

PARAMETERS OF ANTIOXIDANT ACTIVITY OF *GALEGA OFFICINALIS* L. AND *GALEGA ORIENTALIS* LAM. (*FABACEAE* LINDL.) PLANT RAW MATERIAL

Olena Vergun, Oksana Shymanska, Dzhamal Rakhmetov, Olga Grygorieva, Eva Ivanišová,
Jan Brindza

ABSTRACT

The plant raw material of *Galega officinalis* L. (goat's rue) and *Galega orientalis* Lam. (fodder galega) investigated in this study. These species are known as fodder crops with high productivity of green mass and as medicine plants. The current study was aimed to evaluate an accumulation in dry raw of selected plants the total content of phenolic acids (TPA) and flavonoids (TFC) as compounds with antioxidant activity (AA) by spectrophotometric method. AA by DPPH-method and phosphomolybdenum method (reducing power (RP)) was measured. Study of ethanolic extracts of *G. officinalis* showed accumulation of TPA in different organs in range from 3.65 to 15.17 mg.g⁻¹ caffeic acid equivalent (CAE) and TFC from 10.08 to 65.75 mg.g⁻¹ quercetin equivalent (QE), AA by DPPH-method from 6.02 to 8.45 mg.g⁻¹ Trolox equivalent (TE) and RP of extracts by phosphomolybdenum method from 86.56 to 288.15 mg TE.g⁻¹. In extracts of *G. orientalis* was identified TPA from 3.52 to 18.52 mg CAE.g⁻¹ and TFC from 6.09 to 46.72 mg QE.g⁻¹, antioxidant activity by DPPH-method from 6.80 to 8.48 mg TE.g⁻¹ and antioxidant capacity by phosphomolybdenum method from 52.52 to 188.51 mg TE.g⁻¹. It was established that less concentration of studied compounds found in the stems for both species. It should be noted that the content of phenolic acids in the leaves was decreased and flavonoids in stems increased during vegetation for both species. Content of phenolic acids in the generative organs and flavonoids in the leaves decreased in raw of *G. orientalis* during vegetation. Pearson's correlation analysis demonstrated very strong relations between TFC and AA by DPPH, TPA and RP, TFC and RP for *G. officinalis* extracts. Very strong correlation in the extracts of *G. orientalis* found between TFC and RP, TPA and RP. Obtained results can be used in the further biochemical and pharmacological study.

Keywords: *Galega officinalis*; *Galega orientalis*; antioxidant activity; flavonoids; phenolic acids

INTRODUCTION

Study of the antioxidant activity and compounds that cause it very widespread and actually in modern biological science (Carocho and Ferreira, 2013; Kumar, Sharma and Vasudeva, 2017). Plant raw material of medicinal (Adámková, Kouřimská and Kadlecová, 2015; Vergun et al., 2019b), food (Frusciante et al., 2007; Mendelová et al., 2016), forage (Sang et al., 2014; Petrović et al., 2016; Vergun et al., 2018), fruit (Ivanišová et al., 2017; Horčinová Sedláčková et al., 2018; Brindza et al., 2019) and other plant groups and their products are a valuable source of antioxidant compounds of different nature. Leguminous plants (*Fabaceae* Lindl.) are a perspective group of crops, which ecological and economic function is important in agriculture. It is one of the most important plant families in the production of food for humans and livestock, as well as in the production of industrial products. These crops have provided interesting as forage grasses with high productivity and play an important role

as N fixators (Peiretti, 2009; Teleută et al., 2015). Plants from the *Fabaceae* family are of interest in relation to biologically active compounds, especially individuals, in different organs (Danilčenko et al., 2017).

Among economically important leguminous plants can be highlight goat's rue (*Galega officinalis* L.) and fodder galega (*Galega orientalis* Lam.). Plants of species of *Galega* L. are valuable perennial and productive crops with the protein-rich chemical composition of plant raw material (Baležentienė, 2008). They widespread in natural flora and are characterized by high productivity of seeds (Tkacheva, Vinogradova and Pavlova, 2011). Results obtained by Peiretti (2009) showed that *G. officinalis* has the potential for large-scale ensiling if plants are harvested at the budding stage or during regrowth. These species cultivated as medicinal plants due to the biochemical composition of plant raw material and as garden plants (Baležentienė and Spruogis, 2011; Kumar et al., 2012). As reported Kiselova et al. (2006), plants of *G. officinalis*

use in traditional phytotherapy due to hypoglycemic and diuretic properties. Also, the hypoglycemic and weight-reducing ability of this species was described in some reports (Lemus et al., 1999; Hasani-Ranjbar et al., 2009; Shojaee et al., 2015).

