

Composition of some macro and micro elements, polyphenol content, antimicrobial and antioxidant activity of *Achillea millefolium* (L.) grown in Kosovo

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This study aimed to determine the composition of several macro and micronutrients (N, P, K, Ca, Mg, Fe, Zn, Cu, Mn, Co, Ni, Cr, Cd and Pb), the composition of polyphenols (total polyphenols, total flavonoids and total phenolic acids), antimicrobial activity and antioxidant activity of the plant *Achillea millefolium* (L.) grown in Kosovo (the eastern part of Kosovo). The concentrations of the analysed macro and micronutrients are as follows: N 9.95 g/kg, P 6.15 g/kg, K 23.85 g/kg, Ca 21.98 g/kg, Mg 6.95 g/kg, Fe 59.85 mg/kg, Zn 35.42 mg/kg, Cu 10.75 mg/kg, Mn 31.87 mg/kg, Co 1.15 mg/kg, Ni 3.01 mg/kg, Cr 0.45 mg/kg, Cd 0.035 mg/kg and Pb 0.65 mg/kg. Potassium and calcium were the most abundant macroelements, while iron and zinc were the most abundant micronutrients. The antibacterial efficacy of *A. millefolium* (L.) was examined using methanol, ethyl acetate, diethyl ether, and water extracts and tested against *Staphylococcus aureus* (ATTC 25923), *Listeria monocytogenes* (WSLC 1042) and *Escherichia coli* (ATCC 25922). Antibacterial activity was determined using the agar disk diffusion method. The inhibition zones from the extracts were compared with that of penicillin G as the standard. Ethyl acetate and diethyl ether extracts showed antibacterial activity against *S. aureus* and *L. monocytogenes*, while methanol extracts showed antibacterial activity against *S. aureus*. The total content of polyphenols (determined using the Folin–Ciocâlteu reagent) was 17.5 ± 0.45 mg GAE/g (GAE-gallic acid equivalent). The total content of flavonoids (determined using the aluminum chloride method) was 8.45 ± 0.65 mg QE/g (QE-quercetin equivalent), and the total phenolic acid content (determined using Arnova reagent) was 13.2 ± 0.17 mg CAE/g (CAE-caffeic acid equivalent). The antioxidant activity determined using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) and phosphomolybdenum methods had values of 9.85 ± 0.15 and 185.7 ± 1.75 mg TE/g, respectively.

Keywords: *Achillea millefolium* (L.), antibacterial activity, antioxidant activity, macroelements, microelements, AAS

INTRODUCTION

Achillea spp., commonly known as yarrow, is widely used as a medicinal and aromatic herb worldwide [1]. There are about 110–140 known species

in the genus *Achillea* (Asteraceae). Most of them are located in Europe, Asia and North Africa [2, 3]. *Achillea millefolium* (L.) (*A. millefolium*) is an herbaceous plant that grows up to 80 cm tall.

The stem of this plant is erect, slender, and without branches, with white flowers arranged in

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umbels like an umbrella and gray-green leaves. It is widespread in mountainous areas, meadows, and occasionally in forests. It is found in low areas (500–600 m above the sea level) and reaches up to mountain altitudes of 2400 m above the sea level. It grows well in soils with a low fertility and a medium composition. This plant is highly resistant to drought and adapts to dry, barren soils, also resisting frost, although it prefers low mountainous areas. The plant is very common during the months of May and June and grows in all parts of Kosovo.

Achillea millefolium (L.) is used in both folk and official medicine [1]. *Achillea millefolium* (L.) has traditionally been used against skin inflammations, hepato-biliary and gastrointestinal complaints [4–6]. *Achillea millefolium* (L.) has been used internally as herbal tea and externally in lotions and herbal medications [1, 7–8].

Plants with potential antimicrobial activity should be tested against a suitable model microbe to confirm their activity [9]. The antimicrobial effect of plant extracts has been studied by several researchers in different parts of the world [10–13].

