Biologically Active Compounds and Antioxidant Activity of the Plants from CSBG SB RAS Collection of the Asteraceae Family

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Abstract. The article presents data on the content of biologically active compounds (BAC) and the total phenolic antioxidants activity (TPA) evaluated for leaves and inflorescences of Achillea millefolium, Anthemis tinctoria, Artemisia absinthium, A. dracunculus, Leucanthemum vulgare, Pyrethrum balsamita, P. macrophyllum, Tanacetum vulgare. The leaves and inflorescences of the plants contain flavonols, catechins, tannins, carotenoids, pectin substances. High content of phenolic compounds was found in Artemisia dracunculus and Achillea millefolium (352.34 – 398.99 mg/g of DW). The highest level of catechins was registered in inflorescences (8.01 mg/g) and leaves (4.58 mg/g) of Artemisia absinthium. The content of pectin substances in the studied plants was quite high, with the biggest share of propectins (50.6 – 95.9 mg/g). The maximum quantity of pectins was found in the leaves and inflorescences of Leucanthemum vulgare (11.2-12.7 mg/g), propectins – in the leaves of Tanacetum vulgare (95.9 mg/g). The highest content of carotenoids was found in the leaves of plants, except for Leucanthemum vulgare. The highest antioxidant activity was recorded for water-ethanol extracts from leaves of Achillea millefolium (up to 1.09 mg/g) and inflorescences of Artemisia dracunculus (0.94 mg/g) plants due to the increased content of phenolic compounds, in particular flavonols and tannins, compared to the other species.

The Asteraceae family belongs to the largest family of flowering plants and contains more than 24,000 species belonging to 1,400 genera [1-2]. Tribe Anthemideae Cass. comprising 111 genera with c. 1,800 species, distributed worldwide (extratropical), but with main concentrations in central Asia, the Mediterranean region, and southern Africa [2]. Members of the tribe are well known as aromatic plants, and some are utilized for their pharmaceutical and/or pesticide value. Three main classes of chemical substances are of considerable systematic significance: acetylenes, sesquiterpene lactones, and flavonoids [2].

Eight species of the tribe Anthemideae presented in the CSBS collection were selected for the study of the BAC content and the total activity of phenolic antioxidants: Achillea millefolium L., Anthemis tinctoria L., Artemisia absinthium L., A. dracunculus L., Leucanthemum vulgare L., Pyrethrum balsamita (L.) Wild., P. macrophyllum (Waldst. & Kit.) Willd., Tanacetum vulgare L. Many of these plants have a sufficient raw material

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base, are used in official and folk medicine, have shown promise in experimental studies for various types of activity. At the same time, biologically active compounds are of particular interest from this point of view, in particular, phenolic compounds, pectin substances, carotenoids, which have a wide spectrum of biological activity. In addition, recently there has been a growing interest in the study of the antioxidant and antiviral activity of plants for the creation of new drugs. The purpose of this research was a comparative study of the content of biologically active compounds, and the total activity of phenolic antioxidants in leaves and inflorescences of Asteraceae plants (tribe Anthemideae), successfully introduced in CSBG SB RAS.

1. Materials and Methods

The materials were selected from plants cultivated on the introduction plot of the CSBG during the flowering phase in 2019. The raw materials were divided into leaves, and inflorescences, dried and crushed to pick representative samples for analysis.

The total flavonol content of the extracts was determined spectrophotometrically according to the aluminum chloride colorimetric method. The amount of flavonols was expressed as milligrams of rutin equivalent per gram of dry weight (mg of RE/g of DW) through the calibration curve with rutin [3]. The content of catechins was detected by the method based on the ability of catechins to give crimson staining with a solution of vanillin in concentrated hydrochloric acid. The amount of catechins was expressed as milligrams of (+)-catechin equivalent per gram of dry weight (mg of CE/g of DW) through the calibration curve with catechin [4]. Detection of tannins (hydrolyzable tannins) was performed using a spectrophotometric method. The tannin content of the extracts was calculated using a tannin calibration curve. The results were expressed in mg of tannin equivalents (TE/g of DW) [5].