As described in some reports, plants of *G. officinalis* use in traditional phytotherapy due to hypoglycemic, diuretic properties, and weight-reducing ability (Modak et al., 2007; Hasani-Ranjbar et al., 2009; Shojaee et al., 2015). These plants as a source of metformin use to treat diabetes and use in the pharmacology (Umashanker and Shruti, 2011; Khodadidi, 2016; Abhati-Evari et al., 2017; Luka, Adoga and Istifanus, 2017). Leaves of *G. officinalis* are a source of bioactive secondary metabolites (Pehlivan Karakas, Sahin and Türker, 2016a).

Biochemical composition of *Galega* species raw is ascorbic acid, carotene, soluble sugars, lipids, protein, ash, alkaloids, macroelements, etc. (Symanowicz and Kalembasa, 2012; Vergun, Shymanska and Rakhmetov, 2012; Shymanska et al., 2017). Also, the phytochemicals screening revealed that in aqueous, methanolic, ethanolic and acetone extracts were found flavonoids, tannins, cardiac glycosides, triterpenoids, and steroids. Methanolic extracts of goat's rue significantly improved the lipid profile in a clinical study (Peirs et al., 2006; Luka, Adoga and Istifanus, 2017). As pointed out Pehlivan Karakas, Yildirim and Türker (2012), different extracts of *G. officinalis* showed broad-spectrum activity against both gram-positive and gram-negative bacteria. Moreover, different extracts of goat's rue exhibited cytotoxic, anti-inflammatory and antioxidant activity (Pehlivan Karakas et al., 2016b).

As reported by Meripöld et al. (2017), the first cut of *G. orientalis* advisable to use as a bioenergy crop and the second cut as forage. Also, fodder galega was the object of allelopathic study. Experimental evidence obtained by Baležentienė (2009) and Baležentienė and Kusta (2011) suggests that shoots of fodder galega are the main source of its allelochemicals, especially at flowering stage. As emphasizes Ignat, Volf and Popa (2011), the main group of biochemical compounds among allelochemicals is phenolic compounds. According to Symanowicz et al. (2015), nitrogen fertilization significantly increased the dry matter yield of fodder galega.

Investigations of oxidative properties of *Galega* species

indicated that plant raw material is the source of antioxidants with different natures (Maslennikov, Chupakhina and Skrypnik, 2014; Shymanska et al., 2018a; Shymanska et al., 2018b). Also, high antioxidant activity found in the seed extracts (Vergun et al., 2019a).

Nonetheless, it is necessary to carry out a study with plants of a genus of *Galega* as a source of important biologically active compounds. The aim of this study was to determine the peculiarities of accumulation of compounds with phenolic nature that can detect the antioxidant status of investigated plants as important crops.

Scientific hypothesis

Comparative assessment of the accumulation of phenolic compounds and determination of the antioxidant activity of two species of *Galega* L. genus during vegetation.

MATERIAL AND METHODOLOGY

Conditions of plant growing

The plants were grown in 2017 – 2018 at the experimental fields of the M. M. Gryshko National Botanical Garden of the NAS of Ukraine in the Kyiv city (50°24'55"N, 30°33'45"E).

Biological material

Observation on plants was conducted in the experimental collection of the Cultural Flora Department of M. M. Gryshko National Botanical Garden of the NAS of Ukraine (Figure 1). Plant raw material of two species – *Galega officinalis* and *G. orientalis* were collected in the stages according to Biologische Bundesanstalt, Bundessortenamt and Chemische Industrie (BBCH) coding system (Meier, 2018). According to the BBCH scale, plant samples were taken at the phenological growth stages described for faba bean (*Vicia faba* L.). Four principal growth stages were assigned: leaf development (19 – nine or more leaves infolded), inflorescence emergence (50 – flower buds present, still enclosed by leaves), flowering (65 – full flowering: flowers open on 5 racemes per plant), and ripening (80 – beginning of ripening: seed green, filling pod cavity). For chemical analyses plant raw material was dried at 35 °C for three days (Müller and Heindl, 2006). After this, the samples were milled in the powder condition. All biochemical analyses were done in the Slovak University of Agriculture



Figure 1 *Galega officinalis* L. and *Galega orientalis* Lam. in the stage of flowering.

in Nitra (Slovak Republic).

