

## Phytochemical study of a plant composition with hypoglycemic action

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## Phytochemical study of plant compositions with hypoglycemic action

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### RESUME

A study of qualitative and quantitative composition of biologically active substances of plant composition having hypoglycemic action was done. With the use of thin-layer chromatography we established the presence of quercetin, caffeic acid, fructose. Spectrophotometrically quantified flavonoid content in terms of quercetin ( $0.66 \pm 0.02\%$ ), phenol carbonic acids in terms of caffeic acid ( $1.39 \pm 0.07\%$ ), low and high fructosan and fructose ( $2.56 + 0, 11\%$ ), free fructose ( $0.10 \pm 0.01\%$ ) and polysaccharides by gravimetric method ( $7.80 + 0.38\%$ ).

The results can be used to develop normative documents on plant composition having hypoglycemic action and preparations obtained based on it.

Keywords: diabetes, herbal medicine, botanical products standardization.

### SUMMARY

The study of the qualitative and quantitative composition of biologically active substances of a plant composition with a hypoglycemic effect has been carried out. Using thin layer chromatography methods, the presence of quercetin, caffeic acid, fructose was established. Spectrophotometric method was used to determine the quantitative content of flavonoids in terms of quercetin ( $0.66 \pm 0.02\%$ ), phenol carboxylic acids in terms of caffeic acid ( $1.39 \pm 0.07\%$ ), low- and high-molecular fructosans and fructose ( $2.56 \pm 0.11\%$ ), free fructose ( $0.10 \pm 0.01\%$ ) and the gravimetric method of polysaccharide content ( $7.80 \pm 0.38\%$ ).

The results obtained can be used to develop regulatory documents for a plant composition with a hypoglycemic effect and preparations based on it.

Key words: diabetes mellitus, herbal medicine, herbal remedies, standardization.

### Introduction

According to the World Health Organization, 347 million people worldwide suffer from diabetes. 3.4 million people die from complications of the disease every year. The most common is type 2 diabetes mellitus associated with the development of relative insulin deficiency and accounting for 90% of all cases of the disease [1].

Type II diabetes mellitus is especially dangerous because its symptoms in most cases are much less pronounced than that of type 1 diabetes, as a result of which the disease is most often diagnosed after the development of complications in the later stages [2].

As a prophylaxis and in the early stages of development of type 2 diabetes mellitus, diet therapy and phytotherapeutic agents are mainly used [1]. On the Russian pharmaceutical market there are such hypoglycemic preparations as Arfazetin, Mirfazin, Sakharonorm, as well as antidiabetic teas and drinks. Currently, it is advisable to expand the range of herbal medicines with hypoglycemic action.

At the Department of Pharmacognosy with a Course of Botany and Fundamentals of Phytotherapy, SBEI HPE BSMU, a herbal collection has been proposed, including 7 components, which, according to the literature, have both direct and indirect hypoglycemic effects.

The effectiveness of the proposed herbal composition was studied in an experiment on rats with streptozocin-induced diabetes mellitus at the Institute of Organic Chemistry, Ufa Scientific Center of the Russian Academy of Sciences (Ufa). The course of experimental diabetes was monitored by an increase in the level of glucose and alpha-amylase in the blood, as well as the physical condition of the animals (decrease in body weight, increase in consumption

water and urine output). The death of rat pups was observed in all groups of animals with induced diabetes. By the seventh day of the experiment, all animals in the control group died, while the survival rate in the main group was 58.3%. By the 14th day, the level of alpha-amylase and glucose in the blood of animals of the main group decreased to normal, which allows us to conclude about the effectiveness of the developed plant composition [3].

The aim of this study is a phytochemical study of a plant composition with a hypoglycemic effect.

#### Methods

Samples of medicinal plants for phytochemical analysis were collected in typical habitats in various regions of the Republic of Bashkortostan (2010 - 2012). Raw materials were stored in accordance with the requirements of regulatory documents (OST 64-4-143-75 and GF USSR XI ed.) At room temperature, in a dry, well-ventilated room, not infected with barn pests, without direct sunlight.

Qualitative detection of flavonoids and phenolcarboxylic acids was carried out by ascending chromatography in a thin layer of sorbent (TLC) - on plates "Silufol UV-254" (Czech Republic).

The following solvent systems were used in the experiment: 1)

- 1) butanol - acetic acid - water (4: 1: 5);
- 2) chloroform - ethanol - water (26: 20.3: 3);
- 3) chloroform - acetic acid - water (10: 6.5: 1);
- 4) ethyl acetate - formic acid - water (8: 1: 1);
- 5) ethyl acetate - formic acid - water (10: 2.5: 3);
- 6) ethyl acetate - methanol - water (100: 17: 13);
- 7) toluene - ethyl acetate - water (4: 4: 2).

