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A comparative study of *Silphium* spp. antioxidant activity

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This study aimed to evaluate the parameters of antioxidant activity of different parts of *Silphium* spp. during budding and flowering periods. The four species of *Silphium* L. genus from the experimental collection of the Cultural Flora Department of M.M. Gryshko National Botanical Garden of the NAS of Ukraine were prepared and used in this research. Total polyphenol content was from 12.32 (*S. asperrimum*, stems at the budding) to 95.21 (*S. laciniatum*, leaves at the flowering stage) mg GAE.g^{-1} DW depending on the organ and stage of vegetation. The total content of phenolic acids in ethanol extracts of investigated plants was from 3.52 (*S. asperrimum*, buds at the budding) to 34.58 (*S. laciniatum*, leaves at the flowering) mg CAE.g^{-1} DW. The total flavonoid content in investigated extracts was from 3.67 (*S. perfoliatum*, stems at the budding) to 57.31 (*S. laciniatum*, leaves at the budding) mg QE.g^{-1} DW. Antioxidant activity was determined by two assays namely the DPPH method and molybdenum-reducing power of extracts and was from 3.33 (*S. trifoliatum*, stems at the budding) to 9.11 (*S. laciniatum*, flowers at flowering) mg TE.g^{-1} and from 29.11 (*S. trifoliatum*, stems at the budding) to 185.22 (*S. laciniatum*, leaves at the flowering) mg TE.g^{-1} DW, respectively. It was found a very strong correlation between total polyphenol compounds, flavonoids, phenolic acids, and molybdenum reducing power r = 0.715–0.905. A weak or moderate correlation was found between DPPH scavenging activity and investigated phenolic compounds (r = 0.110–0.522). Obtained data can be useful for further deep biochemical investigation of plant raw of *Silphium* spp. and in livestock nutrition branch.

Keywords: *Silphium* spp., polyphenols, flavonoids, phenolic acids, correlation

Introduction

Genus *Silphium* L., commonly known as rosin-weed, belongs to Asteraceae Bercht. & J. Presl family includes 33 species and originated from Northern America (Clevinger and Panero, 2000; Peni et al., 2020). Representatives from this plant family are well-known as medicinal plants with different biological activities such as antioxidant (Shymanska et al., 2020), antifungal, and immunosuppressive (Wolski and Kędzia, 2018).

S. perfoliatum L. is a high-productive crop (Šiaudinis et al., 2012; Ţîţei et al., 2013; Peni et al., 2020) that can grow for over 15 years (Bury et al., 2020) and be harvested for different purposes such as fodder

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(Pichard, 2012; Shalyuta and Kostitskaya, 2018), honey, ornamental (Jucsor and Sumalan, 2018), medicinal (Kowalska et al., 2022), energetic (Mueller et al., 2019; Šiaudinis et al., 2019; Cossel et al., 2020; Cumpido-Marin et al., 2020; Rakhmetov et al., 2020) plants. The high productivity and calorific value of *S. perfoliatum* allow using plant raw on biogas (Gansberger et al., 2015) and solid (Jasinskas et al., 2014) production. According to Rakhmetov et al. (2020), the yield of energy of plant raw of various *Silphium* genotypes in the conditions of Forest-Steppe of Ukraine was from 43.81 (*S. asperrimum* Hook) to 149.27 (*S. integrifolium* Michx.) Gcal.ha^{-1} at the budding-flowering stage. It is reported about phytoremediation uses *S. perfoliatum* (Peni et al., 2020).

The chemical composition of raw *Silphium* plants is protein, amino acids, fat, cellulose, water-soluble sugars, and minerals (especially potassium, calcium, magnesium, iron, and manganese) (Kowalska et al., 2020). The biochemical compound content of *S. trifoliatum* L. decreased with plant development and the stage of growth before the flowering was the best for harvesting (Kowalski, 2007). Seeds of *Silphium* spp. contained 33.5% of protein, 24.1% of fat, 9.58% of water-soluble sugars, and 25.4% of cellulose. Among the amino acids of seeds, glutamic acid (up to 23%) and leucine (7.76%) were predominant. Main seed fatty acids are determined linolic (44.4%) and oleic (13.2%) (Kowalski & Wiersiński, 2004). In the petroleum ether extracts of leaves and inflorescences identified α-amyrine, heptacosane, stigmasterol, γ-sitosterol, β-marine, etc. The labdane type diterpene and sesquiterpene dominated in rhizome extracts (Kowalski, 2005). According to Ţîţei (2014), the raw protein content of *S. perfoliatum* was 16.33%, and in amino acid composition prevailed glutamine, leucine, and asparagine. Investigation of the above-ground part of *Silphium* spp. raw showed a content of dry matter 21.14–29.02%, water-soluble sugars 3.54–12.17%, crude fiber 29.46–48.24%, ascorbic acid 77.12–296.35 mg%, β-carotene 0.23–1.54 mg% (Rakhmetov et al., 2019).

Leaf, inflorescence, and rhizome extracts of *S. perfoliatum* exhibited activity against some gram-positive and gram-negative bacteria, and rhizome alcohol extracts had the highest antibacterial effect (Kowalski and Kędzia, 2007). Investigations showed that ethanol extract of *S. perfoliatum* inhibited the growth of fungi species *Alternaria alternata* and *Colletotrichum coccodes* that can be used as biological preparation for the management of plant diseases (Jamiołkowska and Kowalski, 2012).