Sample preparation

For planned analyses, 0.2 g of milling fraction was extracted with 20 mL of 80% ethanol for 24 hours. After centrifugation at 4000 g with Rotofix 32 A (Hettich, Germany) for 20 min, the supernatant was used for measurement (phenolic acids, flavonoids, antioxidant activity by DPPH-method and reducing power of extracts).

Total phenolic acid content (TPAC)

The content of phenolic acids was determined using **Farmakopea Polska (1999)**. 0.5 mL of sample extract was mixed with 0.5 mL of 0.5 M hydrochloric acid, 0.5 mL Arnova reagent, 0.5 mL of 1 M sodium hydroxide (w/v) and 0.5 mL of distilled water. Absorbance at 490 nm was measured using the spectrophotometer Jenway (6405 UV/VIS, England). Caffeic acid 1 – 200 mg L⁻¹ ($R^2 = 0.999$) was used as a standard. The results were expressed in mg.g⁻¹ caffeic acid equivalents (CAE).

Total content of flavonoids (TFC)

Analise was conducted according to the procedure which was described by **Shafii et al. (2017)**. 0.5 mL of sample extract was mixed with 0.1 mL of 10% (w/v) ethanolic solution of aluminium chloride, 0.1 mL of 1 M sodium acetate and 4.3 mL of distilled water. After 30 min. in darkness the absorbance at 415 nm was measured using the spectrophotometer 6405 UV/VIS (Jenway, England). Quercetin 0.01 – 0.5 mg L⁻¹ ($R^2 = 0.997$) was used as the standard and the results were expressed in mg.g⁻¹ quercetin equivalents (QE).

Antioxidant activity (AA)

Radical scavenging activity of samples was measured using 2,2-diphenyl-1-picrylhydrazyl (DPPH) according to **Sánchez-Moreno, Larrauri and Saura-Calixto (1999)** with slight modification. The ethanol extract (1 mL) was mixed with 4 mL of DPPH solution (0.025 g of radical in 100 mL of ethanol). The absorbance of the sample extract was determined using the spectrophotometer Jenway (6405 UV/VIS, England) at 515 nm. Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) 10 – 100 mg L⁻¹ ($R^2 = 0.983$) was used as a standard and the results were expressed in mg.g⁻¹ Trolox equivalents (TE).

Reducing power of extracts

Reducing power of extracts was determined by the phosphomolybdenum method of **Prieto, Pineda and Aguilar (1999)** with slight modifications. The mixture of 1 mL of sample, 2.8 mL of monopotassium phosphate (0.1 M), 6 mL of sulfuric acid (1 M), 0.4 mL of ammonium heptamolybdate (0.1 M) and 0.8 mL of distilled water was incubated at 90 °C for 120 min, then rapidly cooled and detected by monitoring absorbance at 700 nm using the spectrophotometer Jenway (6405 UV/VIS, England). Trolox 10 – 1000 mg L⁻¹ ($R^2 = 0.998$) was used as the standard and the results were expressed in mg.g⁻¹ TE.

Statistical analysis

The statistically treated data are given in tables as the arithmetical mean values and their standard errors. Data were submitted ANOVA and differences between means compared through the Tukey-Kramer test ($\alpha = 0.05$). Correlation analysis was performed using Pearson's criterion.

RESULTS AND DISCUSSION

Antioxidant compounds common nowadays play an important role in protecting factors that explain reducing the risk of different chronic diseases and belong to various classes of biochemical compounds. Phenolic compounds are widespread secondary metabolites in plant extracts, and it possesses various biological activities such as antioxidant, anticarcinogenic, antimicrobial, antiallergic, antimutagenic, anti-inflammatory etc. (**Tatiya et al., 2011; Najafabad Morabbi and Jamei, 2014**). In addition, they play an important role in plant resistance (**Kulbat, 2016**). The antioxidant compounds are the natural defense system to protect the plants from abiotic and biotic stresses such as salinity and drought. They play a key role in defense mechanisms against the free radicals which cause the deleterious effect to plant organisms (**Govindaraj et al., 2017**). A high level of antioxidant agents in medicinal plants can be proposed as an effective therapeutic approach (**Saeed, Khan and Shabbir, 2012**).