Regarding the bioactivity of *A. millefolium* (L.), recent studies have reported antioxidant and antimicrobial activities, antiphlogistic, hepatoprotective, gastrointestinal, antispasmodic, diuretic, urinary antiseptic, and calcium antagonist activities of its polar extracts [14–17]. It has also been reported that the essential oil of *A. millefolium* (L.) possesses disinfectant properties and has been used as a hemostatic agent [1, 7–8].

This study aims to determine the content of some macroelements (N, P, K, Ca and Mg), some microelements (Cu, Zn, Mn, Fe, Cr, Ni, Co, Pb and Cd), the content of polyphenols (total polyphenols, flavonoids and phenolic acids), antioxidant activity (DPPH and phosphomolybdenum methods) and the microbial activity of the plant *A. millefolium* (L.) grown in the Republic of Kosovo.

EXPERIMENTAL

MATERIALS AND METHODS

Plant material

The aerial part of the plant *A. millefolium* (L.) grown in the region of Anamorava in the south-eastern part of Kosovo (42°27'10"N, 21°30'25"E, Malishevë) was collected in May of the year 2023.

Voucher specimens were deposited in the herbarium of the Department of Plant Protection, University of Prishtina. The plant material was air-dried and ground in a mixer.

Determination of the content of mineral compounds

The material digestion was conducted using a microwave. For analysis, samples of approximately 0.5 g were placed in Teflon digestion vessels. Then, 7 mL of 65% HNO₃ and 1 mL of 30% H₂O₂ were added. The vessels were subsequently closed, tightened and placed in the rotor of the Analytikjena microwave digestion system. The digestion process followed this programming: Step 1 involved maintaining a temperature of 180°C for 10 min at 500 W power and 45 bar pressure. Step 2 maintained the same temperature for 15 min under the same power and pressure conditions. Finally, the vessels were cooled, carefully opened, and their contents quantitatively transferred into 50 mL calibrated flasks. Phosphorus concentration was determined colorimetrically using a spectrophotometer set to a wavelength of 882 nm [18]. Nitrogen concentration was determined using the Kjeldahl method [19]. Analysis of K, Ca, Mg, Cu, Zn, Mn, Fe, Cr, Ni, Co, Pb and Cd content was performed using a M Series spectrophotometer, model GE650416v1.26, in the Flame Mode. The device's working parameters (air, acetylene, optics and electronics) were adjusted to achieve the maximum absorption for each element. Analytical grade standard solutions (1000 mg/L) were sourced from Riedel de Haen (Germany). The experiment utilised ultra-pure-grade 65% nitric acid solution (Merck, Germany), and all solutions were prepared using deionised water. Each measurement was conducted three times.

Antibacterial activity

A portion of the finely powdered material (100 g) was extracted three times with 70% methanol (MeOH, 2 L) for 24 h. After removing the MeOH under reduced pressure, the aqueous phase was successively extracted with four solvents of increasing polarity (methanol, ethyl acetate, diethyl ether, and water). The extraction continued until a colourless extract was obtained. The residue was the aqueous extract. The solvents were evaporated using a vacuum rotary evaporator (EYELA

N-1000, Japan). The extraction process yielded methanol (10.39 g), ethyl acetate (4.0 g), diethyl ether (6.24 g), and water (12.68 g) extracts. Solvents (analytical grade) for extraction were obtained from commercial sources (Sigma-Aldrich, Merck).

The antibacterial activity of the methanol, ethyl acetate, diethyl ether, and water extracts of *A. millefolium* (L.) was determined using the Kirby–Bauer method [20] or the disk method (diameter = 5.5 mm, maximum capacity 10 mg). Organic extract samples were tested *in vitro* against bacterial strains: *S. aureus* (ATTC 25923), *L. monocytogenes* (WSLC 1042) and *E. coli* (ATCC 25922). Discs were previously wetted with a dimethylformamide (DMF) solution of the organic extracts at three different concentrations: 1, 3 and 5 mg/mL, and then placed in a Petri dish (diameter = 15 cm). The disks were incubated at 37°C for 48 h; a control was also maintained with penicillin G similarly dissolved in DMF.