The extracts of these plants and their components such as polysaccharides (Shang et al., 2017) exhibited antioxidant activity (Borchardt et al., 2009; Kowalska et al., 2022). Kowalski and Wolski (2003a, 2003b) identified in phenolic acid fractions caffeic, *p*-coumaric, *p*-hydroxybenzoic, ferulic, and vanillic acids and the most predominant was caffeic acid. The study of the phenolic acid complex of *S. trifoliatum* extracts showed the presence of protocatechuic and salicylic acids besides the above. The phenolic acid complex is found both in free and bounded forms in the leaves, inflorescences, and rhizomes of *S. trifoliatum* (Kowalski, 2007). The flavonoid fraction of *S. trifoliatum* leaf and inflorescences extracts were flavonoid glycosides and kaempferol (Kowalski, 2007), the total content of flavonoids was 0.87 and 1.05% for *S. perfoliatum* and *S. integrifolium* (Kowalski and Wolski, 2003a; Kowalski, 2004). Also, among polyphenol compounds determined 7.34–11.24% of tannins (Kowalska et al., 2020).

This study dwells on the investigation of the polyphenol content and antioxidant activity of ethanol extracts of different organs of *Silphium* spp. as a source of antioxidants.

Biological material

In this study investigated plants of *Silphium* L. (Figure 1) growing in the M.M. Gryshko National Botanical Garden of NAS of Ukraine in Kyiv (NBG). Some species were investigated in an experimental study in 2019–2020, including *S. asperrimum* Hook., *S. laciniatum* L., *S. perfoliatum* L., and *S. trifoliatum* L.

Figure 1 *Silphium* spp. at the flowering stage
1 – *S. asperrimum* Hook.; 2 – *S. laciniatum* L.; 3 – *S. perfoliatum* L.; 4 – *S. trifoliatum* L.

All biochemical analyses were conducted at the Slovak University of Agriculture in Nitra (Slovak Republic).

Chemicals

All chemicals used were of analytical grade and were purchased from Sigma-Aldrich (St. Louis, MO, USA) and CentralChem (Slovakia).

Preparations of extracts

An amount of 0.25 g of each sample was extracted with 20 mL of 80% ethanol for 2 h in a laboratory shaker GFL 3005 (GFL, Burgwedel, Germany). Then, the samples were centrifuged at 4605 RCF (Rotofix 32 A, Hettich, Germany) for 10 min and the supernatant was used for measurement of FRSA (antiradical activity) using DPPH, MRAP (antioxidant activity) using phosphomolybdenum method and measurement of other antioxidant properties (detection of total polyphenol, total flavonoid, and phenolic acid content).

Total polyphenol content of extracts

The total polyphenol content (TPC) was measured by the method of Singleton and Rossi (1965) using the Folin-Ciocalteu reagent. A quantity of 0.1 mL of each sample was mixed with 0.1 mL of the Folin-Ciocalteu reagent, 1 mL of 20% (w/v) sodium carbonate, and 8.8 mL of distilled water. After 30 min in darkness, the absorbance at 700 nm was measured with the spectrophotometer Jenway (6405 UV/Vis, England). Gallic acid (25–300 $mg.L^{-1}$; R^2 = 0.998) was used as the standard. The results were expressed as $mg.g^{-1}$ DW gallic acid equivalent.

Total phenolic acid content

The content of phenolic acids (TPAC) 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 flavonoid content of extracts

The total flavonoid content (TFC) was determined by the modified method described by Shafii et al. (2017). An aliquot of 0.5 mL of the sample was mixed with 0.1 mL of 10% (w/v) ethanolic solution of aluminium chloride, 0.1 mL of 1 M potassium acetate, and 4.3 mL of distilled water. After 30 min in darkness, the absorbance at 415 nm was measured using the spectrophotometer Jenway (6405 UV/Vis, England). Quercetin (1–400 $mg.L^{-1}$; R^2 = 0.9977) was used as the standard. The results were expressed in $mg.g^{-1}$ DW quercetin equivalent.

Free radical scavenging activity

Free radical scavenging activity (FRSA) of samples (antiradical activity) was measured using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) (Sánchéz-Moreno et al., 1998). An amount of 0.4 mL of sample was mixed with 3.6 mL of DPPH solution (0.025 g DPPH in 100 mL ethanol). The absorbance of the reaction mixture was determined with 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 = 0.989) was used as the standard and the results were expressed in $mg.g^{-1}$ DM Trolox equivalents.

Molybdenum-reducing power of extracts

The molybdenum-reducing power (MRP) of samples was determined by the method of Prieto et al. (1999) with slight modifications. The mixture of the sample (1 mL), monopotassium phosphate (2.8 mL, 0.1 M), sulfuric acid (6 mL, 1 M), ammonium heptamolybdate (0.4 mL, 0.1 M), and distilled water (0.8 mL) was incubated at 90 °C for 120 min, then cooled to room temperature. The absorbance at 700 nm was detected with 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 $mg.g^{-1}$ DM Trolox equivalent.

Statistical analysis

The results are expressed as mean values of three replications ± standard deviation (SD); Data were analyzed with the ANOVA test and differences between means were compared through the Tukey-Kramer test ($p < 0.05$).

