe^{2+}/L hydrolate. Additionally, the correlation between certain phenolic compounds and antioxidant activity has been noted.

Acknowledgments

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ANTIOKSIDATIVNA AKTIVNOST HIDROLATA DOBIJENOG IZ NADZEMNOG DELA BOSILJKA *OCMIUM BASILICUM* L.

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Ocimum basilicum je poznata aromatična, lekovita i kulinarska biljka. Etarsko ulje dobijeno parnom destilacijom iz listova i vršnih cvetova bosiljka ima antiseptička, antimikrobna, antioksidativna, antivirusna i antiinflamatorna svojstva. Hidrolati se mogu dobiti kao proizvodi hidrodestilacije aromatičih biljaka. Za potrebe ove studije izvršene su spektrofotometrijska kvantifikacija ukupnog sadržaja fenola, tanina, netanina i flavonoida u hidrolatu dobijenom iz nadzemnog dela biljke *O. basilicum*, procena antioksidativne aktivnosti i korelaciona analiza fenolnih jedinjenja i antioksidativne aktivnosti. Kvantitativnom analizom ukupnih fenola, flavonoida, tanina i netanina u hidrolatu utvrđene su sledeće koncentracije: 151,91 mg CE/L ± 23,491 mg CE/L, 23,34 mg CE/L ± 3,978 mg CE/L, 119,75 mg CE/L ± 8,09 mg CE/L i 0,86 mg CE/L ± 0,07 mg Ru/L, redom. Hidrolat je pokazao antioksidativni potencijal u trima testovima: sposobnost uklanjanja slobodnih 2,2-diphenyl-1 picrylhydrazyl (DPPH) radikala, IC_{50} 0,51% ± 1,07%; ukupni antioksidativni potencijal 392,15 mmol Fe^{2+}/L ± 16,299 mmol Fe^{2+}/L i prevencija lipidne peroksidacije na način koji zavisi od koncentracije. Osim toga, zabeležena je korelacija sadržaja fenolnih jedinjenja i antioksidativne aktivnosti u hidrolatu. Pokazana antioksidativna aktivnost hidrolata *O. basilicum* može biti važna za njegovu buduću upotrebu u svojstvu potencijalnog prirodnog antioksidansa.

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Ključne reči: *antioksidans, antioksidativna moć redukcijom gvožđa, hidrosol, polifenoli, bosiljak*

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