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PHYTOCHEMICAL INVESTIGATION OF GRASS OF INULA BRITANNICA L.

Yerenko Olena

Ph.D., Assistant of the
Department of Management and Pharmacy
Economics and Medicinal Preparations Technology
Zaporizhzhya State Medical University

The *Inula* L. genus of the Asteraceae family contains over 200 species, more than 30 of which sprout in Ukrainian flora. Of all 30 species of *Inula* L. genus, growing on the territory of Ukraine. But during the harvesting phylogenetically close species are constantly found: *I. britannica* L., *I. oculus* – Christi L., *I. germanica* L., *I. salicina* L., *I. aspera* Poir., *I. sabuletorum* Czern. ex Lavr., *I. vulgaris* Lam., *I. thapsoides* Bieb. ex Willd. Spreng., *I. ensifolia* L. and *I. macrophylla* Kar. et Kir. The plants are most often found and reproduce their thickets in Dnipro, Zaporizhzhya, Vinnytsya, Donetsk, Khmelnytskyi, Odessa, Ternopil and Chernivtsi regions [1, 2].

The representatives of *Inula* L. genus are perspective phytotherapy objects, which are traditionally used in scientific medicine and ethnoscience of many countries as expectorants during respiratory diseases, as diuretic, choleretic, anthelmintic and gall-forming agents. Thereby, the species containing a lot of biologically active compounds that have antiphlogistic, gastroprotective, antioxidant effects, are of particular interest now, namely *Inula britannica* L. [3, 4, 5, 6, 7, 8].

The purpose of this work is making qualitative analysis of ascorbic, organic acids and tannic substances content in the raw material of investigated species of *Inula britannica* L.

One of the most important physiological functions of ascorbic acid and free organic acids is the ability to regenerate the tissues and to normalize capillary permeability through collagen and procollagen synthesis. Their participation in blood coagulation mechanisms is of particular importance since the activity of platelets, prothrombin complex and thromboplastin depends on their presence. Scientific sources confirm vitamin C presence in raw material of many plant species. However, there is almost no information about ascorbic acid quantitative content.

The literature sources investigated indicate the presence and accumulation of ascorbic, free organic acids and tannic substances in the grass of many species of *Inula* L. genus. However, the quantitative content of substances and their accumulation during vegetation are not established.

The presence of organic acids in raw material of investigated species of *I. britannica* L. was established with the help of paper chromatography and thin-layer chromatography methods in system ether petroleum - acetic acid - water (13:3:1) using compounds of working reference substances (Ascorbic acid Sigma-Aldrich (Germany)).

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The quantitative determination of total organic acids was carried out according to XI edition of State Pharmacopoeia methods in terms of malic acid (Malic Acid DL (China)). In order to eliminate impact on accuracy during ascorbic acid determination, the concentrations of this substance (titration with 2,6-dichloro-sodiumphenolindophenolate solution) were taken into account in calculations [9].

Water extracts were studied for the tannins presence in raw material of *I. britannica* L. investigated species. Compounds identification was carried out through chemical reactions in accordance with Ukrainian State Pharmacopeia.

The quantitative tannins content in raw material was carried out according to the XI edition of State Pharmacopoeia methods.

The presence of ascorbic and free organic acids in the grass of investigated species of *I. britannica* L. was determined on the previous step using paper chromatography and thin-layer chromatography methods in systems acetic acid 15%, ether petroleum - acetic acid - water (13:3:1) [10, 11]. At the same time, the chromatography of working reference substances was carried out. In order to display the ascorbic acid spots, 0.001 N solution of 2,6-dichloro-sodiumphenolindophenolate were used.

Maximum substances accumulation was observed during the period of mass vegetation and bud formation. Thus, the concentration of ascorbic acid in *Inula britannica* L. grass was established equal up to $0.48 \pm 0.05\%$ and $0.47 \pm 0.05\%$. At the same time, the content of free organic acids in *Inula britannica* L. grass was equal up to $0.19 \pm 0.02\%$ and $0.18 \pm 0.02\%$. High concentrations of compounds in the investigated species' vegetalraw material persisted until the end of flowering period.

In order to segregate tannins, the investigated raw material was extracted with the help of hot water; then the extract was purified from the related compounds presence by successive action of ether petroleum benzene, followed by a mixture of benzene and chloroform (1:1) and, finally, by petroleum ether and ethyl acetate. The preliminary extraction of investigated raw material with the help of nonpolar or low polar organic solvents was used to separate chlorophyll, lipids and terpenoids. Then, extraction with the help of ethanol with purification by column chromatography on polyamide was performed with the purpose of tannins segregation. Catechins were detected in the form of three pale blue spots ($R_f = 0.44, 0.56$ and 0.73). The chromatograms were dried, treated with iron (III) chloride solution, and a brownish-red stains colouring was observed. Catechin, catechin gallates and gall acid solutions (+) were used as working reference substances.

It was found out that raw material of *I. britannica* L. investigated species contained the following compounds from condensed tannins class: (+) catechin, catechin gallat. The tannins concentration in raw material is determined by gravimetry, photometry, spectrophotometry and other methods.

Significant tannins concentrations in raw material of *I. britannica* L. investigated species are important in the development of hepatoprotective effect of lyophilic extracts' biologically active compounds complex. The tannins content in *Inula britannica* L. grass during the flowering period was respectively equal to $9.30 \pm 0.92\%$ and $8.62 \pm 0.81\%$.

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Thus, the *Inula britannica* L. grass contained quite significant concentrations of ascorbic acid, free organic acids and tannic substances, which greatly influence the biological effect of herbal medicinal products made of raw material.

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