Results and discussion

The phytochemical investigation of plant raw materials demonstrated the therapeutical properties of various plant products. Some plants from Asteraceae, as reported in previous reviews, have medicinal applications (Bessada et al., 2015). Last time, numerous studies showed that plants from this family are a rich source of biologically active compounds that caused the antioxidant activity of plant raw (Vijaylakshmi et

al., 2009; Bakar et al., 2015; Kumar et al., 2019a), and their preparations (Rolnik et al., 2021). The antioxidant activity of plant extracts has been correlated with polyphenol compounds of different natures (Piluzza and Bullitta, 2009; Spiridon et al., 2011; Terpinc et al., 2012). It has been determined diversity of polyphenol compounds of different Asteraceae species has pharmacological importance and systematic value (Williams et al., 2009; Pavlenko-Badnaoui et al., 2021).

Polyphenols are a widely distributed group of biologically active compounds in plant raw, products and plant waste that includes flavonoids and phenolic acids (Abbas et al., 2017; Mourtzinos and Goula, 2019). Some authors divided polyphenols into flavonoids and non-flavonoids (Stagos, 2020). These compounds possess various functions among which are antioxidant and health-promoting (Maqsood et al., 2014). Nutritionists' attention to plant polyphenols is explained by their health effects of it such as anticancer (Dai and Mumper, 2010). Phenolic compounds can be used as chemical markers in botanical chemosystematics studies on different taxonomic levels (Míka et al., 2005). It is found a negative correlation between polyphenol consumption with cardiovascular diseases, cancer, and diabetes (Abbas et al., 2017).

The polyphenol, flavonoids, and phenolic acids content of four species of *Silphium* changed during vegetation. The accumulation of total polyphenol content in our study was from 12.32 (*S. asperrimum*, stems at the budding) to 95.21 (*S. laciniatum*, leaves at the flowering stage) mg GAE.g^{-1} DW depending on the organ and stage of vegetation (Figure 2). Leaves of investigated *Silphium* spp. accumulated polyphenols from 38.58 to 83.32 mg GAE.g^{-1} DW at the budding stage and from 69.13 to 95.21 mg GAE.g^{-1} DW at the flowering stage. TPC in the buds was from 34.87 to 46.58 mg GAE.g^{-1} DW at the budding and in flowers from 42.11 to 52.67 mg GAE.g^{-1}. This parameter in stems at the budding and flowering stage was 12.32–38.36 and 18.23–41.22 mg GAE.g^{-1}, respectively. As shown in Figure 2, at the budding and flowering stages the total content of polyphenol compounds was the least accumulated in the stems of all investigated plants except plants of *S. perfoliatum* buds at the stage of budding. At both budding and flowering stages, the most polyphenols accumulated in the leaves.

Due to limited information about *Silphium* spp. polyphenol content comparison with other results is difficult. Existing data about such a large group as Asteraceae, on the whole, showed that polyphenol content significantly varied. For example, *Aster scaber* leaf extracts were investigated in different solvents, and the maximal value of polyphenol content found in ethyl acetate (322.43 mg GAE.g^{-1}), minimal in water (23.67 mg GAE.g^{-1}) (Thiruvengadam et al., 2014). The polyphenol content of *Chrysanthemum parthenium* was 3.48 mg GAE 100 g^{-1} and the flavonoid content was 1.27 mg RE 100 g^{-1} (Hanganu et al., 2016). As reported Azzouzi et al. (2016), this parameter in *Centaurea choulettiana* Pomel extracts detected 325.81 mg GAE.g^{-1}. As resulted by Indradi et al. (2017), the TPC of four species from Asteraceae was from 3.19 to 16.48 mg GAE 100 g^{-1} depending on extracts. The TPC in ethanol extracts of *Cichorium itybus* L. was 33.91 mg GAE.g^{-1} (Vergun et al., 2019). A study of other Asteraceae representatives showed that the TPC of *Helianthus tuberosus* was 7.9–11.1 mg GAE.g^{-1} in leaf

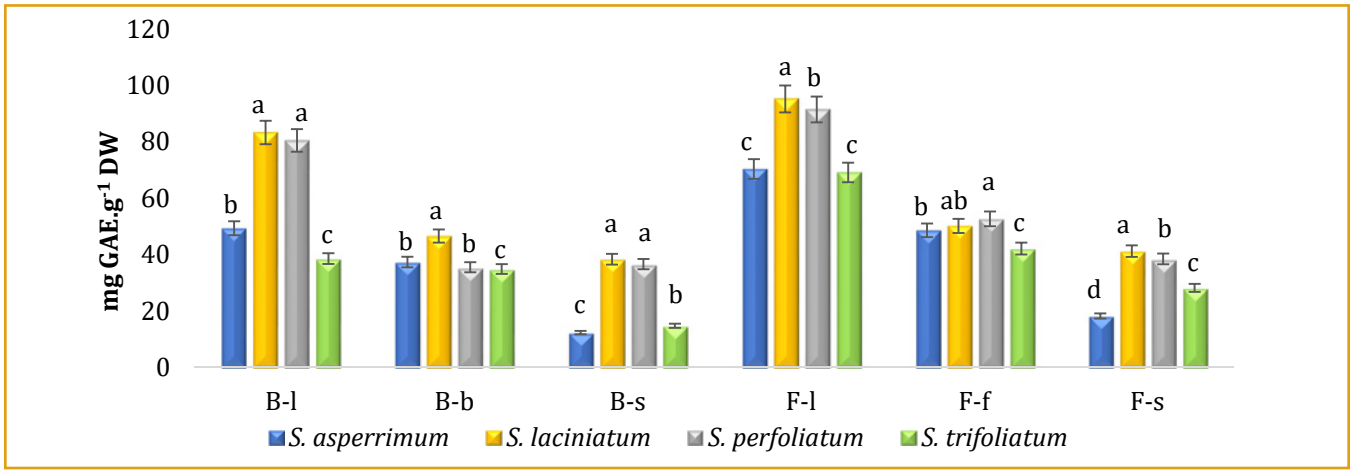

Figure 2 The total content of polyphenol compounds of ethanol extracts of *Silphium* spp.
GAE – gallic acid equivalent, B-l – leaves at the budding; B-b – buds at the budding; B-s – stems at the budding; F-l – leaves at the flowering; F-f – flowers at the flowering; F-s – stems at the flowering. Means in each column followed by different letters are significantly different ($p < 0.05$)

