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# Phenolic Compounds of Plants *Bidens tripartita* (L.) and *Bidens pilosa* (L.) from Different Locations

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Abstract—*Bidens tripartita* L. and *Bidens pilosa* L. are potential sources of biologically active substances with antimicrobial, antidiabetic, anticancer, anti-inflammatory, antioxidant, and other activities. These types of strings are widely used in different countries in phytomedicine. It was established that the studied species are rich in a variety of phenolic compounds, and plants growing in temperate continental (Tatarstan) and tropical (Burundi) climates differ slightly in the content of phenolic compounds, which indicates a genetically determined narrow amplitude of variability in the metabolism of these species. Qualitative analysis of phenolic compounds showed that the studied plant species synthesize certain groups of compounds for adaptation to specific environmental conditions. Kirimiro in the Republic of Burundi and Spassky raion in the Republic of Tatarstan can be considered as promising areas for growing and collecting the plant species under study. Temperature, altitude, rainfall, and soil composition are key factors affecting phenolic content in *B. pilosa* and *B. tripartita* plants.

**Keywords:** *Bidens pilosa* (L.), *Bidens tripartita* (L.), soil composition, temperature, phenolic compounds **DOI:** 10.1134/S1021443723700243

## **INTRODUCTION**

*Bidens* is one of the genera of the aster family (Asteraceae), which includes approximately 280 species, including, *B. tripartita* and *B. pilosa*. Species of *Bidens* are widely used in folk and traditional medicine to treat various diseases in different countries of the world.

According to the literature, plants of the genus *Bidens* contain a variety of compounds, including flavonoids, phenylpropanoids, triterpenoids, alkaloids, and organic acids, of which flavonoids are the main effective components [1].

Modern pharmacological studies show that it is these compounds in plants *Bidens* that have antiinflammatory, antioxidant, analgesic, antibacterial, antitumor, hypolipidemic, antidiabetic, and hepoprotective functions [2].

In turn, the main function of secondary metabolites in plants is protection from the adverse effects of various factors of abiotic and biotic nature throughout life, thus ensuring the fitness and survival of plants [3, 4]. To cope with stress, plants are forced to induce or reduce the synthesis of secondary metabolites in the body [5]. Various studies indicate a significant influence of individual environmental factors on the accumulation of substances of secondary metabolism in plants. For example, drought has been shown to cause oxidative stress with an increase in phenolic compounds and vitamins in *Amaranthus tricolor* L. [6]. Elevated temperatures and high light intensity increase the synthesis and accumulation of various secondary metabolites, including quercetin, tannins, ascorbic acid, and triterpenoids, many of which have antioxidant properties [7].

In this regard, the composition and quantity of biologically active substances that characterize the effectiveness of medicinal plants differ depending on the type and organ of the plant, the period of collection, and habitat conditions (climatic conditions, geographical location, type and composition of soil, etc.) [8, 9].

The goal of our work was to identify a set of environmental factors and promising regions for growing and collecting *B. pilosa* and *B. tripartite* medicinal plants with the highest content of biologically active substances. To achieve this goal, a comparative study of the content of phenolic compounds of two plant species (*B. pilosa* and *B. tripartita*) growing in different regions of the Republic of Burundi and the Republic of Tatarstan was carried out and the degree of influence of ecological and geographical factors on the content of biologically active substances in the studied plants was assessed.

*Abbreviations*: WPC—water-soluble phenolic compounds; RB—Republic of Burundi; RT—Republic of Tatarstan.

Area	Altitude above sea level, m	Precipitation amount, mm	tempera	pН	Content mineral elements in soils of the studied regions					
					$NO_3^-,\%$	$PO_4^{3-}, md$	K <sup>+</sup> , mEq/100 g	Ca <sup>2+</sup> , mEq/100 g	Mg <sup>2+</sup> , mEq/100 g	
Kirimiro	1603	1321	19	4.9	0.13	5.94	0.36	4.7	0.63	
Mugamba	2097	1598	15.7	4.0	0.11	7.56	0.24	0.45	0.19	
Buragane	1550	1233	21.5	5.7	0.19	8.45	0.57	3.44	0.58	

Table 1. Geo-ecological characteristics of the regions of Kirimiro, Mugamba, and Buragane (annual average for 2019)

