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ASSESSMENT OF THE CONTENT OF BIOLOGICALLY ACTIVE SUBSTANCES IN FRESH AND DRIED RAW MATERIALS OF SWEET BASIL (*OCIMUM BASILICUM* L.)

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In this article the authors conducted a qualitative and quantitative determination of a number of known groups of biologically active substances, such as essential oils, polyphenols and anthocyanins. Qualitative analysis was performed using characteristic qualitative reactions and thin-layer chromatography. Quantitative evaluation of the groups studied was carried out using well-known Pharmacopoeia methods (essential oils, anthocyanins) and the Follin – Chicalteu method, which is widely used in the food industry (polyphenols). The maximum yield of substances is typical for fresh raw materials of Pepper Basil, in which the content of essential oil was 0.82%, polyphenolic compounds – 6.42%, anthocyanins – 0.560%.

Keywords: basil, essential oil, polyphenolic substances, anthocyanins, antibacterial activity, TLC, spectrophotometry, quality factors

Ocimum basilicum L. (Lamiaceae) is a plant with a long history of use as a food and medicinal plant raw material. The description of the plant

is found in the works of Hippocrates, Galen, and Dioscorides.

The ancient Sanskrit medical book of India "Yajur-Veda", written before our era, contains recommendations for the use of basil, the flowers of which are recommended as a diuretic and sedative, the fruit – for gonorrhoea, the roots – for the treatment of intestinal disorders [1]. The great doctor and thinker of the East, Avicenna in "Canon of medical science" separately describes various types of basil – fibrous, hairy, mountain, sweet, garden. Basil is very popular in folk medicine in the Balkans. Herbalists since the time of the Ottoman Empire have recommended the juice of fresh basil leaves to be used for purulent inflammation of the middle ear, and externally – for non-healing wounds [2].

Over 800 years ago Ganjevi Nizami, the famous Azerbaijani scientist and writer, in his works, summarized national data on effect of medicinal plants of the Caucasus, among which basil used as a diuretic, anti-inflammatory and febrifuge, occupies an important place, [3].

For many years, basil has been included in the Russian State Pharmacopoeia, is a pharmacopoeial raw material in France, and is included in the Pharmacopée Française. Currently, in Russia, basil is cultivated as a spice and oilseed raw material and is used in the food and cosmetic industry. However, raw materials and essential oil of basil are widely studied by scientists, primarily as an antibacterial agent. Thus, the presence of high antibacterial activity of *Ocimum basilicum* essential oil against the *S. Aureus* strain was revealed. This study also proved the presence of a synergistic effect when using antibiotics and basil essential oil together [4,5].

In the studies of scientists from South India, the antitumor activity of the essential oil from *Ocimum basilicum* Linn was revealed. Methylthiazole-tetrazolium assay was used for in vitro cytotoxicity screening against human cervical cancer cell line (HeLa), human laryngeal epithelial carcinoma cell line (HEp-2), and mouse embryonic fibroblasts NIH 3T3. As a result of the study, it was proved that basil oil has a powerful cytotoxicity [6].

An interesting area of research is the study of the compositional analysis of basil essential oil, conducted by scientists from different countries with samples of basil introduced in their localities, which revealed the presence of significant differences in the compositional analysis of basil grown in different latitudes [7,8].

It should also be noted that there is a significant chemical variation of basil chemotypes, among which linalool and eugenol varieties are most common [9].

Also in recent years, researchers have been interested in studying the substances of polyphenolic nature and anthocyanin glycosides, the presence of which causes anti-inflammatory and capillary-strengthening activity of ethanol extracts of basil herb [10].

Despite the large number of scientific publications characterizing the chemical

composition of various groups of biologically active substances (BAS) of basil herb and features of the pharmacological action of this raw material, it is important to conduct studies aimed at the choice of criteria of quality of fresh and dried basil herbs for following inclusion into the proposed regulations, because today the basil herb is standardized according to the requirements of GOST R 56562–2015 "Fresh basil – greens. Technical specifications", which includes the definition of quality factors such as the appearance of raw materials, color, smell, taste, mass fraction of defected plants, mass fraction of weeds, the presence of agricultural pests, the presence of mineral impurities and which does not allow to assess the content of biologically active substances and regulate the quality of raw materials.

The purpose of the study is to substantiate methods for identifying the main groups of BAS of basil herb, quantify and evaluate the effect of drying on the content of essential oil, polyphenolic substances and anthocyanins in basil herb.

MATERIALS AND METHODS

As the objects of study, we used fresh basil herb sold in food stores in Moscow that meets the requirements of GOST 56562-2015 "Fresh basil – greens", fresh basil herb of the "Pepper" variety, cultivated in pot culture, as well as dried basil raw material sold as spice by KAMIS, "Home kitchen", "MAGIC TREE" companies (packing – 10 g).