extracts, 4.0–5.3 mg GAE.g^{-1} in flower extracts, and 0.9–1.7 mg GAE.g^{-1} in stem extracts (Showkat et al., 2019). In *Matricaria recutita* L. extracts determined 21.4 mg GAE.g^{-1} of polyphenol compounds (Al-Dabbagh et al., 2019). In extracts of *Helianthus annuus* L. determined the TPC during stem extension was 21.9 mg GAE.g^{-1}, visible bud period 17.7 mg GAE.g^{-1}, at the early, mid-, and late flowering 20.4, 29.3, and 21.7 mg GAE.g^{-1}, respectively (Gai et al., 2020). *Matricaria chamomilla* L. extracts showed a TPC of 3.72–7.94 mg GAE.g^{-1} (Hassanpour et al., 2020). As reported by Muhtadi (2021), the TPC of *Helianthus annuus* leaves, inflorescences, and bark extracts was 35.14, 24.88, and 17.46 mg GAE.g^{-1}, respectively. Thus, the total content of polyphenols and antioxidant activity depended on the solvent, for example, as shown in a study with *Vernonia blumeoides* (Asteraceae) where in n-butanol fraction determined the total polyphenol content was approximately 4 times more than in ethanol crude extracts (Aliyu et al., 2011).

One of the most important classes of polyphenols is phenolic acids and their derivates (Mourtzinos and Goula, 2019). They play an important role in the human diet and demonstrated antimicrobial and strong antioxidant activity (Heleno et al., 2015). Among phenolic acids of *Silphium* spp. leaves identified complexes of benzoic and cinnamic acids (Williams et al., 2009). Among benzoic acids identified *p*-hydroxybenzoic, protocatechuic, isovanilic, gallic, ellagic, vanillic, syringic, and salicylic; among cinnamic acids found chlorogenic, rosmarinic, *p*-coumaric, caffeic, hydrocaffeic, ferulic, isoferulic, *m*-coumaric (Kowalska et al., 2022).

The total content of phenolic acids in ethanol extracts of investigated plants was from 3.52 (*S. asperrimum*, buds at the budding) to 34.58 (*S. laciniatum*, leaves at the flowering) mg CAE.g^{-1} DW depending on the species and part of the plant (Figure 3). At the budding stage, TPAC in the leaves was from 6.8 to 23.89 mg CAE.g^{-1} DW, and at the flowering from 13.36 to 34.58 mg CAE.g^{-1} DW. Buds accumulated TPAC from 3.52 to 13.68 mg CAE.g^{-1} DW and flowers from 9.08 to 21.14 mg CAE.g^{-1} DW. In stem extracts, TPAC was 3.72–8.57 mg CAE.g^{-1} DW at the budding and 5.86–14.12 mg CAE.g^{-1} DW at the flowering. Stem extracts showed TFC from 3.72 to 8.57 mg QE.g^{-1} at the budding stage and from 5.86 to 14.12 mg QE.g^{-1} DW at the flowering. Most contents of flavonoids for all investigated species accumulated in the leaves, the least in the stems. However, exclusion was found for *S. asperrimum*, stem ethanol extracts of which demonstrated higher TPAC than bud extracts.

According to Kowalski and Wolski (2003b), the content of phenolic acids in dry extracts of *S. perfoliatum* leaves and inflorescences were 23.2 and 25.5 mg 100 g^{-1}, respectively. The leaf fraction of *S. trifoliatum* contained up to 21.15 mg 100 g^{-1}, inflorescences up to 52.73 mg 100 g^{-1}, and rhizomes up to 10.33 mg 100 g^{-1} of phenolic acids, among which caffeic acid was predominant (Kowalski, 2007). It was determined of 4.56 mg CAE.g^{-1} of TPAC in the ethanol extracts of *Cichorium intybus* (Vergun et al., 2019).

Flavonoids are a group of secondary metabolites with numerous functions such as survival and reproductive fitness (Williams et al., 2009). As many authors emphasize, flavonoid action is not clear full, however, they may regulate the molecular and cellular processes (Spencer et al., 2009; Kumar et al., 2019b). Asteraceae representatives are characterized by the presence of flavonoid aglycons such as kaempferol, apigenin, gekwanin, acacetin, luteolin, nepetin, eupaletin, etc.