**Table 2.** Landscape and climatic (autumn-spring period 2019) characteristics of the regions of Tatarstan. <a href="https://nuipo-goda.ru/">https://nuipo-goda.ru/</a>

Area	Quantities precipitation, mm	Average temperature, °C	Klimatiche Sky zone	pН	Content mineral elements V soils of the studied regions					
					$\mathrm{NO}_3^-,\%$	$PO_4^{3-}, md$	K <sup>+</sup> , mEq/100 g	Ca <sup>2+</sup> , mEq/100 g	Mg <sup>2+</sup> , mEq/100 g	
Spassky	189	12.5	Forest-steppe	7.4	0.23	0.29	1.84	15.03	12.00	
Vysokogorsky	261	14	Coniferous-wide	6.1	0.18	0.31	1.27	15.63	1.56	
			natural							

# MATERIALS AND METHODS

**Object of study.** The object of the study was the aerial part of two Bidens species: B. tripartita and B. pilosa from different regions of the Republic of Burundi (RB) (Table 1): Kirimiro (place of plant collection was dry meadow with high illumination, altitude 1603 m above sea level), Mugamba (place of plant collection was dry meadow with high illumination, altitude 2097 m above sea level), Buragane (place of plant collection was dry meadow with high illumination, height above sea level 1550 m); and the Republic of Tatarstan (RT) (Table 2): Spassky raion (place of plant collection was a sedate meadow with high illumination) and Vysokogorsky raion (place of plant collection was roadside depression with high illumination). Plants were collected in July 2019 at the flowering stage. The composition of soils from the collection site of RB plants was determined in the agrochemical laboratory for soil and food analysis of the Institute of Agronomic Sciences of Burundi (ISABU). The altitude above sea level at the collection site was determined using the offlinemaps application. Temperature and precipitation data were provided by the Geographical Institute of Burundi (IGEBU). The composition of soils from the site where plants were collected in the Republic of Tatarstan was determined in the agrochemical laboratory of the Rosselkhoznadzor of the Republic of Tatarstan.

Plant material was dried in the shade until air-dry; samples were stored in paper bags without exposure to direct sunlight. The leaves of the plants were ground to a powder. Next, the resulting crushed raw materials were used to prepare extracts. Determination of the total content of water-soluble phenolic compounds. The total content of water-soluble phenolic compounds (WPC) was determined by the spectrophotometric method using the reagent Folina-Denis in the presence of NaHCO<sub>3</sub>. Approximately 50 mg of crushed plant material was placed in an Eppendorf tube, 1.5 mL of distilled water was added and incubated in a water bath for 45 min at 70°C, after which it was centrifuged at 15000 rpm for 5 min. The aqueous extract was stored at +4°C without access to light for further use.

To determine WPC, 75  $\mu$ L of the aqueous extract was taken into 1.5 mL test tubes; 75  $\mu$ L of the Folin-Denis reagent was added, mixed, and 120  $\mu$ L saturated NaHCO<sub>3</sub> solution (10%) was added after 3 min, stirred, 1.2 mL of water was added, and centrifuged after 45 min at 16000 rpm for 2 min. Optical density was measured using a spectrophotometer at a wavelength of 725 nm. As a control, 75  $\mu$ L of water was used instead of the extract. The total content of soluble phenolic compounds in the studied raw materials was calculated using the formula:

$$C = \frac{EKRV}{m \times 1000}$$

where *C* is concentration of phenolic compounds, mg/g dry mass; *E* is optical density at 725 nm; *K* is conversion factor to the reference substance—(-)-epicatechin (480); *R* is dilution; *V* is extract volume, mL; and m is the mass of a sample of plant material, g [10].

**Determination of flavonoid content.** The extract for the determination of flavonoids in medicinal raw

Page 3 of 9 160 is the volume of 0.02 M potassium

materials was prepared according to the method described in [11]. Approximately 1 g of crushed plant material was placed in a 150-mL conical flask, 30 mL of 90% ethyl alcohol containing 1% concentrated hydrochloric acid (HCl) was added, and it was heated in a boiling water bath for 30 min. The flask was then cooled to room temperature and the resulting extract was filtered through a paper filter. The extraction was repeated twice. It was filtered through the same filter into the same flask, then the filter was washed with 90% ethyl alcohol and the volume of the filtrate was brought to the mark (solution A).

