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THE OPTIMIZING THE EXTRACTION PROCESS OF BIOLOGICALLY ACTIVE SUBSTANCES OBTAINED FROM DANDELION LEAVES (*TARAXACUM OFFICINALE* WIGG)

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Abstract

Purpose: *to optimize the process of separation of biologically active substances from medicinal dandelion leaves.* ***Methods:*** *Different methods for the selection of biologically active substances from the medicinal dandelion (Taraxacum oficinale Wigg.), Including ultrasound treatment, were performed at 4, 5 and 6 W/m^2 at 5, 10, 15, 20, 25 minutes.* ***Results:*** *The aquatic extracts of dandelion officinalis (Taraxacum officinale Wigg) have been obtained for selecting biologically active substances, namely vitamin C. The optimal conditions for the extraction process are chosen: the ratio of raw material:extractant 1:20, extraction time - 30 minutes, temperature - 25 ° C . The extraction was performed with distilled water.* ***Discussion:*** *It was investigated that the extraction of medicinal plant material results in the diffusion of biologically active substances from the internal structures of the material particle. This process has its own peculiarities. First of all, the presence of porous septum, intercellular space and cellular movements reduces the diffusion rate. Secondly, in the porous partition, only those substances whose particles do not exceed pore sizes can penetrate. Also, the phenomenon of desorption observed in the cell after penetration of the extractant into it is characteristic. The optimal conditions for the extraction of biologically active substances from medicinal dandelion (Taraxacum officinale Wigg) were determined: - extractant water; - ratio of extractive raw material (1:20); - time of infusion 30 minutes; - temperature 25 °C. Ultrasonic fluctuations optimize the process of vitamin C extraction from dandelion leaf extracts and as a result, it has been found that the highest yield of ascorbic acid in extracts treatment by ultrasound for 25 minutes with a power of 6 W/m^2 and is 0.077 mg per 100 g of product.*

Keywords: dandelion officinalis (*Taraxacum officinale* Wigg); biologically active substance; ascorbic acid; extraction; ultrasound.

1. Introduction

Much attention has recently been paid to the extraction and study of biologically active substances (BAS) of wild plants, which are widely used in the food, perfumery, cosmetic and pharmaceutical industries. Natural regeneration and adaptability to the environment conditions make wildlife plants an inexhaustible raw material for BAS production [9].

The search and creation of new herbal medicinal products, the development of methods for controlling their quality with the use of modern methods of pharmaceutical analysis, as well as organizing their industrial production, is considered as an important task of biotechnology. The particular attention is paid to plants with centuries-old experience of using in folk medicine, including dandelion officinalis (Taraxacum oficinale Wigg).

Medicinal Dandelion is a perennial herb of Compositae family. It is spread everywhere and blossoms from spring to late fall. The plant has choleretic, antipyretic, expectorant, sedative, antispasmodic and mild hypnotic effects. Roots, leaves, grass, dandelion juice are all used for therapeutic purposes. The leaves of medicinal Dandelion, which contain hydroxycinnamic acids, flavonoids, amino acids, fatty acids, sugars, sterols, coumarins and vitamin C, are particularly rich in biologically active substances [12].

Multicomponent medical preparations, containing both a below-ground part (Hepatophyte, Detoxifit, Tonifit, Nephrophyte) and an above-ground part of medicinal Dandelion (Tonzilgon), are registered and widely used on the Ukrainian pharmaceutical market. The medications are used in case of acute and chronic respiratory disorders, diseases of the urinary tract, urinary-stone disease, as well as chronic cholecystitis, dyskinesia of the biliary tract, gall bladder, etc.

Among medical drugs of domestic production, there are currently no preparations made from the above-ground part of medicinal Dandelion, despite the wide spread of the plant throughout the area of Ukraine.

Methods of identifying and quantifying of biologically active substances (BAS) of medicinal Dandelion have not been elaborated sufficiently. Therefore, the development of BAT research methods and introduction of medicinal plant material into the biotechnological production are thought to be an actual direction of today's research [12].