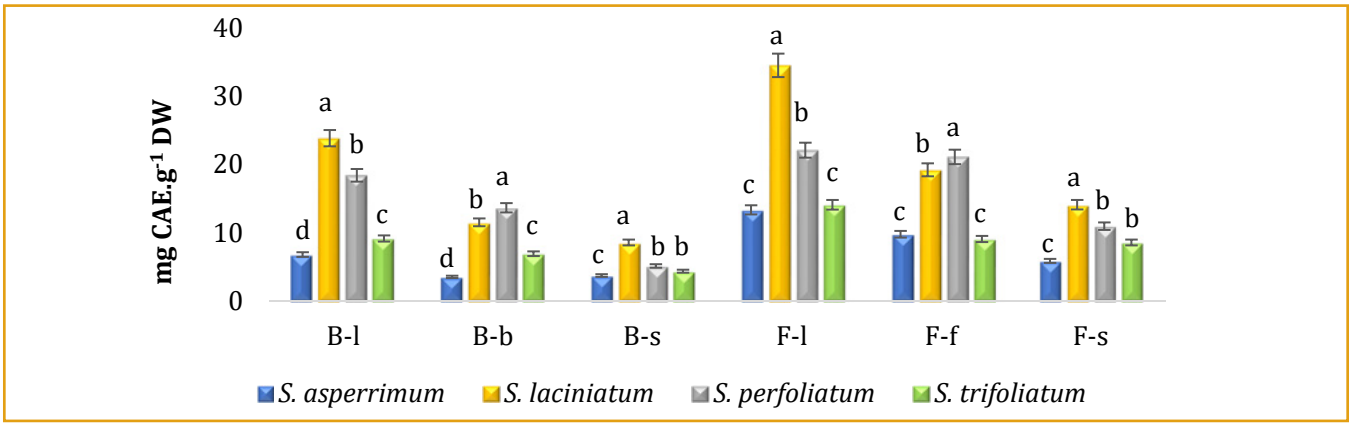

Figure 3 The total content of phenolic acids in ethanol extracts of *Silphium* spp.
CAE – caffeic acid equivalent, B-l – leaves at the budding; B-b – buds at the budding; B-s – stems at the budding; F-l – leaves at the flowering; F-f – flowers at the flowering; F-s – stems at the flowering. Means in each column followed by different letters are significantly different ($p < 0.05$)

Considering the results of the genus *Silphium* should be noted that *S. terebinthinaceum* does not contain aglycones that are quite incoming among Asteraceae (Valant-Vetschera and Wollenweber, 2007). The flavonoid composition of *Silphium* spp. are derivates of the flavonols quercetin, isorhamnetin, and kaempferol (Williams et al., 2009).

TFC in investigated extracts was from 3.67 (*S. perfoliatum*, stems at the budding) to 57.31 (*S. laciniatum*, leaves at the budding) mg QE.g^{-1} DW depending on species and period of growth (Figure 4). Leaf extracts at the budding and flowering stage had TFC 29.31–57.31 and 36.15–51.78 mg QE.g^{-1} DW, respectively. In the buds and flowers, this parameter was 11.12–25.45 and 24.29–32.2 mg QE.g^{-1} DW, respectively. TFC in the stems at the budding and flowering stage was 3.67–22.53 and 11.07–20.96 mg QE.g^{-1} DW, respectively. TFC in the leaf extracts depends on the period of growth and species was maximal and in stem extracts was minimal.

According to Azzouzi et al. (2016), the TFC of *Centaurea choulettiana* Pomel extracts was 236.73 mg QE.g^{-1}. Extracts of *Matricaria recutita* L. showed a TFC of 157.9 mg QE.g^{-1}. As reported by Indradi et al. (2017), different representatives of Asteraceae had the TFC from 0.83 to 23.49 mg QE 100 g^{-1} depending on species and extracts. The TFC in the extracts of *Cichorium intybus* was 26.29 mg QE.g^{-1} (Vergun et al., 2019). According to Hassanpour et al. (2020), in extracts of *M. chamomilla* L. the TFC was 1.37–2.98 mg RE.g^{-1} (rutin equivalent). Muhtadi (2021) determined 10.91, 4.58, and 2.59 mg QE.g^{-1} of TFC in the leaf, inflorescence, and bark extracts of *Helianthus annuus*, respectively.

This study demonstrated that leaves of all investigated *Silphium* species accumulated the highest content of total polyphenols, flavonoids, and phenolic acids. The comparable analysis of reviews about different Asteraceae representatives and obtained data showed that the content of polyphenol compounds, flavonoids, and phenolic acids depends on many factors such as species, extracts, conditions of growth, methods of detection, etc. Due to the lack of sufficient data on a concrete plant species, it is difficult to compare obtained results, however, this gives a general idea about an accumulation of certain groups of polyphenol compounds and their antioxidant activity.

Exist numerous antioxidant activity methods (Alam et al., 2013; Pisoschi et al., 2016; Romulo, 2020) that can indicate that polyphenol compounds are present (Mourtzinos and Goula, 2019). According to Alam et al. (2013), ethanol extracts are most commonly used for antioxidant capacity studies. Antioxidant activity depends on species, populations (Hassanpour et al., 2020), and extracts (Chatha et al., 2006; Wong et al., 2006; Borah et al., 2012). Among antioxidant activity methods, the most widely used is the DPPH (α,α-diphenyl-β-picrylhydrazyl) method based on electron donation and the reaction of discoloration of DPPH radical (Brand-Williams et al., 1995; Alam et al., 2013; Shahidi and Zhong, 2015).