The content of carotenoids was detected in acetone-ethanol extraction [6-7]. Pectin substances (pectins and protopectins) were determined by a carbazole-free method based on obtaining specific yellow-orange staining of uronic acids with thymol in a sulfuric acid medium [8]. To detect the total activity of phenol-type antioxidants, an operational amperometric method was used [9]. The measurements were performed on the device “Tsvet Jauza-01-AA”. Before the measurement, a calibration curve was constructed to determine the dependence of the signal of the comparison sample (Gallic acid) on its concentration. The amount of total activity of phenolic antioxidants was expressed as milligrams of (+)-gallic acid equivalent per gram of dry weight (mg of GAE/g of DW) through the calibration curve with gallic acid. All biochemical parameters were calculated for the mass of absolutely dry raw materials. Detection of the content of BAC was carried out in threefold repetition, TPA – in fivefold. Statistical data processing was based on descriptive statistics using Statistica 8.0 and Microsoft Excel 2010 programs.

2. Results and Discussion

It was found that the leaves and inflorescences of the plants contain a complex of biologically active substances.

**Phenolic compounds.** The leaves and inflorescences of the plants contained flavonols, tannins, and catechins. Their content varied depending on the type and organ of the plant (Table 1). The content of flavonols and catechins in inflorescences was usually higher than in leaves, while tannins content, on the contrary, was higher in leaves. The highest content of flavonols was observed in leaves of Achillea millefolium (135.4 mg of RE/g) and inflorescences of Artemisia dracunculus (109.4 mg of RE/g). A high content of flavonols (more than 40 mg of RE/g) was found in Achillea millefolium and Artemisia
dracunculus plants and the inflorescences of *Anthemis tinctoria* (47.3 mg of RE/g). Low content of flavonols, less than 10 mg of RE/g, noted in leaves of *Anthemis tinctoria* and *Artemisia absinthium* plants. Tannins were predominantly accumulated in leaves compared to inflorescences, except for *Anthemis tinctoria*. High content of tannins (more than 200 mg of TE/g) is found in leaves and inflorescences of *Artemisia dracunculus*, in leaves of *Achillea millefolium* and *Pyrethrum macrophyllum*, in inflorescences of *Anthemis tinctoria*. Two species *Artemisia* showed significant catechin content from 2.10-4.58 mg of CE/g in leaves to 3.38-8.01 mg of CE/g in inflorescences.

In general, *Artemisia dracunculus* и *Achillea millefolium* accessions characterized in this study indicated a great deal of phenolic content.

**Table 1.** The content of phenolic compounds in aboveground organs of plants

| Plants                  | Underground organs | Flavonols (mg of RE/g of DW±SD) | Tannins (mg of TE/g of DW±SD) | Catechins (mg of CE/g of DW±SD) |
|-------------------------|--------------------|--------------------------------|------------------------------|-------------------------------|
| *Leucanthemum vulgare*  | Leaves             | 13.6±0.1                       | 194.2±6.3                    | 1.03±0.02                     |
|                         | Inflorescences     | 30.2±0.2                       | 116.6±0.4                    | 0.80±0.01                     |
| *Pyrethrum balsamita*   | Leaves             | 19.0±0.1                       | 175.9±5.8                    | 1.97±0.05                     |
|                         | Inflorescences     | 12.9±0.1                       | 102.1±3.6                    | 1.40±0.04                     |
| *Pyrethrum macrophyllum*| Leaves             | 32.6±0.1                       | 208.4±9.4                    | 0.80±0.03                     |
|                         | Inflorescences     | 22.5±0.2                       | 104.6±2.8                    | 0.91±0.03                     |
| *Artemisia dracunculus* | Leaves             | 63.2±1.0                       | 327.6±10.3                   | 2.10±0.03                     |
|                         | Inflorescences     | 109.4±2.0                      | 286.2±8.9                    | 3.38±0.04                     |
| *Artemisia absinthium*  | Leaves             | 6.7±0.1                        | 108.1±1.8                    | 4.58±0.03                     |
|                         | Inflorescences     | 13.0±0.1                       | 96.4±2.1                     | 8.01±0.05                     |
| *Achillea millefolium*  | Leaves             | 135.4±4.2                      | 215.6±9.8                    | 1.34±0.03                     |
|                         | Inflorescences     | 74.9±1.1                       | 94.4±3.1                     | 2.01±0.02                     |
| *Anthemis tinctoria*    | Leaves             | 9.3±0.3                        | 156.3±4.0                    | 0.53±0.02                     |
|                         | Inflorescences     | 47.3±0.5                       | 271.6±10.6                   | 0.55±0.02                     |
| *Tanacetum vulgare*     | Leaves             | 37.7±0.3                       | 194.9±9.1                    | 1.15±0.02                     |
|                         | Inflorescences     | 25.6±0.4                       | 125.9±2.9                    | 0.98±0.02                     |