Preparation of extracts

A 0.2 g sample was extracted with 20 mL of 80% ethanol for 2 h. The supernatant obtained by centrifugation was used to measure antioxidant activity, polyphenols, flavonoids, and phenolic acids. The extraction was performed in triplicate.

Total polyphenol content

Total polyphenol content was measured using the Folin Ciocalteu reagent according to Singleton and Rossi [21]. A 0.1 mL sample was mixed with 0.1 mL of Folin Ciocalteu reagent, 1 mL of 20% (w/v) sodium carbonate and 8.8 mL of distilled water, and left in the dark for 30 min. Absorbance at 700 nm was measured using a spectrophotometer. Gallic acid was used as a standard, and the results were expressed in mg/g gallic acid equivalents. The assay was performed in triplicate.

Total flavonoid content

Total flavonoids were determined using a method modified by Willett [22]. A 0.5 mL sample was mixed with 0.1 mL of a 10% (w/v) ethanolic solution of aluminum chloride, 0.1 mL of 1 M potassium acetate and 4.3 mL of distilled water. After 30 min in the dark, the absorbance was measured at a wavelength of 415 nm using a spectrophotometer. Quercetin was used as a standard, and the results

were expressed in mg/g quercetin equivalents. The assay was performed in triplicate.

Total phenolic acid content

Total phenolic acid content was determined using the method of Jain et al. [23]. A 0.5 mL extract sample was mixed with 0.5 mL 0.5 M hydrochloric acid, 0.5 mL Arnova reagent (10% NaNO_2 + 10% Na_2MoO_4), 0.5 mL sodium hydroxide 1 M (w/v) and 0.5 mL water. Absorbance was measured at a 490 nm wavelength using a spectrophotometer. Caffeic acid was used as a standard, and the results were expressed in mg/g caffeic acid equivalents. The assay was performed in triplicate.

Antioxidant activity by DPPH assay

The radical-scavenging activity of the samples was measured using 2,2-diphenyl-1-picrylhydrazyl (DPPH) [24]. 0.4 mL of the sample was mixed with 3.6 mL of DPPH solution (0.025 g of DPPH in 100 mL of ethanol). The absorbance of the reaction mixture was determined at a wavelength of 515 nm using a spectrophotometer. Trolox was used as a standard, and the results were expressed in mg/g Trolox equivalents. The assay was performed in triplicate.

Phosphomolybdenum method

The reducing power of the extracts was determined using the phosphomolybdenum method developed by Prieto et al. [25], with minor modifications. A mixture of sample, K_2HPO_4 (2.8 mL, 0.1 M), H_2SO_4 (6 mL, 1 M), $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24}$ (0.4 mL, 0.1 M) and distilled water (0.8 mL) was incubated at 90°C for 120 min and then rapidly cooled. The absorbance was measured at 700 nm using a spectrophotometer. Trolox served as the standard, and the results were expressed in mg/g equivalents. The assay was performed in triplicate.

Statistical analysis

Statistica 6.0 software, developed by Stat Soft, Inc., was utilised for all statistical calculations to determine the basic statistical parameters in this research [26].

RESULTS AND DISCUSSIONS

The composition of macroelements in the plant *A. millefolium* (L.) grown wild in the eastern part of Kosovo is shown in Table 1.

Table 1. Concentrations of some macroelements in *A. millefolium* (L.) in g/kg

Element	g/kg
N	9.95 ± 0.95
P	6.15 ± 0.45
K	23.85 ± 0.35
Ca	21.98 ± 1.15
Mg	6.95 ± 1.27

In our research, the concentration of macroelements in *A. millefolium* (L.) is as follows: N 9.95 g/kg, P 6.15 g/kg, K 23.85 g/kg, Ca 21.98 g/kg and Mg 6.95 g/kg (Table 1 and Fig. 1). The composition order of macroelements is as follows: K > Ca > N > Mg > P.