Antioxidant activity by the DPPH method of extracts was from 3.33 (*S. trifoliatum*, stems at the budding)

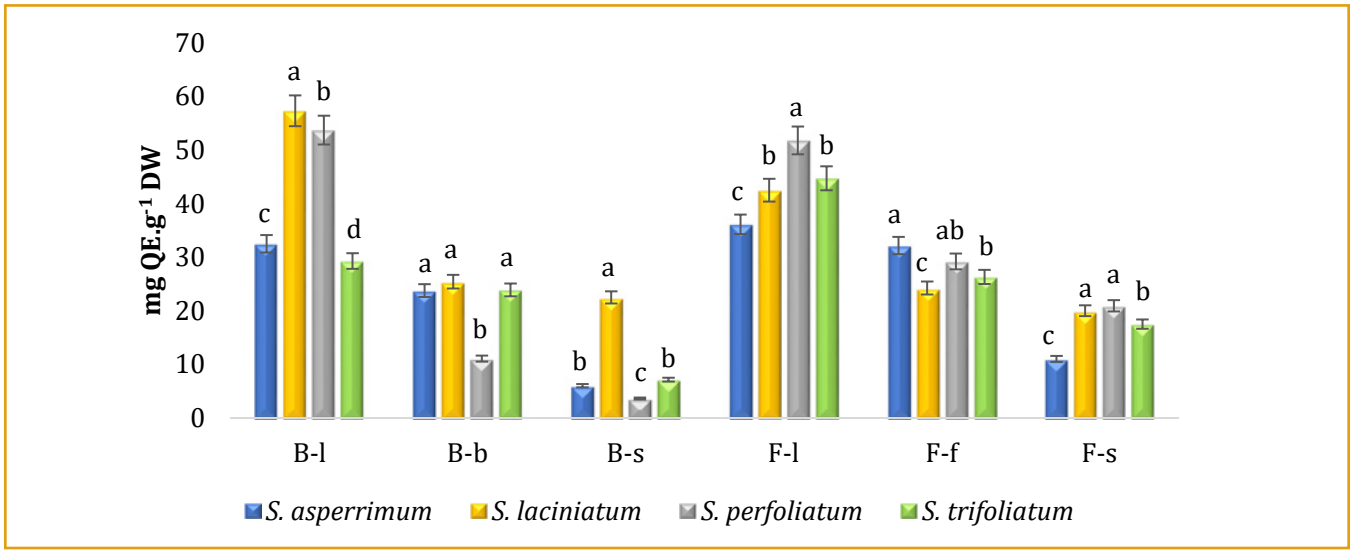

Figure 4 Total content of flavonoids of ethanol extracts of *Silphium* spp.
QE – quercetin equivalent, B-l – leaves at the budding; B-b – buds at the budding; B-s – stems at the budding; F-l – leaves at the flowering; F-f – flowers at the flowering; F-s – stems at the flowering. Means in each column followed by different letters are significantly different ($p < 0.05$)

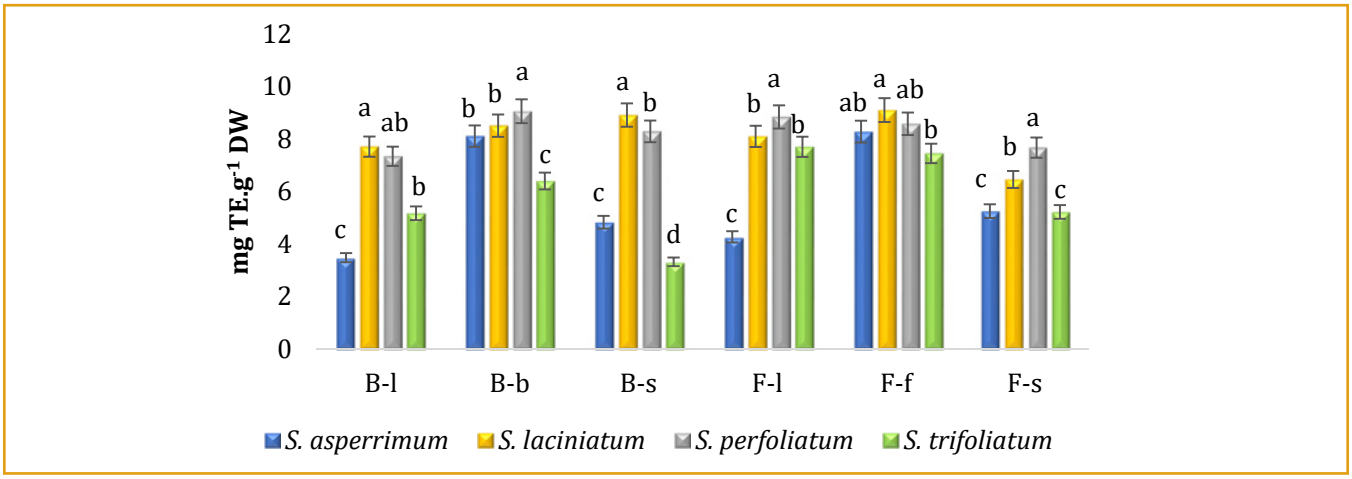

Figure 5 DPPH-radical scavenging activity of ethanol extracts of *Silphium* spp. by DPPH method
TE – Trolox equivalent, B-l – leaves at the budding; B-b – buds at the budding; B-s – stems at the budding; F-l – leaves at the flowering; F-f – flowers at the flowering; F-s – stems at the flowering. Means in each column followed by different letters are significantly different ($p < 0.05$)