The presence of essential oil in the raw material was determined by steam distillation in accordance with the OFS.1.5.3.0010.15 "Determination of the content of essential oil in medicinal plant raw materials and medicinal plant products".

Qualitative analysis of substances of anthocyanin nature was performed by TLC in the system of solvents "n-butanol – glacial acetic

acid – water" (4:1:2) with detection in the visible range of the spectrum by the method described earlier for identification of anthocyanin pigments [11].

Quantitative determination of polyphenolic substances in basil herb was performed by the Folin-Ciocalteu method, which is widely used for evaluating the quality of food raw materials [12].

Quantitative evaluation of anthocyanin compounds was performed by the method of spectrophotometry [11,13], widely used for the analysis of medicinal plant raw materials. About 1 g (exact weight) of crushed basil herb was transferred to a conical flask provided with a 100 ml ground joint, then, 50 ml of 70% ethyl alcohol containing 1% hydrochloric acid was poured, after which the flask was attached to the return condenser and kept in a boiling water bath for half an hour. At the end of the extraction time, the flask with extract was cooled to room temperature, the missing extractant was added and filtered using a paper filter. 1 ml of filtered extract was placed in a 25 ml volumetric flask and brought to the mark with a 1% solution of hydrochloric acid in 70% ethyl alcohol. The

optical density was measured at wavelength of 538 nm, using 70% ethyl alcohol containing 1% HCl as the reference solution. The content of the sum of anthocyanins in basil herb as a percentage (X) in terms of cyanidin-3-O-glucoside was calculated using the formula:

$$X = \frac{A \times 25 \times 50}{m \times 1 \times 100},$$

where A is the optical density of the test solution, m is the weight of the raw material, g, 100 is the specific absorption rate of cyanidin-3-O-glucoside at 538 Nm in 70% ethyl alcohol containing 1% hydrochloric acid.

RESULTS AND DISCUSSION

The use of laboratory express methods allowed us to identify the content of essential oil in all the samples studied. The complex of traditional qualitative reactions confirmed the presence of tannins. When carrying out a qualitative reaction with lead (II) acetate in the presence of a formed amorphous precipitate, the solution above the precipitate changed to

Table

RESULTS OF DETERMINING THE MAIN GROUPS OF BAS IN BASIL HERB

Study sample	Content of essential oil, %	Content of the sum of polyphenols, %	Content of the sum of anthocyanins, %
Fresh basil herb sold in food stores in Moscow "Home kitchen"	0,72	6,04	0,480
Fresh basil herb of the "Pepper" variety, cultivated in pot culture	0,82	6,42	0,560
Dried raw basil sold as a spice by KAMIS	0,43	4,25	0,039
Dried raw basil sold as a spice by "Home kitchen" company	0,39	3,89	0,064
Dried raw basil sold as a spice by "MAGIC TREE" company	0,47	4,76	0,053

a pronounced pink color, which indicates the presence of anthocyanins, identified by TLC. Chromatograms of extracts obtained from basil leaves show pink spots with values of Rf-0.51, Rf-0.46, Rf – 0.36 (dominant spot), Rf – 0.31 and Rf – 0.21. Comparison with the Rf standard identified cyanidin-3-O-glucoside (Rf-0.36) (standard sample of cyanidin-3-O-glucoside of Xian Le Sen Bio-Technology Co).

The results of quantitative determination of essential oil, the total content of substances of polyphenolic nature and anthocyanins are presented in the table.

Taking into account the results obtained, we found that the maximum content of all BAS groups is typical for fresh basil herb raw

materials, along with this the pot basil culture of the "Pepper" variety shows the highest content for all the studied groups, which can be explained by the fact that the raw materials for analysis were cut off immediately before the experiment, while fresh basil raw materials sold in food stores were cut off a day and more earlier. Raw basil sold as a spice was dried in the conditions indicated on the package, which suggests the need for further studies aimed at studying the effect of the drying conditions on the yield of biologically active substances. The greatest losses when drying basil are observed among the class of anthocyanins.

As it is known, anthocyanins are representatives of a class of polyphenolic secondary

