

Study of phenolic compounds of herb and meal of Wormwood
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The study of phenolic compounds of herb and schrot of wormwood
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RESUME

Analysis of quantity and quality portions of phenolic components of herb and schrot of wormwood was done using HPLC method. The water-spirit fraction of herb contains 20 substances, 15 of them were identified. The water-spirit extract of schrot contains 13 substances and 9 of them were identified.

According to differential spectrophotometry data quantitative content of flavonoids in the raw material in terms of rutin was 0.44% and 0.14% for the herb and schrot wormwood respectively.

Keywords: wormwood, wormwood schrot, HPLC, phenolic components, spectrophotometry, flavonoids.

SUMMARY

The HPLC method was used to analyze the qualitative content and quantitative ratio of phenolic compounds of grass and wormwood meal. 20 substances were found in the alcohol-water fraction of wormwood herb, of which 15 were identified. 13 substances were found in the alcohol-water extraction of wormwood meal, of which 9 compounds were identified.

According to differential spectrophotometry, the quantitative content of the sum of flavonoids in the raw material in terms of rutin was 0.44% and 0.14% for grass and wormwood meal, respectively.

Key words: wormwood herb, wormwood meal, HPLC, phenolic compounds, spectrophotomerism, flavonoids.

Introduction

Expanding the range of domestic pharmaceuticals is one of the pressing problems of modern medicine. In our opinion, in addition to the introduction of new plants that are not used in official medicine, a rational solution to this problem can be the resource-saving use of waste from the industrial processing of medicinal plant raw materials.

In this regard, the purpose of this work was a comparative study of the components of the polyphenolic fraction of the aboveground part of wormwood and its meal, which remained after receiving the tincture of wormwood.

Materials and research methods

The analysis of compounds of the polyphenol fraction was carried out on a high-performance liquid chromatograph (GILSTON), model 305 (France), manual injector, model RHEODYNE 7125 USA, followed by computer processing of the research results using the Multichrom for Windows program. The stationary phase was Kromasil C 18 sorbent, particle size 5 microns, metal column 4.6 × 250 mm. The mobile phase is a mixture of methanol - water - phosphoric acid (conc.) In a ratio of 40: 60: 0.5. The analysis was carried out at room temperature for 70 minutes. The flow rate of the eluent is 0.8 ml / min. The detection of substances was carried out using a UV detector "GILSTON" UV / VIS, model 151, at a wavelength of 254 nm.

For research, grass and meal were crushed to a particle size passing through a sieve with a hole diameter of 2 mm (GOST 214-83). Weighed portions of grass and wormwood meal (10.0 g) were placed in flasks with a capacity of 200.0 ml, 60 ml of 70% ethyl alcohol were added,

to reflux condensers and heated in a boiling water bath for 1 hour from the moment the alcohol-water mixture boiled in flasks. After cooling, the mixtures were filtered through paper filters into volumetric flasks with a volume of 100.0 ml and the volumes of the extracts were brought to the mark with 70% ethyl alcohol (test solutions).

At the same time, a series of 0.05% solutions of standard samples (GSO) in 70% ethyl alcohol was prepared: rutin, quercetin, luteolin, luteolin-7-glycoside, hesperidin, apigenin, hyperoside, dihydroquercetin, kaempferol, vitexin, isovitexin, naringenin, bainetoram, gallic, coffee, chlorogenic, chicory, cinnamic, ferulic, ellagic, okumaric acids, umbelliferone, esculetin, coumarin, methoxycoumarin, epigallocatechin gallate, epicatechin. 50 μ L of the test solutions and GSO were introduced into the chromatograph and chromatographed according to the above method [2, 4]. The quantitative ratio of substances of polyphenolic nature in the studied extracts was established by the method of normalization according to the areas of chromatographic peaks [3].

The quantitative determination of the sum of flavonoids in the alcohol-water extract of wormwood herb and its meal was carried out by the spectrophotometric method [1, 5]. We chose the reaction of the interaction of a complex of flavonoids with aluminum chloride in a medium of 70% alcohol as the basis for the analysis. The concentration of flavonoid compounds was determined in terms of GSO rutin.

The content of the sum of flavonoids in the herb and meal of wormwood in terms of rutin and absolutely dry raw materials in percent (X) was calculated by the formula:

$$X = \frac{D \times m_0 \times V_{k1} \times V_{k2} \times V_{n0} \times 100 \times 100}{D_0 \times m \times V_{k3} \times V_{k4} \times V_{n1} \times (100 - W)}$$

where D is the optical density of the test solution; D₀ - optical density of GSO routine; m is the weight of the sample of raw materials in grams; m₀ - weight of sample of GSO routine in grams; W - loss in weight on drying in percent; V_{k1} - the volume of the extractant for extracting flavonoids from raw materials; V_{k2} - the volume of the flask for diluting the extract; V_{k3} - the volume of the flask for the 1st dilution of the GSO rutin; V_{k4} - the volume of the flask for the 2nd dilution of the GSO rutin; V_{n1} - the volume of the pipette (the volume of the extract from the herb and wormwood meal taken for breeding); V_{n0} - the volume of the pipette (the volume of the GSO rutin solution was taken for dilution).