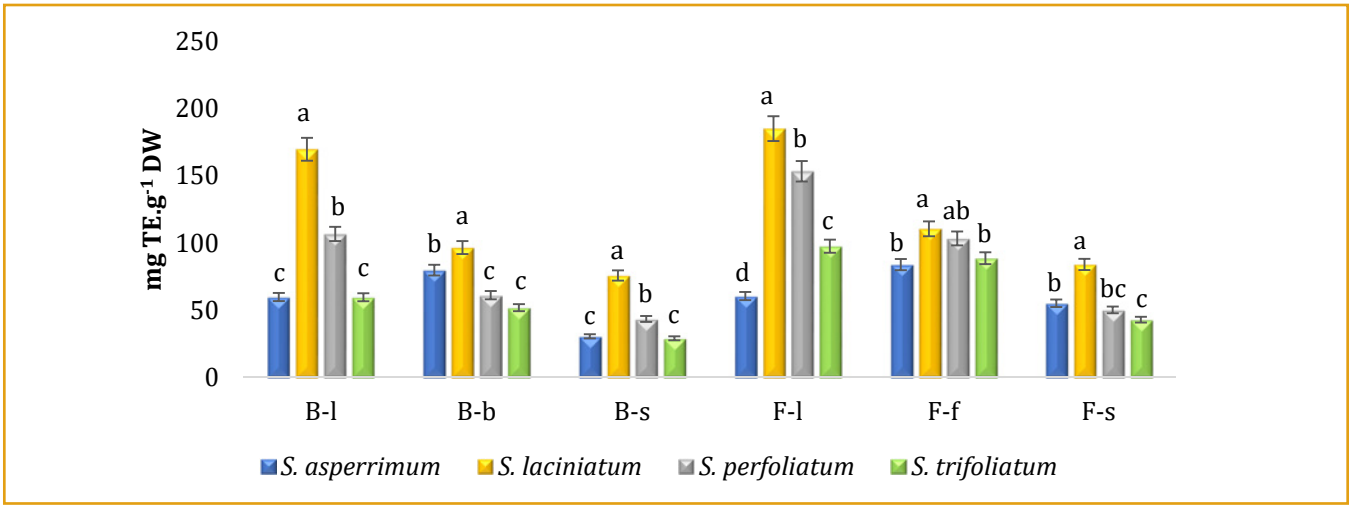

Figure 6 Molybdenum reducing power of ethanol extracts of *Silphium* spp.
TE – Trolox equivalent, B-l – leaves at the budding; B-b – buds at the budding; B-s – stems at the budding; F-l – leaves at the flowering; F-f – flowers at the flowering; F-s – stems at the flowering. Means in each column followed by different letters are significantly different ($p < 0.05$)

to 9.11 (*S. laciniatum*, flowers at flowering) mg TE.g^{-1} DW depending on stage and organ (Figure 5). RP of leaf extracts at the budding and flowering was 3.49–7.72 and 4.28–8.85 mg TE.g^{-1} DW, respectively. Bud and flower extracts showed the RP of 6.41–9.07 and 7.46–9.11 mg TE.g^{-1} DW, respectively. Stem extracts exhibited RP from 3.33 to 8.92 mg TE.g^{-1} DW at the budding and from 5.23 to 7.68 mg TE.g^{-1} DW at the flowering.

Extracts of *Elephantopus scaber* L., *Eclipta alba* (L.) Hassk., *Pluchea indica* (L.) Less, and *Taraxacum officinale* Weber ex F. H. Wigg had DPPH scavenging activity from 16.66 to 16.48 µg.ml^{-1} (Indradi et al., 2017). DPPH-radical scavenging activity of ethanol extracts of *Cichorium intybus* was 8.35 mg TE.g^{-1} (Vergun et al., 2019). Antiradical activity of different extracts of *Silphium* spp. showed that methanol extracts of four investigated species stem had less values, the maximum values demonstrated water extracts of leaves, buds, or inflorescences (Shymanska et al., 2020).

Antioxidant capacity by phosphomolybdenum method based on the reduction of Mo (VI) to Mo (V) by sample extract and described by Prieto et al. (1999). Use of different temperatures (from 40 to 100 °C) at the leaf's extraction of *Gynura divaricate* the highest antioxidant activity by phosphomolybdenum method was at 90 and 100 °C (Wan et al., 2011). In total, the antioxidant activity by phosphomolybdenum method of extracts was from 29.11 (*S. trifoliatum*, stems at the budding) to 185.22 (*S. laciniatum*, leaves at the flowering) mg TE.g^{-1} DW depending on the species and

stage of vegetation (Figure 6). MRP of leaf extracts at the budding and flowering stage was 59.65–169.85 and 60.53–185.22 mg TE.g^{-1} DW, respectively. Extracts of *S. asperrimum* showed the highest MRP in the generative organs, whereas, all other species had the highest values in the leaves at both periods. In this case, the lowest MRP was in the stem extracts of all investigated plants.

Plants of *Scorzonera hispanica* L. at the start of spring vegetation had antioxidant activity by phosphomolybdenum method 125.46 mg TE.g^{-1} (Vergun et al., 2018). Extracts of *Cichorium intybus* at the flowering stage showed 93.01 mg TE.g^{-1} of MRP (Vergun et al., 2019). According to Alper et al. (2021), ethanol extracts of *Centaurea solstitiales* and *Urospermum picroides*, other species of Asteraceae, showed antioxidant activity by this method of 49.23 and 42.13 mg TE.g^{-1}, respectively. In this study, different parts of *Silphium* spp. at the flowering stage showed activity by this method from 42.97 to 185.22 mg TE.g^{-1}. This study showed that extracts of *S. laciniatum* exhibited the highest molybdenum-reducing power depending on the plant part and stage of growth.