For the development of chromatograms, a 3% solution of aluminum chloride was used.

Substances were detected by viewing chromatograms in visible and UV light, before and after development. For this, a mercury-quartz lamp with a light filter at a wavelength of 365 nm was used.

For identification, we used available authentic samples of flavonoid substances (rutin, quercetin), phenol carboxylic acids (chlorogenic, gallic, caffeic).

The detection of fructose and fructosans by ascending thin layer chromatography was carried out in the ethyl acetate - methanol - water system (100: 17: 13). Fructose was used as a standard substance.

To develop spots, chromatograms were treated from a spray bottle with 0.1% alcohol solution of resorcinol, allowed to dry, and placed for 5–7 minutes over a Petri dish with concentrated hydrochloric acid poured onto the bottom of the container with a layer of 3 mm. Viewed in visible light.

The study of UV spectra of extracts from the plant composition, hypoglycemic action ~~possessing~~ carried out on a spectrophotometer brand "Schimadzu 1800" at analytically significant wavelength intervals - 240-270 nm; 310-450 nm.

The spectrophotometric method was used to determine the quantitative content of the sum of flavonoids in terms of quercetin [4], the amount of fructosans and fructose in terms of inulin [5, 6], phenol carboxylic acids, in terms of caffeic acid [7].

The measurements were carried out using a Schimadzu 1800 spectrophotometer.

The determination of the sum of polysaccharides was carried out according to the method described in article No. 20 "Folia Plantaginis majoris. Leaves of the big plantain" GF XI ed. Part 2 [8].

Statistical processing of the experimental data (P = 95%) was carried out using the Student's test with the calculation of the boundary values of the confidence interval of the mean result and the determination of the error of a single determination.

#### results

For chromatographic studies, a sample of raw materials was extracted with 70% ethanol in a 1: 1 ratio. The resulting extract was concentrated and used for analysis.

The best separation of the total BAS was observed in the ethyl acetate - methanol - water system (100: 17: 13). 10 spots were observed on the chromatogram (Table 1).

The presence of quercetin and caffeic acid was established by chromatographic behavior and in comparison with witnesses.

The presence of fructosans and fructose was confirmed by TLC. When the obtained chromatogram was viewed in visible light, a spot of dirty crimson light with an R<sub>f</sub> of 0.15 was observed, the value of which coincided with the fructose standard.

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As a result of a phytochemical study of the plant composition using the above methods, the content of some groups of biologically active substances was determined and it was found that polysaccharides and phenol carboxylic acids are predominant (Table 2).

#### Discussion and conclusions

Thus, the study of the qualitative and quantitative composition of biologically active substances of the plant composition with hypoglycemic action was carried out. The presence of quercetin, caffeic acid, fructose was established. The quantitative content of some groups of biologically active substances has been determined.

Table 1

Chromatographic characteristics of biologically active substances 70% ethanol extract from a herbal composition with hypoglycemic action

№ п/п	R <sub>f</sub>	Окраска в видимом свете		Окраска в УФ-свете	
		до обр. AlCl <sub>3</sub>	после обр. AlCl <sub>3</sub>	до обр. AlCl <sub>3</sub>	после обр. AlCl <sub>3</sub>
1	0,99	Бледно-зеленая	Бледно-зеленая	оранжевая	оранжевая
2	0,94	–	–	красная	красная
3	0,84	–	желтоватая	бледно-зеленовато-желтая	ярко-зеленовато-желтая
4	0,76	–	–	голубая	голубая
5	0,70	–	–	зеленая	зеленая
6	0,55	–	желтая	бледно-желтая	ярко-желтая
7	0,42	–	–	голубовато-зеленая	голубовато-зеленая
8	0,37	–	–	голубая	голубая
9	0,19	–	–	зеленая	зеленая
10	0,07	–	–	бело-голубая	бело-голубая

table 2

The quantitative content of biologically active substances in the collection of hypoglycemic

BAV group	Content, %
Fructosans are low and high molecular weight and fructose	2.56 ± 0.11
Free fructose	0.10 ± 0.01
Polysaccharides	7.80 ± 0.38
Flavonoids in terms of quercetin	0.66 ± 0.02
Phenol carboxylic acids	1.39 ± 0.07

The results obtained can be used to develop regulatory documents for a plant composition with a hypoglycemic effect and preparations based on it.

#### Literature

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