For determination, 2 mL of solution A was placed in a flask, 1 mL of 1% aluminum chloride solution (AlCl<sub>3</sub>) in 95% ethyl alcohol was added, and the volume of the solution was brought to the mark with 95% ethyl alcohol. Then, after 20 min, the optical density of the solution was determined using a spectrophotometer at a wavelength of 430 nm. As a control, we used a solution consisting of 2 mL of solution A adjusted with 95% ethyl alcohol to a given volume.

The content of the total flavonoids in terms of quercetin and absolutely dry raw materials in percent (X) was calculated using the formula:

$$X = \frac{D \times 25 \times 100 \times 100}{764.6m \times 2(100 - W)},$$

where D is the optical density of the test solution; 764.6 is specific absorption rate of the quercetin complex with aluminum chloride at 430 nm; m is mass of raw materials in grams, g; and W is weight loss during drying of raw materials, %.

Determination of the amount of tannins in terms of tannin. The content of tannins was determined by titration with a solution of potassium permanganate. To extract tannins, 2 g of dry crushed raw material was placed in a 500 mL flask, 250 mL of boiling water was added, and it was heated under reflux in a water bath for 30 min with occasional stirring. Next it was cooled to room temperature, filtered through cotton wool, and filled with water to the required volume. Then 25.0 mL of aqueous extract was taken into a flask, 500 mL of water and 25 mL of indigosulfonic acid solution were added and titrated with constant stirring with potassium permanganate solution (0.02 mol/L)until it was golden yellow. As a control, 25 mL of indigo sulfonic acid solution was added to 525 mL of water and titrated with constant stirring with a solution of potassium permanganate (0.02 mol/L) until the color turned golden yellow.

The content of total tannins in terms of tannin in absolutely dry raw materials in percent (X) was calculated using the formula

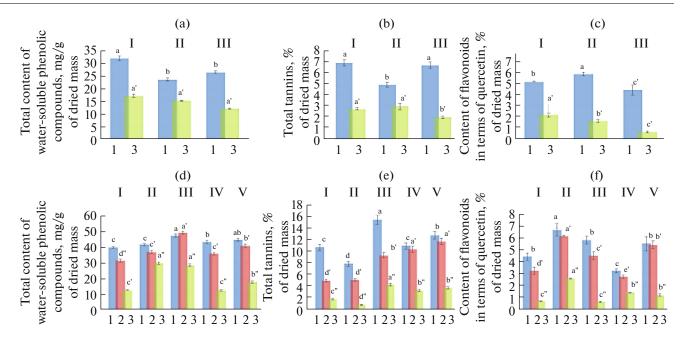
$$X = \frac{(V - V_1) \times 0.004157 \times 250 \times 100}{a \times 25(100 - W)},$$

where V is the volume of 0.02 M potassium permanganate solution used for titration of the aqueous extract, mL;  $V_1$  is the volume of 0.02 M potassium permanganate solution used for titration in the control experiment, mL; 0.004157 is the amount of tannins corresponding to 1 mL of 0.02 M solution of potassium permanganate (in terms of tannin), g; a is the weighed portion of raw materials or medicinal herbal preparation, g; W is humidity of medicinal plant raw materials or medicinal herbal preparation, %; 250 is the total volume of aqueous extraction, mL; and 25 is the volume of aqueous extract taken for titration, mL.

To prepare a solution of indigo sulfonic acid, 1.0 g of indigo carmine was dissolved in 25 mL of concentrated sulfuric acid, then an additional 25 mL of sulfuric acid was added and diluted with water to 1000 mL [12].