Numerous studies demonstrated a positive correlation between total polyphenol content and antioxidant activity. However, this depends on the assay of determination and plant species. A strong correlation between antioxidant activity by the phosphomolybdenum method of plant extracts and total phenolic content and total flavonoid content was found in some studies (Khan et al., 2012). A very strong correlation between investigated parameters at the budding stage was found between TPC and TFC (r = 0.924), TPC and MRP (r = 0.890), TPAC and MRP (r = 0.869), TPC and TPAC (r = 0.864), TFC and MRP (r = 0.858), TPAC and TFC (r = 0.793) (Table 1).

At the flowering stage, a very strong correlation was found between TPC and TFC (r = 0.941), TPAC and MRP (r = 0.905), TPC and MRP (r = 0.825), TPC and TPAC (r = 0.810), TFC and MRP (r = 0.715) (Table 2).

The study of different Asteraceae species showed the existence of a correlation between parameters of antioxidant activity. Aktumsek et al. (2013) found for *Centaurea* L. taxa a strong correlation between antioxidant activity and total phenolic content and flavonoid content (r = 0.84–0.96). The reducing power of extracts in that study also showed a strong correlation with total phenolic content. Muhtadi (2021) found a very strong correlation between DPPH antioxidant activity and TPC (r = 1.000) and with TFC (r = 0.962).

Opposite, the study of *Helichrysum* spp. didn't show a correlation between total phenolic content and antioxidant activity by the phoshomolybdenum method and DPPH method (Albayrak et al., 2010). Negative correlations between DPPH scavenging activity and TPC (r = -0.180–0.952) were found in extracts of Asteraceae species (Indradi et al., 2017). The study of *Cichorium intybus* extracts demonstrated

Table 1 Correlation between antioxidant parameters of *Silphium* spp. at the budding stage

Parameter	TPC	TPAC	TFC	DPPH	MRP
TPC	1.000				
TPAC	0.864**	1.000			
TFC	0.924**	0.793**	1.000		
DPPH	0.342*	0.378*	0.110*	1.000	
MRP	0.890**	0.869**	0.858**	0.451*	1.000

TPC – total phenolic content; TPAC – total phenolic acid content; TFC – total flavonoid content; DPPH – DPPH-radical scavenging activity; MRP – molybdenum reducing power of extracts; ** – correlation is significant at the level of 0.01; * – correlation is significant at the level of 0.05

Table 2 Correlation between antioxidant parameters of *Silphium* spp. at the flowering stage

Parameter	TPC	TPAC	TFC	DPPH	MRP
TPC	1.000				
TPAC	0.810**	1.000			
TFC	0.941**	0.617*	1.000		
DPPH	0.397*	0.522*	0.428*	1.000	
MRP	0.825**	0.905**	0.715**	0.648*	1.000

TPC – total phenolic content; TPAC – total phenolic acid content; TFC – total flavonoid content; DPPH – DPPH-radical scavenging activity; MRP – molybdenum reducing power of extracts; ** – correlation is significant at the level of 0.01; * – correlation is significant at the level of 0.05

a strong positive correlation between antioxidant activities and polyphenol compound groups: between MRP and TFC (r = 0.997), between DPPH and TPC (r = 0.996), between DPPH and TFC (r = 0.976), between DPPH and TPAC (r = 0.971), between MRP and TPC (r = 0.971), between MRP and TPAC (r = 0.884) (Vergun et al., 2019).

According to this study's results, between DPPH scavenging activity of extracts and different polyphenol compounds existed weak or moderate relation depending on the stage of growth (r = 0.110–0.378 at the budding and r = 0.397–0.522 at the flowering) whereas between molybdenum reducing power and polyphenol compounds found a strong correlation (r = 0.859–0.890 at the budding and r = 0.715–0.905 at the flowering). Between the two methods of antioxidant activity, determination found a moderate correlation. It should be concluded from these data that total phenolic compounds contribute highly to the antioxidant activity by the phosphomolybdenum method and contributes slightly to the antioxidant activity by the DPPH method of investigated species of *Silphium*.

Conclusions

Obtained data on antioxidant parameters of *Silphium* spp. showed that its promising crops with antioxidant activity. The content of polyphenol compounds and phenolic acids in ethanol extracts of all investigated plants was the highest in the leaves and the maximal value had plants *S. laciniatum* (95.21 and 34.58 mg CAE.g^{-1}, respectively). The accumulation of flavonoids was uneven. In this case, plants *S. asperrimum* and *S. trifoliatum* accumulated maximal total flavonoid content in the leaves at the flowering stage, and in its turn, *S. laciniatum* and *S. perfoliatum* in the leaves at the budding stage. The highest content of flavonoids was determined in raw *S. laciniatum* (57.31 mg QE.g^{-1}). The highest antioxidant activity by the DPPH method was found at the budding stage for *S. perfoliatum* (buds) and flowering stage for *S. asperrimum* (stems), *S. laciniatum* (flowers), *S. trifoliatum* (leaves). Ethanol extracts of all investigated plants demonstrated maximal values of molybdenum-reducing power at the flowering stage but *S. asperrimum* in the flowers and *S. laciniatum*, *S. perfoliatum*, and *S. trifoliatum* in the leaves. This research can be useful for further deep biochemical investigation of plant raw of *Silphium* spp. and in livestock nutrition branch.

Conflicts of interest

The authors declare no conflict of interest.

Ethical statement

This article doesn't contain any studies that would require an ethical statement.

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