Research methods and materials. The features of BAS extraction from materials with cellular structure are related to the fact that on the way to the substances contained in the cell, there is a cell wall, whose structure and physiological state may be different. Currently, the vast majority of extractive preparations are obtained from dried plant material, that is, dehydrated by natural or thermal drying. When dried, fresh plants lose water. The protoplasm shrinks and turns into a relatively small lump, the cellular juice passes into dry residue, and the cell's inner part is usually filled with air. Biologically active substances in dried raw materials are found in the form of dry conglomerates in the cell's cavity or adsorbed on cell's walls.

In many enterprises, the technology of long-term infusion of raw materials with an extractant (aqueous or hydroalcoholic solution with a 40-80% alcohol volume fraction) is widely used. The disadvantage of these processes is time duration, the need for a large number of solvents, which requires additional costs. Therefore, the scientists nowadays have developed a wide range of methods that contribute to intensifying the extraction process [5]. Among them, a great deal of attention is focused on the physical method, namely the use of ultrasound.

2. Materials and methods

Medicinal Dandelion raw material (leaves) was collected in the fall of 2016, when the leaves were fading. The raw plant material was dried to constant weight. The extraction of biologically active substances in the medicinal Dandelion raw material was carried out as follows: 2 g of crushed raw material (the degree of crushing is 2-3 mm) was placed into a conical flask of 100 ml capacity, then 40 ml of distilled water was added into it and the extracts were kept at room temperature for 30 minutes, and finally the extracts were processed by ultrasound.

3. Methods for determining vitamin C

There were placed from 1 to 10 cm3 of the test solution with the use of a pipette into a flask of 50 or 100 cm3 capacity, the volume was adjusted to 10 cm3 with the extractant and was titrated with a solution of 2.6-dichlorophenolindophenolate sodium, until a pale pink color appeared and did not disappear within 15-20 seconds.

At the same time, control studies on the content of reducing agents were performed. For doing this, the same amount of extract was placed to the flask, and as in the previous definition there were added the same amount of acetate buffer solution and 36-

40% solution of formaldehyde in the volume corresponding to the half volume of acetate buffer solution, then it was stirred and kept for 10 minutes, previously closing the flask with a plug. After that, the content was titrated with a solution of 2,6-dichlorophenolindophenolate sodium.

The calculation of ascorbic acid (X) output in mg per 100 g of the product is carried out by the formula:

$$X = \frac{(Y_1 - Y_2) \cdot T \cdot Y_3 \cdot 100}{Y_4 \cdot m}$$

4. Results and discussion

It is effective to apply ultrasonic vibrations in order to intensify the extraction process. At the same time, extraction is accelerated and the completeness of extraction of biologically active substances is achieved.

The greatest effect of ultrasound is detected when the cell of the extracted material is well saturated with an extracted ultrasound agent. Emerging ultrasound waves create cavitation. As a result, the impregnation of material and dissolution of cell's content are accelerated, the flow velocity of raw material particles increases, turbulent and vortex flows in the boundary diffusion layer of the extractant emerge. Molecular diffusion in the material cells and in the diffusion layer changes to convective one, which leads to the mass transfer intensification. The emergence of cavitation causes the destruction of cells. In this case, extraction is accelerated through the washing of extractive substances from destroyed cells. [2].

Ultrasonic extraction is one of the most common methods used in the process of obtaining biologically active substances from plant material. All extraction processes are limited by diffusion at the boundary of phase separation through a diffusion layer with a concentration gradient of extracting substance. The usage of ultrasound can significantly accelerate the extraction process, increase the output and reduce the cost of extracting substance, improve working conditions and increase its productivity.

When applying ultrasound, there is a sound-capillary effect. This leads to an increase in the depth and speed of liquid penetration in capillary channels under the action of ultrasound. In addition to accelerating the displacement of air bubbles, the ultrasound also creates conditions for their dissolution in liquid. It results in a sharp reduction of the extraction process [2].

The process of extraction from medicinal Dandelion leaves was performed under standard conditions, when the extracts were placed into flasks. Ultrasound processing was performed at 4, 5 and 6 W / m2 within 5, 10, 15, 20, 25 minutes. Control samples were exposed under the same conditions without irradiation, and the content of vitamin C was determined in all irradiated and control samples.