Nitrogen is a component of air as well as a component of living organisms [27]. It is a necessary element for the growth and development of plants. Nitrogen is one of the most important compounds in life. In nature, nitrogen is transformed during life processes. The concentration of nitrogen had a value of 9.95 g/kg, which was comparable to the concentration of nitrogen (10.1 g/kg) in research conducted in the city of Sivas in the central part of Turkey [28]. According to Koniecznyński and Wesołowski [29], *A. millefolium* (L.) grown in some cities in Poland contained nitrogen with an average value of 8.09 g/kg, which was comparable to the amount of nitrogen in our research, which had a value of 9.95 g/kg.

Phosphorus is the second-most abundant mineral in our body. About 85% of the total is found in bones, while the rest is deposited in soft tissues and extracellular fluids [27]. In fact, it is an essen-

tial substance in energy transformations at the cellular level. Phosphorus is also very important for the well-being of bones and teeth; it contributes to the pH regulation mechanism and plays an active role in the constitution of DNA and RNA, some sugars, and certain proteins. The concentration of phosphorus was 6.15 g/kg, which is almost the same as 6.3 g/kg in the research conducted in Turkey by Saraç and collaborators [28].

According to Koniecznyński and Wesołowski [29], *A. millefolium* (L.) grown in Poland contained phosphorus with an average value of 4.05 g/kg, which was comparable to the amount of phosphorus in our research, which had a value of 6.15 g/kg. Potassium is found in extracellular fluids and performs the same functions as Na. It also regulates neuromuscular excitation, heart rate, osmotic pressure, acid-base balance, and water retention [30]. The concentration of potassium had a value of 23.85 g/kg, which was comparable to the concentration of potassium in research conducted in the city of Sivas in the central part of Turkey [28].

Calcium is the most abundant cation in the body. It participates in the construction of cell membranes, increases the viscosity of protoplasm, participates in carbohydrate metabolism, and is necessary during fruit formation [31]. Calcium in the soil is found in the form of easily soluble salts (carbonates, sulfates and nitrates) and is easily accessible to plants as a Ca^{2+} ion. The concentration of calcium in our research was 21.98 g/kg, which is comparable to the research conducted in Turkey by Saraç et al. [28].

Magnesium is an essential element in plant and animal life [32]. Chlorophyll is what allows plants

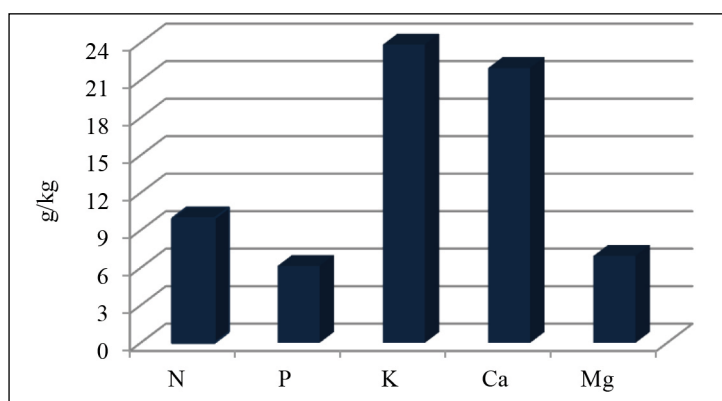


Fig. 1. Concentrations of nitrogen (N), phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg) in the *A. millefolium* (L.) plant

to receive sunlight and allow photosynthesis to occur. Without magnesium, photosynthesis cannot occur, and life as we know it would not exist. In humans, magnesium is essential for the work of hundreds of enzymes. The concentration of Mg was 6.95 g/kg. So, like the concentrations of N, P, K and Ca, the concentration of magnesium is comparable to the research conducted in the central part of Turkey [28]. The composition of micronutrients in the plant *A. millefolium* (L.) is presented in Table 2.