Phytonutrients found in different parts of plants are powerful antioxidants, especially compounds with phenolic nature. Polyphenol compounds from many legumes can be represented by different compounds such as phenolic acids (gallic, ellagic), hydrolyzable tannins (**Akbarirad et al., 2016**). In some studies, flavonoids indicated as the main components responsible for antioxidant capacity (**Zhang et al., 2012**). It is a large group of secondary metabolites, which play a variety of significant functions in plants. They play a role as signal molecules, phytoalexins, detoxifying agents, as UV-filters, pollinator attractants etc. (**Samanta, Das and Das, 2011**). Moreover, as summed up by **Asif (2015)**, flavonoids exhibit a wide spectrum of biological activity such as antioxidant, antibacterial, antiviral, anti-inflammatory, anti-allergic, anti-thrombotic.

Antioxidant activity of plant extracts can be measured by different methods. Free radical scavenging activity by DPPH method is a simple assay for antioxidant activity evaluation (**Marinova and Batchvarov, 2011; Tatiya et al., 2011; Shekhar and Anju, 2014**). DPPH radical shows the reduction capability by the decrease in absorbance induced by investigated plant extract. In our study reducing power of plant extracts measured with Trolox as standard while its can be, for example, ascorbic acid (**Tatiya et al., 2011**).

In Table 1 and Table 2 are shown data of accumulation of total phenolic acids and total flavonoids content in vegetative and generative organs of *G. officinalis* and *G. orientalis* during vegetation. Polyphenol compounds can form several hydrogen bonds and even ionic bonds with most proteins. They modulate the activity of many proteins, involving enzymes, ion channels, etc. As a consequence, many polyphenols are pharmacologically active, being among antioxidants, anti-inflammatory, antibacterial, antifungal, and antiviral (**Wink, 2013**).

Maslennikov, Chupakhina and Skrypnik (2014) reported that *G. officinalis* leaves contain 3.6 mg GAE.g⁻¹ of polyphenol compounds. According to Tusevski et al. (2014), this parameter for *G. officinalis* plants was 32.53 mg GAE.g⁻¹. Pehlivan Karakas, Sahin and Türker (2016a) obtained twenty phenolics compounds from methanolic leaves extracts of *G. officinalis*. The total phenolic content, in this case, was 36.69 mg.g⁻¹ of dry extract. According to Salata and Gruszecki (2010), roots and leaves in vegetative stages of plants contained considerably more phenolic acids than the beginning of the flowering period, while in leaf nodes more polyphenolic compounds were marked during flowering than at the vegetative stage. Total phenolic acids in plant raw material of investigated *G. officinalis* plants were in the range from 3.65 to 15.17 mg CAE.g⁻¹ depending on the phase of growth (Table 1).

Flavonoids belong to derivatives of simple phenols, and their synthesis increases at the stress conditions due to microbial infections, injury, deficiency of nutrients, changing of temperature, etc. (Kulbat, 2016). Flavonoids are biologically active compounds that possess the ability to capture radicals and play a significant role in agriculture and pharmaceutical chemistry as anti-hyperglycemic, anti-cancerous, anti-allergic, anti-viral, immune-stimulating activity (Sulaiman et al., 2013; Marella, 2017).

The level of flavonoid accumulation in raw of *G. officinalis* during vegetation was in the range from 10.08 to 67.75 mg QE.g⁻¹. The study of Tusevski et al. (2014) resulted that the concentration of flavonoids in this plant was 8.95 ± 0.13 mg CAE.g⁻¹.

As shown in Table 2 phenolic acid content of *G. orientalis* plant raw material ranged from 3.52 to 18.52 mg CAE.g⁻¹ during vegetation. The concentration of flavonoids ranged from 6.09 to 46.72 mg QE.g⁻¹. According to Baležentienė (2009) report, the highest total content of phenols was determined at the budding stage which was characterized as the most intensive growth period of the plant shoot.

Also, it should be noted that minimal content of phenolic acids and flavonoids in the plant raw material of both investigated species during vegetation was identified in stems. Obtained data showed that higher accumulation of investigated biochemical parameters was different for two species. So, flavonoids content for *G. officinalis* were maximal in inflorescences and phenolic acids – in leaves (inflorescence emergence stage). For *G. orientalis* total content of phenolic acids and flavonoids was maximal in leaves (inflorescence emergence stage).