Research results and their discussion

Based on the analysis of polyphenolic compounds of grass and wormwood meal by HPLC, it was found that the alcohol-water fraction of the studied raw materials contains from 13 (meal) to 20 (grass) substances (flavonoid compounds, coumarins and phenolcarboxylic acids) (Tables 1, 2).

Table 1

Results of the study of phenolic compounds by HPLC in the grass of wormwood

№	Время удерживания, сек	Количественное соотношение, %	Наименование вещества
1	176,46	4,80	Таннин
2	189,24	12,26	Галловая кислота
3	217,08	1,96	Неидентифиц
4	228,30	3,60	Неидентифиц
5	244,20	7,63	Эпигаллокатехин галлат
6	307,80	17,20	Хлорогеновая кислота
7	399,78	6,24	Кофейная кислота
8	479,10	6,32	Феруловая кислота
9	536,52	1,82	Дигидрокумарин
10	572,94	3,08	Неохлорогеновая кислота
11	680,40	4,60	Эпикатехин
12	882,60	6,18	Гесперидин
13	961,20	4,12	Гиперозид
14	1084,20	2,59	Рутин
15	1217,40	1,95	Неидентифиц
16	1417,80	5,57	Коричная кислота
17	1840,20	4,73	О-метоксикумарин
18	2103,60	0,97	Неидентифиц
19	2430,60	2,04	Катехин
20	2707,20	2,36	Неидентифиц

As can be seen from the data in Tables 1 and 2, the polyphenolic fractions of the studied types of raw materials differ in their qualitative composition. The composition of the polyphenolic fraction of the herb wormwood is more varied than the wormwood meal. In the herb of wormwood, the presence of tannin, epigallocatechin gallate, epicatechin, catechin, dihydrocoumarin and hyperoside was found to be absent or in minimal amounts in the meal. In the meal and grass wormwood, gallic and chlorogenic acids predominate from phenol carboxylic acids, and hesperidin and rutin from flavonoids.

Based on the main criterion - the maximum content of the sum of flavonoids in raw materials and meal in terms of rutin, optimal technological parameters of extraction were selected (type of extractant, its concentration, hydromodule, degree of grinding of raw materials, extraction time and temperature) (Table 3).

table 2

Results of the study of phenolic compounds by HPLC in wormwood meal bitter

№	Время удерживания, сек	Количественное соотношение, %	Наименование вещества
1	190,80	22,54	Галловая кислота
2	202,90	30,11	Неидентифиц
3	310,50	10,12	Хлорогеновая кислота
4	384,20	4,90	Кофейная кислота
5	424,50	3,42	Неидентифиц
6	475,90	6,51	Феруловая кислота
7	579,60	10,87	Неохлорогеновая кислота
8	887,10	3,45	Гесперидин
9	1076,00	2,42	Рутин
10	1245,00	1,70	Неидентифиц
11	1366,00	0,87	Неидентифиц
12	1403,00	2,64	Коричная кислота
13	1813,00	0,45	О-метоксикумарин

Table 3

Conditions for obtaining extracts of herbs and wormwood meal

Польнь горькая	Кратность экстракции	Степень измельчения, мм	Вид экстрагента	Температура, °С	Гидромодуль	Время экстракции, мин
Трава	1	1,0	70 % этанол	100	1:50	30
Шрот	1	1,0	70 % этанол	100	1:10	60

When studying the optimal conditions for the photometric reaction, it was found that the maximum optical density is achieved using 2 ml of a 2% solution of aluminum chloride in 70% ethanol. Stable coloration of solutions of extracts with aluminum chloride occurs after 30 minutes. and lasts for 60 minutes, which is sufficient for analysis.

According to spectrophotometric analysis, the quantitative content of the sum of flavonoids in the raw material in terms of rutin was 0.44% and 0.14% for grass and wormwood meal, respectively. Based on the studies carried out, a spectrophotometric method for the quantitative determination of flavonoids in the herb and meal of wormwood was developed. The proposed technique is reproducible, available and express.

Having analyzed in comparison the composition and quantitative content of the flavonoids of the alcohol-water fraction of the wormwood meal and the wormwood herb, we can conclude that the extraction of flavonoid compounds from the wormwood herb is sufficient in the production of tinctures from it. Consequently, further study of meal in the development of complex use of raw materials is an urgent task for other classes of biologically active compounds.

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