Microelements present in this plant, such as iron, zinc, manganese, nickel, cobalt, copper, iron, chromium, Pb and Cd, perform different and important functions in humans. Manganese is important for enzymatic activities and is also required for the formation of hemoglobin [33]. Copper is a vital component of many enzymes that catalyse redox reactions and is required for iron mobilisation and collagen synthesis [34].

Table 2. Concentrations of some microelements in *A. millefolium* (L.)

Element	mg/kg
Cu	10.75 ± 0.45
Zn	35.42 ± 0.22
Mn	31.87 ± 0.15
Fe	59.85 ± 0.49
Cr	0.45 ± 0.12
Ni	3.01 ± 0.22
Co	1.15 ± 0.14
Pb	0.65 ± 0.33
Cd	0.035 ± 0.11

Zinc is necessary for energy metabolism, tissue repair, and growth [35]. Iron is needed for hemoglobin production and oxygen transport [34]. Cobalt is an integral part of vitamin B-12 and is therefore essential for cell function. It is also involved in the production of red blood cells and the production of antibacterial and antiviral compounds that prevent infections. Cobalt also plays a key role in fat and carbohydrate metabolism as well as protein synthesis [36]. Nickel is also an essential trace element for the proper functioning of the human body, as it increases hormonal activity and is involved in lipid metabolism [37]. Chromium is a mineral required in small amounts by the body. It enables insulin to work. Although

chromium deficiency impairs insulin function, supplementation has not been shown to help people with diabetes, except for small changes in blood sugar [38].

According to this research, the concentrations of trace elements in *A. millefolium* (L.) are as follows: Cu 10.75 mg/kg, Zn 35.42 mg/kg, Mn 31.87 mg/kg, Fe 59.85 mg/kg, Cr 0.45 mg/kg, Ni 3.01 mg/kg, Co 1.15 mg/kg, Pb 0.80 mg/kg and Cd 0.035 mg/kg (Table 2 and Figs 2a, b). The composition order of microelements is as follows: Fe > Zn > Mn > Cu > Ni > Co > Pb > Cr > Cd. According to the research of Ivanišová with collaborators [39], the medicinal plant *A. millefolium* (L.) grown in the Slovak Republic contained Cu 11.52 mg/kg, Zn 39.10 mg/kg, Mn 39.10 mg/kg, Fe 62.60 mg/kg, Cr 0.90 mg/kg, Ni 3.80 mg/kg, Co 1.30 mg/kg, Pb 0.80 mg/kg and Cd 0.06 mg/kg. From this, we can conclude that the concentrations of these microelements were comparable to the concentrations in our research. Saraç et al. [28] found that *A. millefolium* (L.) in the city of Sivas in the central part of Turkey contained 360.4 mg/kg Fe, 47.6 mg/kg Zn, 85.5 mg/kg Mn and 28.3 mg/kg Cu. The concentrations of these trace elements were higher than in our research. According to Gogoasa et al. [40], *A. millefolium* (L.) grown in Romania contained Cu 9.88 mg/kg, Zn 40.2 mg/kg, Mn 84.3 mg/kg, Fe 179 mg/kg, Cr 0.26 mg/kg and Ni 5.54 mg/kg. The concentrations of Fe and Mn were higher, while the concentrations of Zn, Cu, Cr and Ni were comparable to those in our research. According to Koniecznyński and Wesołowski [29], *A. millefolium* (L.) grown in some cities in Poland contained an average zinc content of 44.92 mg/kg, which was almost comparable to the amount of zinc in our research, with a value of 35.42 mg/kg. The iron content in Poland was 21.74 mg/kg, significantly less than the amount found in this research, which had a value of 59.85 mg/kg.