There are a great number of methods for the determination of antioxidant capacity based on different principles. One of them is the DPPH method that is rapid, simple, and accurate (Marinova and Batchvarov, 2011; Pisoschi et al., 2016). The method is based on the scavenging of DPPH through the addition of a radical species or antioxidant that decolorizes the radical solution

(Saeed, Khan and Shabbir, 2012). Trolox Equivalent Antioxidant Capacity assay is widely used to evaluate the antioxidant property of investigated products (Kumar, Sharma and Vasudeva, 2017). Figure 2 demonstrates the antioxidant capacity by DPPH-method of plant raw material of *Galega* species during vegetation.

We found that the antioxidant activity of investigated ethanol extracts of *G. officinalis* was in the range from 6.02 to 8.45 mg TE.g⁻¹. For *G. orientalis* extracts this parameter ranged from 6.80 to 8.48 mg TE.g⁻¹.

Also, the antioxidant capacity can be illustrated by the reducing power of investigated extracts as an important indicator. According to some studies, there is some connection between antioxidant activity and reducing power (Zhang et al., 2012). In our experiment extracts analyzed spectrophotometrically through the phosphomolybdenum method, based on the reduction of Mo (VI) to Mo (V) (Kumar, Sharma and Vasudeva, 2017; Saeed, Khan and Shabbir, 2012; Ravishankar, Kiranmayi and Prasad, 2018). The present study demonstrated that ethanol extracts of *G. officinalis* and *G. orientalis* had antioxidant activity during vegetation ranged between 86.56 – 288.15 and 52.52 – 188.51 mg TE.g⁻¹, respectively (Figure 3).

Correlation analysis was used to explore the relationships between the polyphenols, phenolic, flavonoids compounds and antioxidant capacities (by DPPH and phosphomolybdenum methods) measured for all plant extracts of *Galega officinalis* and *Galega orientalis* (Table 3, Table 4). The results of this study have demonstrated that investigated antioxidant components in two species of *Galega* L. had a correlation between different parameters of an experiment during vegetation. In the period of inflorescence emergence we found a very strong correlation between TPA and TF for both *G. officinalis* and *G. orientalis* (0.985 and 0.950 respectively). Very strong correlation found between AA by DPPH and RP of extracts for *G. officinalis*. The relation between TPA accumulation and RP and TFC and RP was moderate for *G. officinalis* (0.433 and 0.583 respectively). For *G. orientalis* in this period between TPA and RP was strong relation (0.750) and between TFC and RP very strong (0.919). In the period of flowering indicated that very strong correlation found for both species between TFC and RP. Dramatically strong correlation detected between TFC and AA by DPPH (0.999) for *G. officinalis* in this stage. However, in this case for *G. orientalis* found a very weak correlation (0.033). Also, very strong relations found between TPA, TFC and RP for *G. officinalis* (0.924 and 0.851 respectively). In this case values of coefficient of correlation were 0.670 (strong) and 0.953 (very strong) for *G. orientalis*. Different values found for two species regarding the relation between AA by DPPH and RP (very strong for *G. officinalis* and weak for *G. orientalis*).

Table 1 The total content of phenolic acids and flavonoids in plant raw material of *Galega officinalis* L. during vegetation.

Phase of growing	Organ of plant	Total phenolic acids, mg CAE.g ⁻¹	Total flavonoid content, mg QE.g ⁻¹
Leaf development	aerial part	14.13 ±0.89 ^a	44.27 ±2.97 ^b
Inflorescence emergence	leaves	15.17 ±0.12 ^a	55.61 ±0.75 ^a
	stems	6.47 ±0.20 ^c	10.08 ±0.94 ^{ef}
	buds	12.44 ±0.07 ^b	48.91 ±1.14 ^b
Flowering	leaves	11.62 ±0.25 ^b	44.91 ±1.12 ^b
	stems	3.65 ±0.24 ^c	13.18 ±0.86 ^e
	inflorescences	7.70 ±0.48 ^c	67.75 ±5.05 ^a
Ripening	leaves	10.15 ±0.28 ^b	32.24 ±0.29 ^c
	stems	9.62 ±0.19 ^b	24.78 ±0.57 ^d
	fruits	5.89 ±0.29 ^c	16.86 ±0.55 ^e

Note: Means in columns followed by different letters are different at $p = 0.05$; each value represents the mean of three independent experiments (\pm SD); GAE – gallic acid equivalents; CAE – caffeic acid equivalents; QE – quercetin equivalents.

