

The Effect of Technological Extraction Parameters on the Content of Flavonoids and Extractive Substances in Extracts of an Antidiabetic Herbal Mixture

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	Abstract The results of experiments studying the influence of technological extraction parameters (temperature, alcohol concentration, time, and hydromodule) on the yield of total flavonoids and extractive substances from an antidiabetic herbal mixture composed of bean pods, fig leaves, chicory roots, and goat's rue herb are presented. Based on the experimental results, it can be concluded that there is a direct relationship between the technological parameters of the extraction process and the amounts of total flavonoids and extractive substances in the extracts. Under optimal extraction conditions, the total flavonoid yield was about 1.10%, while the yield of total extractive substances was around 15.0%. The quantitative determination of total flavonoids in the extracts was performed spectrophotometrically, recalculated as rutin using aluminum chloride. Keywords: antidiabetic herbal mixture, extraction, maceration, total flavonoids, antioxidant, glycemic, extractive substances, technological parameters, process.
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Introduction

Among natural biologically active compounds, a special place is occupied by the derivatives of phenylbenzopyrone—flavonoids—found in higher plants. They represent one of the most numerous and well-studied groups of natural polyphenolic compounds, which play an important role in plant metabolism and possess a wide range of biological activities. Their study has a history of more than a century. The first compounds were isolated at the end of the 19th and the beginning of the 20th centuries. However, only with the development of organic chemistry and various

instrumental methods (chromatographic and spectroscopic) did it become possible to systematically study their structure, functions, and pharmacological potential [1].

The necessity of studying flavonoids is determined by several factors.

Firstly, they are extremely widespread in the plant world. They are found in leaves, flowers, fruits, seeds, and bark in more than half of the known species of higher plants.

Secondly, flavonoids enter the human body in significant quantities with food as components of vegetables, fruits, grains, and beverages, thereby forming a whole class of nutrients.

Thirdly, numerous studies of recent decades have demonstrated their diverse physiological effects. They possess antioxidant, anti-inflammatory, cardioprotective, antidiabetic, antitumor, and antimicrobial activities [2,3].

From a chemical point of view, flavonoids, as noted above, are derivatives of benzopyran with a general C₆–C₃–C₆ skeleton, including two aromatic rings and a heterocyclic fragment. The variety of structural modifications—hydroxylation, methoxylation, glycosylation, and acylation—determines the enormous chemical and functional diversity of this class of compounds. The presence of specific functional groups and the spatial configuration of the molecule determine both antioxidant potential and interactions with enzymatic targets in the body.

The pharmacological significance of flavonoids extends far beyond dietology. In recent years, they have been considered promising molecules for the development of pharmaceuticals and biologically active supplements. The potential applications of flavonoids are seen in the prevention and treatment of chronic noncommunicable diseases—cardiovascular, neurodegenerative, endocrine, and oncological. At the same time, a number of unresolved problems must be taken into account, such as variability of their content in plant raw materials, low bioavailability, insufficient clinical trials, and technological challenges of their extraction and purification.

This study examines the influence of technological extraction parameters on the yield of flavonoids from an antidiabetic herbal mixture consisting of bean pods (40%), fig leaves (20%), chicory roots (20%), and goat's rue herb (20%) [4]. It is known that type 2 diabetes remains one of the main causes of premature mortality and disability, which stimulates the search for safe phytocomplexes with a multifaceted mechanism of action. The mixture we developed takes into account the diversity of plant organs and spectra of bioactive compounds: flavonoids and phenolic acids (antioxidant, anti-inflammatory, and enzyme-inhibiting effects), inulin (prebiotic and glycemic effect), and alkaloids of the galegine type (historically

associated with prototypes of biguanides). For the technological development of the extraction process, aqueous extraction was excluded, and a hydroalcoholic system was selected as more effective for extracting polyphenolic compounds and low-molecular-weight metabolites of medium polarity [5,6,7,8].

Purpose of the Study

To develop a technology for obtaining an extract from the antidiabetic herbal mixture using hydroalcoholic extraction. The effectiveness of the developed technology was evaluated by the yield of total flavonoids and extractive substances.

Materials and Methods

The preselected components of the mixture, meeting the requirements of regulatory documents and having a residual moisture content of less than 12%, were ground. For the experiments, the aerial parts of the herbs and leaves were ground to particle sizes of 3–5 mm, chicory roots to 2–3 mm, and bean pods to 5–7 mm. For extraction, a mixture of the ground plants in the amount of 30.0 g was taken in the component ratios indicated above and thoroughly mixed.

For the extraction of biologically active substances, hydroalcoholic solutions of various ethanol concentrations (20%, 30%, 40%, and 50%) were used. Based on experience from other processes, the hydromodule was set at 1:10. Extraction was carried out for 24 hours at a temperature of 40 °C. Maceration was performed three times with periodic stirring. The combined extracts were filtered and evaporated to concentrate them. A portion of the extract was evaporated to dryness to determine the amount of extractive substances.