The content of ascorbic acid (X) in mg per 100 g of the product was determined by the formula:

$$X = \frac{(Y_1 - Y_2) \cdot T \cdot Y_3 \cdot 100}{Y_4 \cdot m},$$

Y1 is the volume of 2,6-dichlorophenolindophenolate sodium solution, which was spent on titrating the sample extract, cm3,

Y2 is the volume of 2,6-dichlorophenolindophenolate sodium solution, which was spent on the control test, cm3;

T is the titre of 2,6-dichlorophenolindophenolate sodium solution, mg / cm3;

Y3 is the extract volume obtained during extracting vitamin C from the product's weight, cm3;

Y4 is the extract volume used for titration, cm3;

M is the mass of product's weight, g

On the basis of obtained data, the diagrams of ultrasound influence on vitamin C extraction from medicinal Dandelion leaves were constructed.

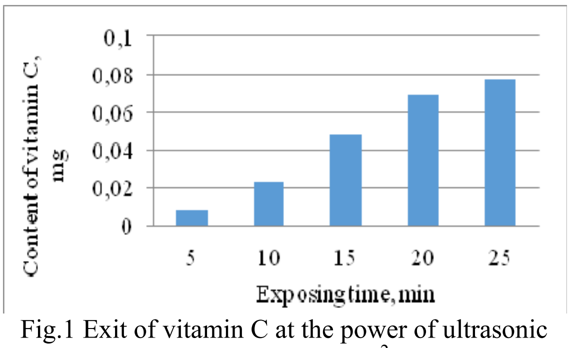

Fig.1 Exit of vitamin C at the power of ultrasonic oscillations 4 W/m^2

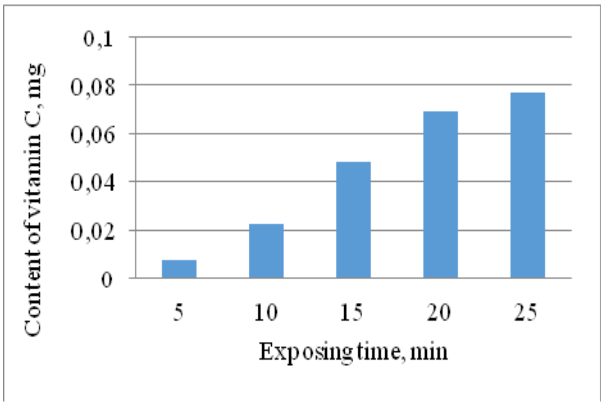

Fig. 2. Exit of vitamin C at the power of ultrasonic oscillations 5 W/m^2

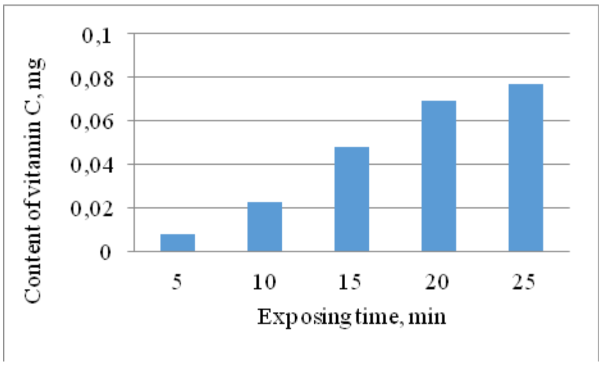

Fig. 3. Exit of vitamin C at the power of ultrasonic oscillations 6 W/m^2

Data in Fig. 1, Fig. 2 and Fig. 3 indicate that the highest content of vitamin C in extracts is observed under the influence of ultrasonic oscillations with a power of 6 W/m^2 for 25 minutes.

5. Conclusions

The optimal conditions for extracting biologically active substances from the leaves of Dandelion officinalis (*Taraxacum officinale* Wigg) were determined:

- extractant-water;
- ratio of raw material - extractant (1:20);
- infusion time - 30 minutes;
- temperature - 25 °C.

It was studied that the ultrasonic vibrations optimize the secretion process of vitamin C from Dandelion leaf extracts.

In addition, it was found that the highest yield of ascorbic acid occurs when processing extracts by ultrasound for 25 minutes with a power of 6 W/m^2 and corresponds to 0.076 mg per 100 g of product.

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Оптимізація процесу екстракції біологічно активних речовин, виділених з листя кульбаби лікарської (*Taraxacum officinale* Wigg).