Table 2 The total content of phenolic acids and flavonoids in plant raw material of *Galega orientalis* Lam. during vegetation.

Phase of growing	Organ of plant	Total phenolic acids mg CAE.g ⁻¹	Total flavonoid content mg QE.g ⁻¹
Leaf development	aerial part	16.73 ±0.52 ^a	38.79 ±0.83 ^b
Inflorescence emergence	leaves	18.52 ±1.64 ^a	46.72 ±0.26 ^a
	stems	4.37 ±0.19 ^d	6.09 ±0.67 ^e
	buds	10.89 ±0.77 ^b	36.21 ±0.55 ^b
Flowering	leaves	16.12 ±0.19 ^a	40.09 ±0.48 ^a
	stems	3.52 ±0.39 ^d	6.74 ±0.51 ^e
	inflorescences	7.38 ±0.16 ^c	32.63 ±0.79 ^{bc}
Ripening	leaves	12.57 ±0.38 ^b	35.59 ±0.48 ^b
	stems	4.05 ±0.30 ^d	14.47 ±0.46 ^d
	fruits	4.25 ±0.24 ^d	9.29 ±0.44 ^d

Note: Means in columns followed by different letters are different at $p = 0.05$; each value represents the mean of three independent experiments (\pm SD); CAE – caffeic acid equivalents; QE – quercetin equivalents.

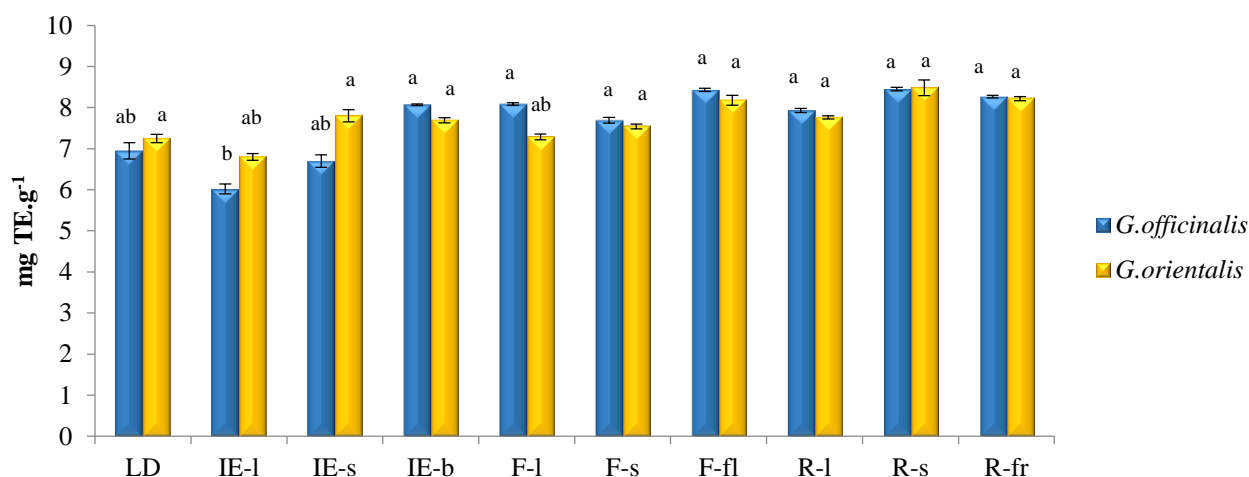


Figure 2 The antioxidant activity of ethanol extracts of *Galega officinalis* L. and *G. orientalis* Lam. by DPPH-method during vegetation. Note: LD – leaf development; IE-l – inflorescence emergence, leaves; IE-s – inflorescence emergence, stems; IE-b – inflorescence emergence, buds; F-l – flowering stage, leaves; F-s – flowering stage, stems; F-fl – flowering stage, inflorescences; R-l – ripening, leaves; R-s – ripening, stems; R-fr – ripening, fruits; means in columns followed by different letters are different at $p = 0.05$; each value represents the mean of three independent experiments (\pm SD).

Table 3 Coefficient of correlation between investigated parameters of *Galega officinalis* L. extracts during vegetation.