Determination of the content of phenolic compounds by HPLC. To carry out chromatographic analysis, we initially carried out alcohol extraction of phenolic compounds in 70% ethanol in a water bath for 90 min [13]. The identification of phenolic compounds was next carried out using high-performance liquid chromatography on a high-pressure chromatography system (Bio-Rad, United States). We used the SN-421001911 original column, 5  $\mu$ m, 4  $\times$  250 mm (United States). Peak detection was carried out using a BioLogic QuadTec UV-Vis dual-wave UV HPLC detector (Bio-Rad, United States) at a wavelength of 260 nm. The mobile phase used was 98% water, 1% acetic acid, and 1% acetonitrile (solution A) and 99% acetonitrile and 1% acetic acid (B). Elution: linear gradient of mobile phase A to phase B was 30-80% from 0 to 9 min, followed by isocratic elution of 80% of phase A to phase B from 9 to 15 min; the flow rate was 1 mL/min. HPLC was carried out at room temperature ( $25 \pm 2^{\circ}$ C). To identify the peaks detected in the chromatogram, working standard solutions of quercetin (CAS 117-39-5,  $\geq 95\%$ , Sigma Aldrich, Germany), protocatechuic acids (CAS 99-50-3, ≥98%, Sigma Aldrich, Germany), chlorogenic acid (CAS 327–97–9, ≥95%, Sigma Aldrich, Germany), luteolin (L9283-10MG,  $\geq 98\%$ , Israel), kaempferol (CAS 520-18-3,  $\geq 90\%$ , Sigma Aldrich, France), coumaric acid (CAS 501– 98-4,  $\geq 98\%$ , Sigma Aldrich, Germany), benzoic acid (CAS 6585–0, ≥99.5%, Merck Millipore, Russia), ferulic acid (CAS 537-98-4, ≥98%, Sigma Aldrich, Germany), caffeic acid (CAS 331-39-5, ≥98%, Sigma Aldrich, Germany), and sinapic acid (CAS 530-59- $6, \geq 98\%$ , Sigma Aldrich, Germany) were used.

Statistical analysis. All experiments were carried out at least five times. Statistical data processing was carried out in the OriginPro 2021 program using a two-tailed Mann–Whitney test with Bonferroni correction for multiple comparisons when P < 0.05. Before analyzing the relationships between environmental factors and the content of phenolic compounds, data were tested for normality of distribution using the Shapiro–Wilk test. The relationship between parameters was assessed using Pearson correlation analysis.



**Fig. 1.** Content of phenolic compounds in *B. pilosa* (a, b, c): I—Kirimiro; II—Mugamba; III—Buragane and *B. tripartita* (d, e, f): I—Vysokogorsky raion; II—Spassky raion; III—Kirimiro region; IV—Mugamba region; V—Buragane region. (1) Leaves; (2) flowers; (3) stem. The same letters indicate the absence of statistically significant differences between samples collected in different areas at R < 0.05.

# RESULTS

Content of phenolic compounds in the studied medicinal plants. In the course of our work, a comparative analysis of the content of WPC, tannins, and flavonoids in the studied medicinal plants collected from different habitats was carried out. It was found that the content of the studied phenolic compounds in the studied species of medicinal plants growing in different regions of the Republic of Tatarstan and the Republic of Burundi varies widely depending on the place of growth and differs in different plant organs (Fig. 1). It has been established that leaves have a higher content of phenolic compounds compared to flowers and stems. As can be seen from Fig. 1, the WPC content in *B. pilosa* plants varied from 23.66 to 32.01 mg/g in leaves and from 12.19 to 17.33 mg/g dry weight in stems. Plants that were collected from the Kirimiro region had the highest WPC content compared to the Mugamba and Buragane regions. Plants from Buragane and Mugamba did not differ in the content of these compounds.

The tannin content ranged from 5.04 to 7.04% dry weight in the leaves and from 2.04 to 3.05% dry weight in the stems. As can be seen from Fig. 1b, a greater amount of tannins accumulates in the leaves compared to the stems. At the same time, plants from Kirimiro and Buragane were characterized by a higher content of tannins than plants from Mugamba.

Flavonoid content in *B. pilosa* leaves was in the range from 4.95 to 5.91% dry weight (in terms of quercetin). The largest amount of quercetin was found in

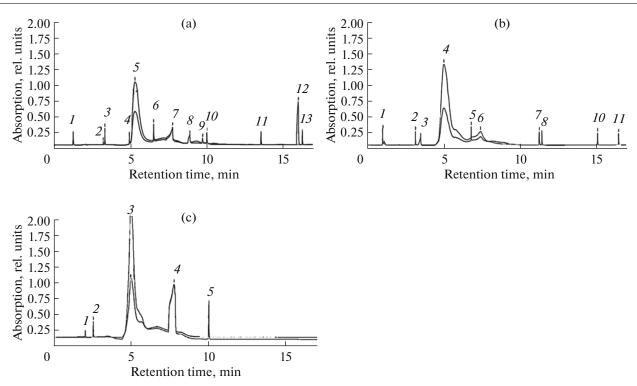
plants from Mugamba (Fig. 1c). Thus, *B. pilosa* plants with the highest levels of phenolic compounds were found in the Kirimiro regions compared to Buragane and Mugambe.