The maximum permissible levels set by the World Health Organization (WHO) for cadmium, chromium and copper in raw plant materials are 0.3, 2 and 20 mg/kg, respectively [41]. This study revealed that micronutrient concentrations were below the values recommended by the World Health Organization (WHO) in 2005 [41].

In this study, the antibacterial activity of methanol, ethyl acetate, diethyl ether, and water

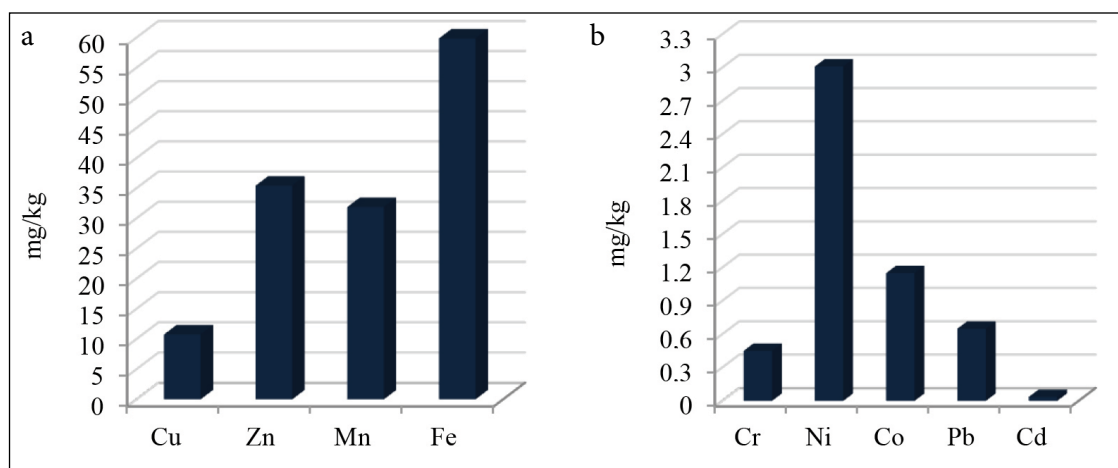


Fig. 2. (a) Concentrations of Cu, Zn, Mn and Fe; (b) concentrations of Cr, Ni, Co, Pb and Cd

extracts of this plant was evaluated against *S. aureus* (gram-positive), *E. coli* (gram-negative) and *L. monocytogenes* (gram-positive). The antibacterial activity was determined using the agar disk diffusion method. The zones of inhibition of the extracts were compared with those of penicillin G as a standard, as shown in Table 3. The methanol extracts (1, 3 and 5 mg/mL), along with diethyl ether and water (5 mg/mL), exhibited antibacterial activity against *E. coli* (Table 3). However, other extracts such as ethyl acetate (1, 3 and 5 mg/mL), diethyl ether and water (1 and 3 mg/mL) did not produce any zones of inhibition against *E. coli*, indicating no antibacterial activity. Notably, the methanol extract at a concentration

of 1 mg/mL demonstrated higher activity (6 mm) compared to penicillin G as a standard (4 mm).

Methanol (3 mg/mL), ethyl acetate, water, and diethyl ether extracts at concentrations of 1, 3 and 5 mg/mL exhibited antibacterial activity against *L. monocytogenes* (Table 3). Moreover, ethyl acetate, diethyl ether, and water extracts at a concentration of 1 mg/mL demonstrated higher activity (6 mm) than penicillin G (2 mm). However, methanol extracts at concentrations of 1 and 5 mg/mL did not produce any zones of inhibition, indicating no activity against *L. monocytogenes*.

Methanol (1 mg/mL), ethyl acetate and diethyl ether (1, 3 and 5 mg/mL) extracts showed antibacterial activity against *S. aureus* (Table 3).