Characters	TPAC	TFC	DPPH
Inflorescence emergence stage			
TFC	0.985*	1	
DPPH	-0.120	0.055*	1
RP	0.433*	0.583	0.843
Flowering			
TFC	0.587	1	
DPPH	0.548*	0.999	1
RP	0.924*	0.851*	0.825
Ripening			
TFC	0.924	1	
DPPH	-0.247	-0.598*	1
RP	0.551*	0.828*	-0.945

Note: Significant according to the t-test ($p < 0.05$).

Table 4 Coefficient of correlation between investigated parameters of *Galega orientalis* L. extracts during vegetation.

Characters	TPA	TFC	DPPH
Inflorescence emergence stage			
TFC	0.950	1	
DPPH	-0.929*	-0.768	1
RP	0.750*	0.919*	-0.454*
Flowering			
TFC	0.863*	1	
DPPH	-0.476	0.033*	1
RP	0.670*	0.953	0.334
Ripening			
TFC	0.979*	1	
DPPH	-0.941	-0.852*	1
RP	0.880*	0.959	-0.669*

Note: Significant according to the t-test ($p < 0.05$).

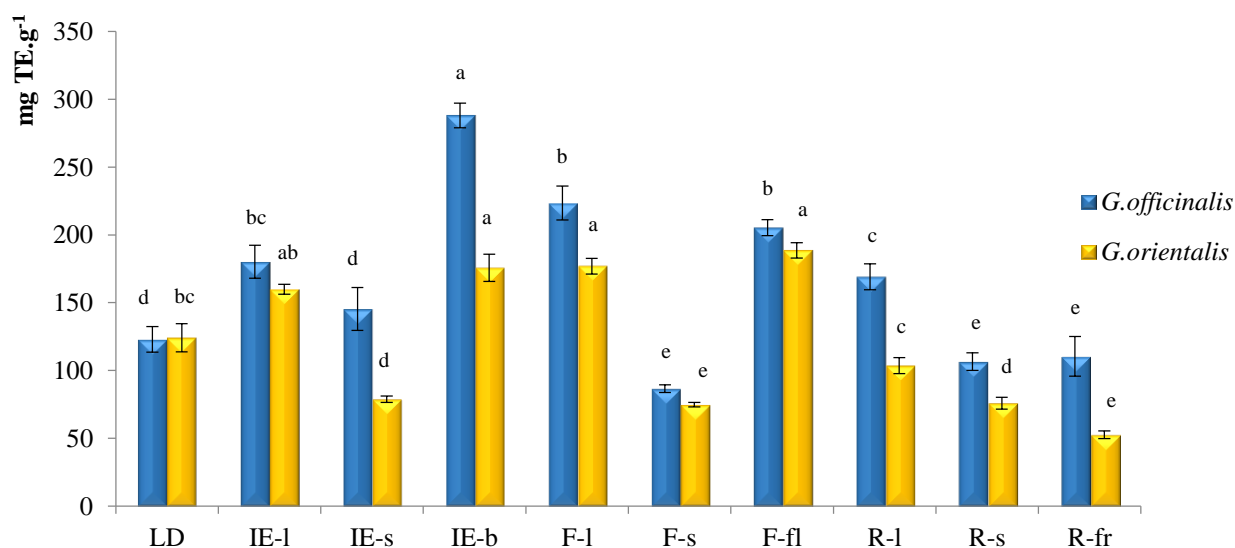


Figure 4 The reducing power of ethanol extracts of *Galega officinalis* L. and *G. orientalis* Lam. during vegetation. Note: LD – leaf development; IE-l – inflorescence emergence, leaves; IE-s – inflorescence emergence, stems; IE-b – inflorescence emergence, buds; F-l – flowering stage, leaves; F-s – flowering stage, stems; F-fl – flowering stage, inflorescences; R-l – ripening, leaves; R-s – ripening, stems; R-fr – ripening, fruits; means in columns followed by different letters are different at $p = 0.05$; each value represents the mean of three independent experiments (\pm SD).

At the period of ripening determined the very strong correlation between TPA and TFC, TFC and RP for both investigated species. Correlation between TPA and RP was moderate for *G. officinalis* (0.551) and very strong for *G. orientalis* (0.880). It should be noted that for both species was found a negative correlation between TPA and AA by DPPH, TFC and AA by DPPH, and between AA by DPPH and RP. The direction of correlation between antioxidant components depends on their nature, thus, different types of phenolics possess different antioxidant activity (Vamanu et al., 2011).