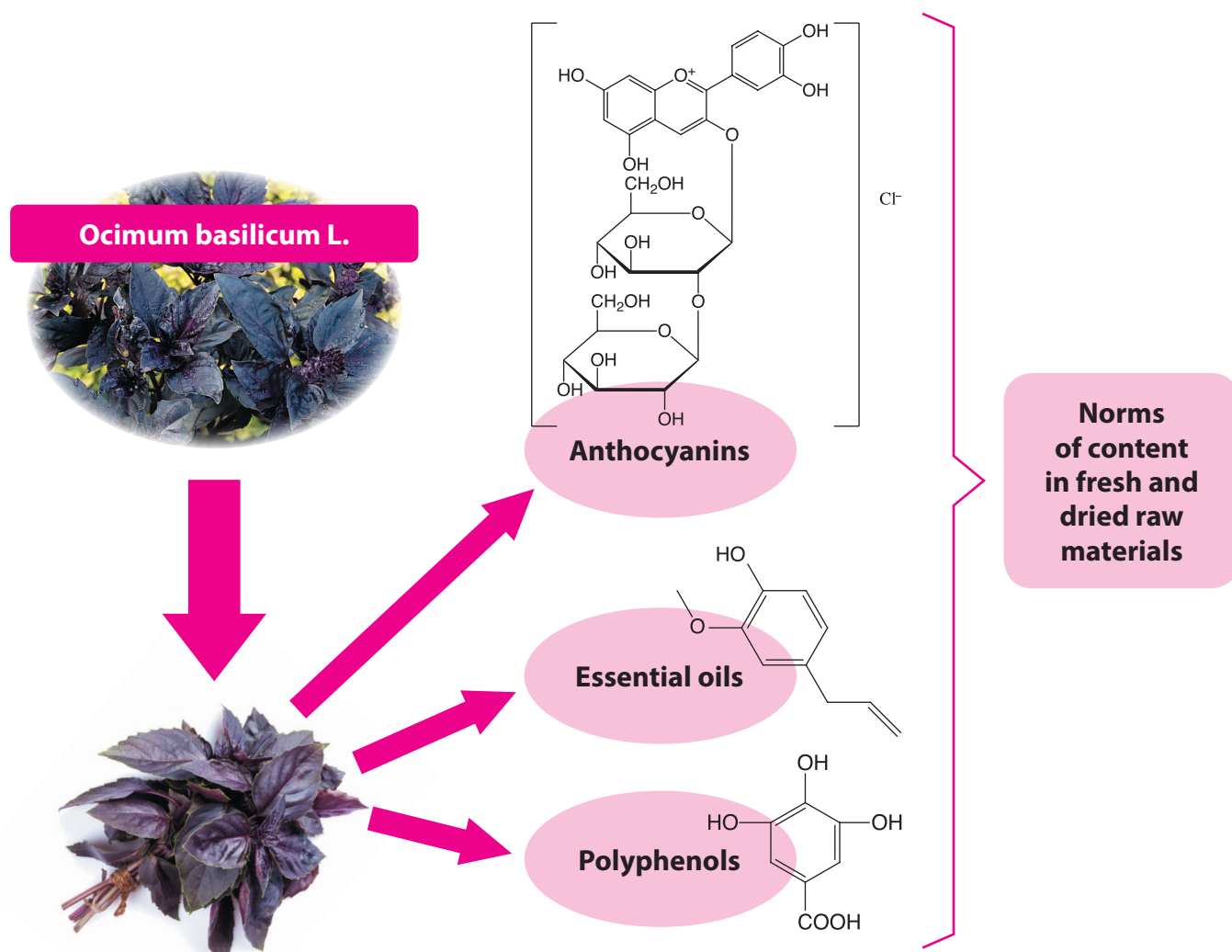


FIG. Selection of BAS groups for the development of standards for the content of BAS in *Ocimum basilicum L.* fresh and dried raw materials for inclusion into the developed regulatory documentation

metabolites-flavonoids, which are widely distributed in the plant world. Under the conditions of modern in vitro and in vivo analysis, the valuable pharmacological properties which are characteristic for anthocyanins, have been identified. The scientific literature often shows the protective function of anthocyanins in the development of oxidative stress, hypoglycemic and hypolipidemic activity, anti-inflammatory effect, the ability to inhibit lipid peroxidation, reducing the permeability and fragility of capillaries.

The need to use fresh basil raw materials as a source of anthocyanins is due to the fact that during the drying of basil leaves, the processes of hydrolysis and polymerization of anthocyanin pigments are carried out, which results in the accumulation of polymeric anthocyanin structures characterized by low bioavailability and absence of pharmacological effects inherent in anthocyanins.

Thus, the quantitative determination of the content of essential oil, polyphenol compounds, and anthocyanins in the raw materials of basil was carried out, on the basis of which it is planned to develop standards for the content of these BASs, according to Fig.

ONCLUSION

During the experiment, the authors determined the content of essential oil, polyphenolic compounds and anthocyanins in fresh and dried basil herb. The identification of these BAS groups was carried out by qualitative reactions and TLC, the Pharmacopoeia method of steam distillation was used for the quantitative determination of essential oil, the polyphenol fraction was determined by the Folin – Chocalteu method, widely used in the food industry, and anthocyanins were determined spectrophotometrically. The maximum yield of substances is typical for fresh raw material

of basil of "Pepper" variety in which the content of essential oil was 0.82%, polyphenolic compounds – 6.42%, anthocyanins – 0.560%.

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