In *B. tripartita* plants, WPC content varied from 40.65 to 48.24 mg/g in leaves, 32.02 to 49.82 mg/g in flowers, and 12.96 to 30.48 mg/g dry weight in stems (Fig. 1d). In general, the leaves and flowers of the *B. tripartita* plant contained approximately the same amount of WPC. Plants collected in the Vysokogorsky and Spassky raions of the Republic of Tatarstan did not differ statistically in this indicator nor did plants from the regions of Kirimiro and Buragane in the Republic of Burundi.

The highest amount of tannins was found in the leaves and the lowest in the stems. In the leaves of the *B. tripartita* plant, the amount of tannins ranged from 8.04 to 15.68% of dry weight. It was found that plants from the Vysokogorsky raion in the Republic of Tatarstan and plants from the Kirimiro region in the Republic of Burundi contained more tannins compared to other regions (Fig. 1e).

The highest value of flavonoids was observed in plants collected from the Spassky raion of the Republic of Tatarstan and from Kirimiro and Buragane regions of the Republic of Burundi. They were most abundant in the leaves and the amount varied from 3.29 to 6.77% of dry weight (in terms of quercetin) (Fig. 1f).

It should be noted that *B. tripartita* plants are characterized by higher amounts of phenolic compounds



**Fig. 2.** HPLC chromatograms of aqueous-alcoholic plant extract *B. pilosa* from different places of growth: (a) Kirimiro region: (2) gallic acid, (4) quercetin, (5) kaempferol, (7) protocatechin, (10) chlorogenic acid, (12) cinaroside; (b) Mugamba region: (2) gallic acid, (4) quercetin, (5) catechin, (8) caffeic acid; (c) Buragane region: (3) quercetin, (4) protocatechin, (5) chlorogenic acid, (6) luteolin.

than *B. pilosa* plants. At the same time, *B. tripartita* plants growing in temperate continental (Tatarstan) and tropical (Burundi) climates have almost the same content of phenolic compounds.

Qualitative composition of phenolic compounds in the studied medicinal plants. When studying the profile of individual phenolic compounds using HPLC in the studied plants, it was found that plants of the same species do not always synthesize the same substances and in the same quantities. Each plant species has a set of individual phenolic compounds that play specific roles in the plant in adaptation and defense against abiotic and biotic stressors.

As can be seen from Figs. 2a–2c, *B. pilosa* plants are rich in various individual phenolic compounds, with the predominance of kaempferol in the Kirimiro region and quercetin in the Mugambe and Buragane regions. It is worth noting that *B. pilosa* plants in the Kirimiro region have the most diverse phenolic composition.

Qualitative analysis of *B. tripartita* showed the greatest diversity of phenolic compounds in plants from the forest-steppe zone of the Spassky raion of the Republic of Tatarstan as well as in plants from the Buragane and Kirimiro regions of the Republic of Burundi (Figs. 3a–3d). The predominance of sinapic acid was found in plants from the Spassky raion and the Kirimiro and Buragane regions.

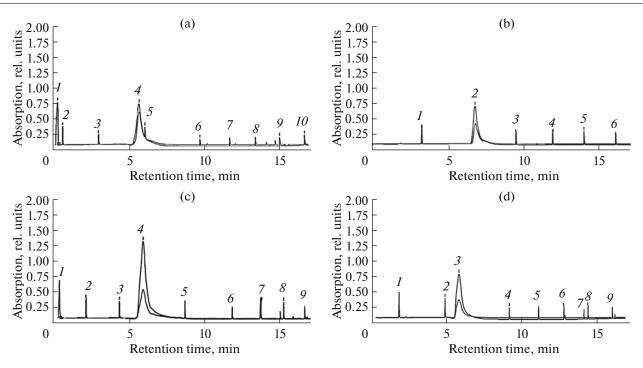
The influence of ecological and geographical conditions on the content of biologically active substances in the studied resource-useful plants. To identify the decisive ecological and geographical factors influencing the level of the studied phenolic compounds in medicinal plants, a Pearson correlation analysis was carried out.