According to previous studies, it should be noted that phenolic extracts exhibited different antioxidant activity that depends on their structure (Tatiya et al., 2011). The study of relationships between phenolic compounds and antioxidant activity demonstrated a significant correlation (Li, Wu and Huang, 2009). Some results, also, not confirmed correlations between the content of phenolic compounds and antioxidant activity (Vamanu et al., 2011). Moreover, according to Vamanu et al. (2011), reducing power and antioxidant activity correlated with extract concentration. In some cases, there wasn't found relationship between phenolic compounds content and antioxidant activity.

CONCLUSION

Based on the results obtained in this study concluded that two investigated species of *Galega* L. as medicine and forage cultures characterized by plant raw material with high antioxidant activity. The maximal content of phenolic acids for both investigated species was found in the leaves in the period at the inflorescence emergence, flavonoids in inflorescences at the flowering stage for *G. officinalis* and in the leaves at the inflorescence emergence period for *G. orientalis*. The least content of phenolic acids and flavonoids identified in the stems of both investigated species. Ethanolic extracts of stems of *G. officinalis* and *G. orientalis* plants exhibited the most antioxidant activity at the ripening stage by DPPH-method. Reducing power of ethanol extracts was higher for *G. officinalis* in the buds, for *G. orientalis* in the inflorescences. Pearson's correlation analysis (at $p < 0.05$) showed very strong values of coefficient of variation between TFC and AA by DPPH (0.999), TPA and RP (0.924), TFC and RP (0.828) for *G. officinalis* depending on the stage of growth. The highest correlation found in extracts of *G. orientalis* between TFC and RP at every investigated stage (0.919, 0.953, and 0.959), between TPA and RP (0.880). Obtained results demonstrated that two investigated species of *Galega* are a good source of antioxidant compounds with polyphenol nature such as phenolic acids and flavonoids. These data can provide further information about an accumulation of flavonoids and phenolic acids in *Galega* spp. raw that possess antioxidant activity and also can be used in pharmacological investigations.

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Contact address:

Mgr. Olena Vergun, PhD, M. M. Gryshko National Botanical Garden of the NAS of Ukraine, Cultural Flora Department, Timiryazevska 1, 04014, Kyiv, Ukraine, Tel.: +380975398541,

E-mail: olenavergun8003@gmail.com

ORCID: <https://orcid.org/0000-0003-2924-1580>

Mgr. Oksana Shymanska, PhD, M. M. Gryshko National Botanical Garden of the NAS of Ukraine, Cultural Flora Department, Timiryazevska 1, 04014, Kyiv, Ukraine, Tel.: +380982284804,

E-mail: galega777@ukr.net

ORCID: <https://orcid.org/0000-0001-8482-5883>

Prof. Dzhamal Rakhmetov, M. M. Gryshko National Botanical Garden of the NAS of Ukraine, Cultural Flora Department, Timiryazevska 1, 04014, Kyiv, Ukraine, Tel.: +380503561930,

E-mail: jamal_r@bigmir.net

ORCID: <https://orcid.org/0000-0001-7260-3263>

*Mgr. Olga Grygorieva, PhD, M. M. Gryshko National Botanical Garden of the NAS of Ukraine, Department of Fruit Plants Acclimatisation, Timiryazevska 1, 04014, Kyiv, Ukraine, Tel.: +380671988082,

E-mail: ogrygorieva@gmail.com

ORCID: <https://orcid.org/0000-0003-1161-0018>

Ing. Eva Ivanišová, PhD, Slovak University of Agricultural in Nitra, Faculty of Biotechnology and Food Resources, Department of Plant Storage and Processing, Trieda Andreja Hlinku 2, 949 76 Nitra, Slovakia, Tel: +421376414421,

E-mail: eva.ivanisova@uniag.sk

ORCID: <https://orcid.org/0000-0001-5193-2957>

doc. Ing. Jan Brindza, PhD, Slovak University of Agricultural in Nitra, Faculty of Agrobiological and Food Resources, Institute of Biological Conservation and Biosafety, Trieda Andreja Hlinku 2, 949 76 Nitra, Slovakia, Tel: +421376414787,

E-mail: Jan.Brindza@uniag.sk

ORCID: <https://orcid.org/0000-0001-8388-8233>

Corresponding author: *