The quantitative determination of total flavonoid content in the extracts was performed spectrophotometrically using aluminum chloride and recalculated as rutin, while the extractive substances were determined by drying to constant weight. The experiment was carried out in triplicate.

Results and Discussion

The quantitative analysis showed that the amount of flavonoids and extractive substances in the extracts varied significantly and depended not only on the technological parameters of the extraction process but also on the concentration of the hydroalcoholic mixture. Data analysis (Table 1) revealed a clear dependence of the yield of biologically active substances on the ethanol concentration in the extractant. The lowest values of total flavonoids (0.59%) and extractive substances

(12.05%) were recorded when 20% ethanol was used for extraction. A gradual increase in alcohol concentration to 25%, 30%, and 35% led to higher yields of both total flavonoids and extractive substances, which is apparently associated with increased solubility of phenolic compounds of medium polarity.

Table 1. Effect of alcohol concentration and temperature on total flavonoid and extractive yields (n = 3)

№	Ethanol, %	Total flavonoids, %	Total extractive substances (w/w),, %
1	20	0,56±0,02	12,05±0,03
2	25	0,59±0,03	12,11±0,04
3	30	1,07±0,03	12,75±0,05
4	35	1,12±0,05	13,07±0,04
5	40	1,19±0,04	14,85±0,05

At an alcohol concentration of 40%, the maximum yield of total flavonoids and extractive substances was observed. Under these conditions, the yield of extractive substances was 14.85%, and the yield of total flavonoids was 1.09%. With a further increase in alcohol concentration to 45%, 50%, and 55%, a gradual decrease in extract yield was observed, which may be associated with the reduced solubility of certain polar components (such as inulin and hydrophilic phenolic acids). Based on the experimental results, 40% ethanol can be considered the most optimal for extracting both polar and moderately lipophilic compounds.

In parallel, the effect of temperature on the efficiency of the extraction process was studied. According to the experimental data, the most optimal temperature was found to be 40 °C. At this temperature, the yield of total flavonoids was 1.07%, and the yield of extractive substances was 15.03%. At lower temperatures, the diffusion rate was apparently much slower, while at higher temperatures (above 45 °C), the yields began to decline.

Table 2. Results of the study on the effect of extraction temperature on the yield of total flavonoids and extractive substances (ethanol 40%, 1:10)

№	Temperature, °C	Total flavonoids, %	Total extractive substances, %
1	20	0,83±0,03	12,95±0,5
2	30	0,94±0,05	14,5±0,5
3	40	1,07±0,02	15,03±0,50
4	50	1,04±0,03	15,01±0,5

It was experimentally established that with an increase in maceration time, the yield of biologically active substances increased almost linearly, reaching its maximum at 24 hours. A further increase in extraction time did not lead to a significant rise in the yield of total flavonoids and extractive substances.

The study of the effect of the hydromodule on the yield of biologically active substances showed that up to a hydromodule value of 1:10, there was a significant increase in the yield of both total flavonoids and total extractive substances.

Table 3. Influence of the process hydromodule on the yield of total flavonoids and extractive substances (ethanol 40%, temperature 40 °C, n = 3)

№	Hydromodule	Total flavonoids, %	Total extractive substances, %
1	1:5	1,01±0,03	12,7±0,04
2	1:10	1,09±0,04	15,01±0,03
3	1:15	1,10±0,03	15,02±0,04
4	1:20	1,11±0,04	15,02±0,03

With a further increase in the hydromodule to a ratio of 1:20, a gradual but insignificant increase in the content of biologically active substances (BAS) in the extract was observed. However, the large volume of solvent required, along with the need for subsequent evaporation, increased energy consumption and led to solvent losses, making the extraction process uneconomical and inefficient. Therefore, in our opinion, the most optimal hydromodule is 1:10.

A comprehensive analysis of the influence of various technological factors (ethanol concentration, extraction temperature, duration and frequency of maceration, and hydromodule) demonstrated that the optimal extraction conditions are as follows:

- Solvent: 40% hydroalcoholic mixture,
- Temperature: 40°C,
- Hydromodule: 1:10,
- Maceration time: 24 hours,
- Number of extractions: two sequential macerations.

Conclusion

Based on the results of the conducted studies, a technology has been developed for obtaining an extract from an antidiabetic herbal mixture consisting of bean pods (40%), fig leaves (20%), chicory roots (20%), and goat's rue herb (20%). The extracts were obtained using a hydroalcoholic mixture. The efficiency of the extraction process was evaluated by determining the content of total flavonoids and extractive substances in the extracts.

It was experimentally established that the optimal technological parameters of the extraction process are as follows: extractant – 40% ethanol; hydromodule – 1:10; temperature – 40 °C; maceration duration – 24 hours; and number of extractions – 2.

A direct relationship between the technological parameters of the extraction process and the yield of total flavonoids in the obtained extracts has been demonstrated.

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