A high correlation was found in *B. pilosa* plants between phenolic content and geo-climatic factors as well as soil mineral elements. According to Table 3, soil nitrogen content and average annual temperature were negatively correlated with quercetin content in these plants, while altitude and precipitation were positively correlated. Content of WPC and tannins in *B. pilosa* plants positively correlated with the content of calcium and magnesium in the soil (Table 3).

Quercetin content in *B. tripartita* plants was positively correlated with average temperature, soil calcium and magnesium content, and altitude above sea level, while an inverse correlation was observed with precipitation. The content of tannins was positively correlated with the content of calcium and magnesium in the soil, and the content of WPC was positively correlated with the content of magnesium in the soil and negatively with the content of phosphorus (Table 4).

#### DISCUSSION

Environmental factors, such as altitude, light, temperature, precipitation, moisture, and soil composi-



**Fig. 3.** HPLC chromatograms of aqueous-alcoholic plant extract *B. tripartita* from different places of growth: (a) Spassky raion: (4) sinapic acid, (6) chlorogenic acid, (7) caffeic acid, (10) luteolin; (b) Vysokogorsky raion: (2) catechin, (4) caffeic acid, (6) cinaroside; (c) Buragane region: (2) cinnamic acid, (4) sinapic acid, (5) cis-5 caffeoylquinic acid, (6) caffeic acid; (d) Kirimiro region: (2) quercetin, (3) sinapic acid, (5) caffeic acid, (7) coumaric acid.

tion, influence the synthesis and accumulation of phenolic compounds in the studied plants. Research by various authors [14] prove that the amount of phenolic compounds can increase significantly under the influence of various environmental stress factors. They are powerful antioxidants and, therefore, effectively provide plant resistance to a number of biotic and abiotic stressors [15]. Their increase affects the stress resistance of plants. Previous studies have shown that temperature stress increases the synthesis of phenolic compounds in plants but to varying degrees depending on the plant species. In general, cold-tolerant plants appear to accumulate higher amounts of phenolic compounds at low temperatures and heattolerant plants at higher temperatures. It is also known that altitude plays an important role in the formation of secondary metabolites that ultimately neutralize free radicals. An increase in altitude above sea level creates a certain stress for the plant, including due to increased UV radiation and decreased temperature, which accelerate the formation of free radicals [16].

Soil composition is another factor that can influence the content of phenolic compounds. According to the literature, the activity of the enzyme phenylalanine ammonia lyase, responsible for the synthesis of polyphenols, increases at high potassium ( $K^+$ ), magnesium ( $Mg^{2+}$ ) and phosphorus ( $PO_4^{3-}$ ) concentrations [17]. Other studies have reported the effect of long-term and constant nitrogen starvation on enhancing the synthesis of carbon-containing compounds, including accumulation of phenols [18–20]. Our results confirm the influence of the above factors on the accumulation of phenolic compounds.

 Table 3. Values of the Pearson correlation coefficient between environmental factors and the content of phenolic compounds in plants *B. pilosa*

DAV	Ecological and geological factors of the habitat									
BAV	temperature	precipitation	height	Mg <sup>2+</sup>	Ca <sup>2+</sup>	$K^+$	NO <sub>3</sub>	$PO_4^-$		
WPC	0.69*	-0.70	-0.43	0.92*	0.99*	-0.08	0.07	-0.67*		
Querce tin	-0.95*	0.97*	0.99*	-0.41	-0.66	0.82	-0.97*	-0.12		
Tanning substances	0.94*	-0.58	-0.78*	0.99*	0.98*	0.55	0.61	-0.26		

\* Statistically significant correlation from (P < 0.05).

DAV	Ecological and geological factors of the habitat									
BAV	temperature	precipitation	Height	Mg <sup>2+</sup>	Ca <sup>2+</sup>	K <sup>+</sup>	NO <sub>3</sub>	PO <sub>4</sub> <sup>3-</sup>		
WPC	0.55	-0.56	-0.68	0.8*	0.73*	-0.03	0.03	-0.78*		
Querce tin	0.85*	-0.94*	0.99*	1*	0.98*	0.56	0.61	-0.27		
Tanning substances	0.48	-0.63	-0.76*	0.87*	0.95*	0.08	0.14	-0.70		

 Table 4. Values of the Pearson correlation coefficient between environmental factors and the content of phenolic compounds in plants *B. tripartita*

\* Statistically significant correlation from ( $P \le 0.05$ ).