R: Rutin equivalents; T: Tannin equivalents; C: catechin equivalent; SD: Standard deviation; DW: Dry weight.

**Pectin substances.** Pectin substances (or pectin) belong to a group of plant polysaccharides. Pectin substances in the form of water-insoluble compounds are known as protopectins. The content of pectin substances in the studied plants was quite high with the main share of protopectins in the total substances (Fig. 1).

The content of pectins in plants varies from 2.9 to 12.7 mg/g, protopectins from 50.6 to 95.9 mg/g. *Leucanthemum vulgare* and *Anthemis tinctoria* plants are distinguished by a significant accumulation of pectins in leaves and inflorescences (11.2-12.7 mg/g). The highest content of protopectins was found in the leaves of *Tanacetum vulgare* and *Artemisia dracunculus* plants (95.9 - 93.7 mg/g), and in the inflorescences of *Pyrethrum balsamita* (95.4 mg/g). Low content of pectin substances was found in the leaves of *Artemisia absinthium* (66.3 mg/g).

**Pigments.** Carotenoids play an important role in photosynthesis, in protection from the harmful effects of UV radiation, and make a certain contribution to the color of tissues containing them. According to our data, the content of carotenoids was usually higher in leaves than in inflorescences, except for *Leucanthemum vulgare* and *Anthemis tinctoria* (fig. 2). The minimum content of carotenoids in leaves and inflorescences was found for *Pyrethrum macrophyllum* plants (0.23 mg/g and 0.06 mg/g). The highest content
of carotenoids was notably observed in leaves of *Achillea millefolium* (0.72 mg/g) and inflorescences of *Leucanthemum vulgare* (0.53 mg/g).

![Fig. 1. Contents of Pectin Substances in Aboveground Organs of Plants (l – leaves, fl – inflorescences)](image1)

**Fig. 1.** Contents of Pectin Substances in Aboveground Organs of Plants (l – leaves, fl – inflorescences)

**Fig. 2.** Content of Carotenoids in Aboveground Organs of Plants

*Antioxidant activity.* The research evaluated the total antioxidant activity of water-alcohol extracts from leaves and inflorescences of the studied plants. It was found that the samples exhibit different antioxidant activity (Fig. 3). High antioxidant activity was shown by plants of *Achillea millefolium* and *Artemisia dracunculus*. The maximum total content of antioxidants was found in leaves of *Achillea millefolium* (1.09 mg of GAE/g).

![Fig. 3. Total Antioxidant Content in Aboveground Organs of Plants](image3)

**Fig. 3.** Total Antioxidant Content in Aboveground Organs of Plants
High antioxidant activity of inflorescences and leaves of *Achillea millefolium* and *Artemisia dracunculus* plants, is associated with an increased content of phenolic compounds (flavonols, tannins) in these samples. Analyzing the correlation between the content of BAC in leaves and inflorescences of the studied plants with anti-oxidant activity, performed by the method of correlation analysis, it was determined that flavonols are most closely associated with anti-oxidant activity (R = 0.92) (Table 2), that is supported by literature data for other plant species [10].

|                | Flavonols | Tannins | Catechin | Pectins | Protopectins | Carotenoids |
|----------------|-----------|---------|----------|---------|--------------|-------------|
| **R**          | **0.92**  | 0.45    | 0.00     | -0.27   | -0.23        | 0.08        |

*Marked correlations are significant at p < 0.05 N=16

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