Table 3. Antibacterial activities in *A. millefolium* (L.) organic extracts

Extract	Concentration, mg/mL	Inhibition zones diameters, mm		
		<i>E. coli</i> (ATCC 25922)	<i>L. monocytogenes</i> (WSLC 1042)	<i>S. aureus</i> (ATCC 25923)
Methanol	1	6	–	6
	3	6	6	–
	5	6	–	–
Ethyl acetate	1	–	6	6
	3	–	6	6
	5	–	6	6
Diethyl ether	1	–	6	6
	3	–	6	6
	5	6	6	6
Water	1	–	6	–
	3	–	6	–
	5	6	6	–
Penicillin	1	4	2	2
	3	6	6	6
	5	8	10	8

Methanol (3 and 5 mg/mL) and water (1, 3 and 5 mg/mL) extracts did not produce any zone of inhibition against *S. aureus*. Methanol, ethyl acetate, and diethyl ether extracts with a concentration of 1 mg/mL showed higher activity (6 mm) than the penicillin G standard with the same concentration (2 mm), while ethyl acetate and diethyl ether extracts with a concentration of 3 mg/mL showed the same activity (6 mm) as penicillin G. On the other hand, from Table 3, it can also be observed that the extracts of ethyl acetate and diethyl ether with a concentration of 5 mg/mL showed lower activity (6 mm) than penicillin G (8 mm). The antibacterial activity of *A. millefolium* (L.) is due to the presence of various secondary metabolites such as phenols and flavonoids. Therefore, this plant can be used to discover bioactive natural products that can serve as guides in the development of new pharmaceuticals. The total composition of polyphenols, flavonoids, and phenolic acids in extracts of *A. millefolium* (L.) grown in Kosovo is shown in Table 4.

Table 4. Total polyphenol, flavonoid and phenolic acid contents of *A. millefolium* (L.)

TPC – total polyphenol content (mg GAE/g dry matter)	17.5 ± 0.45
TFC – total flavonoid content (mg QE/g dry matter)	8.45 ± 0.65
TPAC – total phenolic acid content (mg CAE/g dry matter)	13.2 ± 0.17

Mean ± standard deviation; GAE, gallic acid equivalent; QE, quercetin equivalent; CAE, caffeic acid equivalent.

From Table 4 and Fig. 3, it can be seen that the composition of polyphenols, flavonoids, and phenolic acids is 17.5 ± 0.45 mg GAE/g, 8.45 ± 0.65 mg QE/g and 13.2 ± 0.17 mg CAE/g. According to Ivanišová et al. [38], *A. millefolium* (L.) grown in the Slovak Republic contained 19.65 GAE/g of polyphenols, 9.69 mg QE/g of flavonoids and 14.67 mg CAE/g of phenolic acids. Therefore, the content of polyphenols, flavonoids, and phenolic acids was comparable to the results of our research. Mehmood and collaborators [42] found that *A. millefolium* (L.) grown in Kashmir, Pakistan, contained 20.32 GAE/g of polyphenols, which was comparable to our research, while the content of flavonoids was higher at 25.67 mg QE/g. The content of polyphenols, according to Afshar et al. [43], in *A. millefolium* (L.) grown in Iran was 48.1 mg GAE/g, which is much higher than the amount found by us at 17.5 mg GAE/g. However, the content of flavonoids in Iran was 10.9 mg QE/g, which is comparable to our research, where it had a value of 8.45 mg QE/g. The antioxidant properties of *A. millefolium* (L.) extracts are given in Table 5.

Table 5. Antioxidant activity in *A. millefolium* (L.)

DPPH (mg TE/g dry matter)	9.85 ± 0.15
Reducing power (mg TE/g dry matter)	185.7 ± 1.75

Mean ± standard deviation; DPPH, 2,2-diphenyl-1-picrylhydrazyl; TEAC, Trolox equivalent.