Indeed, the Kirimiro region, which was characterized by high contents of WPC and tannins in *B. pilosa* plants and all studied groups of phenolic compounds in *B. tripartite* plants, is characterized by fairly acidic soils, low nitrogen, and high calcium (Ca<sup>2+</sup>) and magnesium (Mg<sup>2+</sup>). The Mugamba region, in which high values of flavonoids were found in terms of quercetin in *B. pilosa* plants, is located at the highest altitude above sea level with low average daily temperatures, which confirms the protective function of quercetin under stress conditions as a powerful antioxidant [21, 22].

In RT in B. tripartita plants, the highest content of phenolic compounds was found in the Spassky raion compared to the Vysokogorsky raion. The Spassky raion differed from the Vysokogorsky raion in lower temperatures and slightly alkaline soil with a high content of nitrogen and potassium. However, some scientific studies show that polyphenol content increases in response to nitrogen deficiency in plants [24]. It was shown in [25] that the content of flavonoids and water-soluble phenolic compounds increased in Brassica oleracea var. sabellica leaves when adding N: P: K. A high content of phenolic compounds, including flavonoids, have been found in Eleutherine *palmifolia* plants treated with N : K [26, 27]. The observed contradictions in the influence of mineral substances on the content of phenolic compounds in plants may be associated with the presence of other factors (since plants are simultaneously influenced by several factors in the natural environment), the amount and availability of mineral substances, and plant characteristics.

It is worth noting that *B. pilosa* and *B. tripartita* plants growing in temperate continental (Tatarstan) and tropical (Burundi) climates differ slightly in the content of phenolic compounds, which indicates a genetically determined narrow amplitude of variability in the metabolism of these species.

While studying the profile of individual phenolic compounds using HPLC, we found the presence of kaempferol and cynaroside only in *B. pilosa* plants from Kirimiro, caffeic acid and catechin only in plants from Mugamba, and luteolin only in plants from Buragane. Quercetin was present in all samples. It can

be assumed that there is a set of unique environmental factors that contribute to the accumulation of certain phenolic compounds.

As is the case with *B. pilosa, B. tripartita* plants use different phenolic compounds (such as chlorogenic acid and luteolin in plants from the Spassky raion, catechin and cinaroside in plants from the Vysokogorsky raion; cinnamic acid and 5 cis-caffeolquinic acid in plants from Buragane and coumaric acid in plants from Kirimiro) for adaptation to different habitats, which explains their wide distribution. Moreover, in all *B. tripartita* samples (except from Vysokogorsky raion), a predominance of sinapic acid was found. Sinapic acid is one of the most abundant hydroxycinnamic acids in plants, exhibiting powerful antioxidant effects. The antioxidant activity of sinapic acid is comparable to caffeic acid [28].

Thus, it has been established that *B. pilosa* and *B. tripartita* plants from the Republic of Burundi and the Republic of Tatarstan growing in completely different conditions have similar contents of biologically active substances. Similar adaptation mechanisms of the studied species probably ensure their survival in completely different conditions of the Republic of Tatarstan and the Republic of Burundi.

Analysis of plant growth conditions in different regions of the Republic of Burundi revealed that the most favorable conditions for the accumulation of phenolic compounds in two *Bidens* species is the combination of environmental factors in the Kirimiro region. The Kirimiro and Buragane regions are at the same altitude, the amount of precipitation there is approximately the same, but the average daily temperatures are slightly lower in the Kirimiro region and the soils are quite acidic, with low nitrogen content. At the very least, the content of phenolic compounds could be affected by a significantly low level of nitrogen in the soils of this region (Table 1).

Thus, the soil conditions (high magnesium, low nitrogen, and rather acidic soils) and relatively high average temperatures (19°C) characteristic of the Kirimiro region favor the accumulation of biologically active substances in *B. pilosa* and *B. tripartita* plants. For plants in the Republic of Tatarstan, apparently, in addition to the above, an important factor is zoning (forest-steppe zone, Spassky raion).

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### ETHICS APPROVAL AND CONSENT TO PARTICIPATE

This work does not contain any studies involving human and animal subjects.

#### CONFLICT OF INTEREST

The authors of this work declare that they have no conflicts of interest.

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