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Мета: оптимізувати процес виділення біологічно активних речовин з листя кульбаби лікарської. **Методи:** Розглянуто різні методи виділення біологічно активних речовин з кульбаби лікарської (*Taraxacum oficinale* Wigg.), в тому числі обробка ультразвуком, що проводили при потужностях 4, 5 та 6 Вт/м^2 протягом 5, 10, 15, 20, 25 хв. **Результати:** Отримано водні екстракти кульбаби лікарської (*Taraxacum oficinale* Wigg.) для виділення з них біологічно активних речовин, а саме вітаміну С. Підібрано оптимальні умови для процесу екстрагування: співвідношення сировина:екстрагент 1:20, час екстракції – 30 хв, температура – 25 °С. Екстракцію проводили дистильованою водою. **Обговорення:** Досліджено, що при екстрагуванні з лікарської рослинної сировини відбувається дифузія біологічно активних речовин із внутрішніх структур частинки матеріалу. Цей процес має свої особливості. Перш за все, наявність пористої перегородки, міжклітинного простору і клітинних ходів знижує швидкість дифузії. По-друге, у пори перегородки можуть проникати лише ті речовини, частинки яких не перевищують розмірів пор. А також характерним є явище десорбції, що спостерігається в клітині після проникнення в неї екстрагента. Визначено оптимальні умови екстрагування біологічно активних речовин з кульбаби лікарської (*Taraxacum officinale* Wigg): - екстрагент-вода; - співвідношення сировина-екстрагент (1:20); - час настоювання 30 хв; - температура 25 °С. Ультразвукові коливання оптимізують процес виділення вітаміну С з екстрактів листя кульбаби лікарської і в результаті цього встановлено, що найбільший вихід аскорбінової кислоти при обробленні екстрактів ультразвуком протягом 25 хв з потужністю 6 Вт/м^2 і становить 0,076 мг на 100г продукту.

Ключові слова: кульбаба лікарська (*Taraxacum officinale* Wigg), біологічно активна речовина, аскорбінова кислота, екстракція, ультразвук.

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Оптимизация процесса экстракции биологически активных веществ, выделенных из листьев одуванчика лекарственного (*Taraxacum officinale* Wigg).

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Цель: оптимизировать процесс выделения биологически активных веществ из листьев одуванчика лекарственного. **Методы**: Рассмотрены различные методы выделения биологически активных веществ из одуванчика лекарственного (*Taraxacum oficinale* Wigg.), в том числе обработка ультразвуком, проводившие при мощностях 4, 5 и 6 Вт / м^2 в течение 5, 10, 15, 20, 25 мин. **Результаты**: Получено водные экстракты одуванчика лекарственного (*Taraxacum oficinale* Wigg.) для выделения из них биологически активных веществ, а именно витамина С. Подобраны оптимальные условия для процесса экстрагирования: соотношение сырье:экстрагент 1:20, время экстракции - 30 мин, температура - 25 °С. Экстракцию проводили дистиллированной водой. **Обсуждение:** Доказано, что при экстрагировании из лекарственного растительного сырья происходит диффузия биологически активных веществ из внутренних структур частицы материала. Этот процесс имеет свои особенности. Прежде всего, наличие пористой перегородки, межклеточного пространства и клеточных ходов снижает скорость диффузии. Во-вторых, в поры перегородки могут проникать только те вещества, частицы которых не превышают размеров пор. А также характерно явление десорбции, что наблюдается в клетке после проникновения в нее экстрагента. Определены оптимальные условия извлечения биологически активных веществ из одуванчика лекарственного (*Taraxacum officinale* Wigg): - экстрагент-вода; - соотношение сырье-экстрагент (1:20) - время настаивания 30 мин - температура 25 ° С. Ультразвуковые колебания оптимизируют процесс выделения витамина С из экстрактов листьев одуванчика лекарственного и в результате этого установлено, что наибольший выход аскорбиновой кислоты при обработке экстрактов ультразвуком в течение 25 мин с мощностью 6 Вт / м^2 и составляет 0,076 мг на 100г продукта.

Ключевые слова: одуванчик лекарственный (*Taraxacum officinale* Wigg); биологически активное вещество; аскорбиновая кислота; экстракция; ультразвук.

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