The antioxidant activity, determined in the medicinal plant *A. millefolium* (L.) using the DPPH and phosphomolybdenum methods, had values of

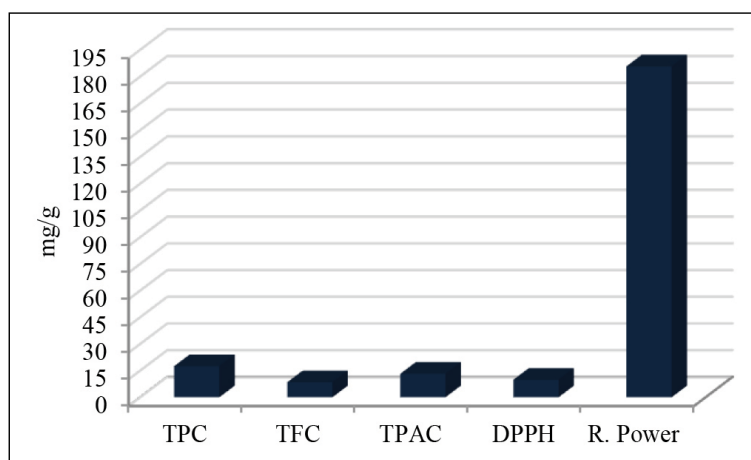


Fig. 3. The content of polyphenols, flavonoids, phenolic acids, and antioxidant activity (DPPH and reducing power) in the plant *A. millefolium* (L.)

9.85 ± 0.15 and 185.7 ± 1.75 mg TE/g, respectively (refer to Table 5 and Fig. 3). These values were comparable to the research conducted in the Slovak Republic [39].

The discrepancies observed between parameters reflecting antioxidant activity obtained using these two methods are mainly the result of methodological differences. Each method is based on a different principle, and each captures a range of different biologically active substances. DPPH[•] is a stable, dark purple free radical. Antioxidant compounds contained in the environment convert the DPPH[•] radical into a more stable molecular product, DPPH[•], by donating an electron or a hydrogen atom. The resulting change in the colour of the reaction solution from dark purple to pale yellow enables the spectrophotometric determination of antioxidant activity [44]. In the phosphomolybdenum method, the reducing power is generally related to the presence mainly of polyphenols, which act with their antioxidant effect in such a way that they break the free radical chain by providing a hydrogen atom [45].

The reducing capacity of the compounds can serve as an important indicator of their potential antioxidant activity [46]. From this, we can conclude that, to obtain the most accurate results, it is necessary to use several methods that are based on different principles when measuring antioxidant activity.

CONCLUSIONS

Nowadays, there is an increase in the use of medicinal and aromatic plants as people move away from synthetic products that contain chemicals and have serious side effects and turn to natural products. Since ancient times, medicinal plants have been used as food and for their healing effects. The use of medicinal plants and active substances obtained from them, due to their many therapeutic properties such as antioxidants, antimicrobials, and anticancer agents, has become inevitable in medicine and pharmacy. Many herbal medicines are sold in pharmacies, and many more are being developed. *Achillea millefolium* (L.) is one of these plants, which is the most widely used among medicinal plants. The determination of the content of macro and micro elements showed that they are present in different concentrations in the medicinal

plant *A. millefolium* (L.). Of the macroelements, the largest amount was potassium (23.85 g/kg) and calcium (21.98 g/kg). Among all micronutrients tested, iron proved to be dominant, with a concentration of 59.85 mg/kg. It is worth mentioning that a significant amount of zinc was also found with a concentration of 35.42 mg/kg. These trace elements are necessary for the human body, but in larger concentrations, they can have a harmful effect. The concentrations of trace elements were below the values recommended by the World Health Organization. Medicinal plants are an excellent source of flavonoids, carotenoids, and phenolic acids, which are powerful antioxidants. The obtained results are evidence of the antioxidant and antimicrobial potential of the medicinal plant *A. millefolium* (L.) and show that the widespread application of medicinal plants in the food, pharmaceutical and cosmetic industries is justified. The possibility of using natural raw materials as an alternative to synthetically produced products should be given more attention.

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