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The development of new approaches of standardization of the species "the Siberian swallow"

Kurkin Vladimir ¹, Shmygareva Anna ^{2*}

1. Samara State Medical University, Samara, Russia.
[Email: Kurkinvladimir@yandex.ru]
2. Orenburg State Medical Academy, Orenburg, Russia.
[Email: a.shmygareva@mail.ru, Tel: +79123401517]

The methodological approaches for standardization of the species "the Siberian swallow" were developed on the base of results of chemical investigation of *Frangula alnus* Mill. barks. These approaches consist in the determination of anthracen derivatives by means of spectrophotometry at analytical wavelength 524 nm and using of frangulin A as standard sample. The contents of the total anthracen derivatives in species "the Siberian swallow" are varied with 3,50 % to 3,63% (calculated on frangulin A).

Keyword: The species "the Siberian swallow", *Frangula alnus* Mill., anthracen derivatives, frangulin A, standardization, spectrophotometry.

1. Introduction

It is so hard to keep a balanced diet for the human in the XXI century, this leads to disruption of the gastrointestinal tract. Today in the correction of functional disorders of the digestive system are widely used herbal laxatives drugs. Species "the Siberian swallow" is popular like a mild laxative ^[1, 2]. "The Siberian swallow" includes Frangulae cortex, Sennae folia, Taraxaci radices, Calendulae Flores, Chamomillae Flores, Menthae piperitae folia, Foeniculi fructus, Millefolii herba, Hiperici herba, Filipendulae herba ^[3, 4, 5, 6, 7].

Known, that laxative effect due to presence anthracen derivatives contained in Frangulae cortex and Sennae folia ^[8]. At present the methods of quantitative determination of anthracen derivatives in the species "the Siberian swallow" was not develop.

2. Materials and methods

The object of the study served the raw materials of the industrial design of "the Siberian swallow" ("OOO Altaiskii kedr") (2011, 2012 and 2013), Sennae folia ("OAO Krasnogorskleksredstva")

(2012) and Frangulae cortex (OOO PKF Phytopharm (2012).

Electronic spectra were measured on the spectrophotometers "Specord 40" (Analytik Jena) and "UNICO -2800" in cells with thickness of a layer 10 mm.

3. Results and Discussion

Previously, the purpose of a substantiation of methodical approaches to standardization the bark of *Frangulae alnus* conducted a study on the isolation of substances from raw materials of this plant ^[9, 10]. It was found that the dominant components are 6-O- α -L-rhamnopyranoside of Frangula-emodin (frangulin A) and 6-O- β -O-apiofuranoside of Frangula-emodin (frangulin B), and was justified use in the methods of analysis of bark of *Frangulae alnus* fragolina A as the standard substance ^[9, 10]. During the development of methods of quantitative determination of the amount of anthracen derivatives in "the Siberian swallow" studied UV spectra of water-alcohol extraction from this species, of water-alcohol extraction from the bark of *Frangulae alnus*,

and also solution of frangulina A. In addition, also studied in comparative terms, the water-alcohol extraction from the leaf of *Senna alexandria*, part of "the Siberian swallow".

The study UV spectra of water-alcohol extraction from "the Siberian swallow" showed (Figure 1) that the characteristic is the presence of maximum absorption at 269 ± 2 nm. A comparative study of UV spectra of initial water-alcohol extraction from the bark of *Frangula alnus* and leaf of *Senna alexandria* suggests (Figure 2 and 3) that the substance of both raw materials largely determine the nature of absorption curve of water-alcohol extraction from "the Siberian swallow" (Figure 1). In the study of alkaline-ammoniac solution of water-alcohol extraction from "the Siberian swallow" in the electron spectrum is found characteristic absorption maximum at length 524 ± 2 nm (Figure 1) A comparative study of UV spectra of alkali-ammonia solutions of water-alcohol extraction from the bark of *Frangulae alnus* and leaf of *Senna alexandria* showed (Figure 2 and 3) that in the case of both types of raw materials takes place contribution to the absorption curve of the substances investigated types of raw materials in

the curve of absorption alkali-ammoniac solution of water-alcohol extraction from "the Siberian swallow" (Figure 1), but the greatest extent correlation is observed with alkali-ammoniac solution of water-alcohol extraction from the bark of *Frangula alnus* (Figure 2). We have shown previously, one characteristic of anthracen derivatives of the *Frangula alnus* barks is frangulin A [9, 10]. In this regard, we conducted a survey of solution of frangulin A, it is shown that in the UV spectrum of alkali-ammoniac solution of this substance is also present characteristic absorption maximum at length 524 ± 2 nm (Figure 4). This gives grounds, conversion of the content of the amount of anthracen derivatives in the species of "the Siberian swallow" implement on frangulin A. Therefore, as in the case of the bark of *Frangulae alnus* [9], the analytical wavelength can be used is 524 nm, and a standard pattern can serve as the dominant anthraglycoside -frangulin A, moreover, in the absence of this standard in the calculation formula can be used theoretical value of the specific absorption index ($E_{1\text{cm}}^{1\%}$) - 180 [9].

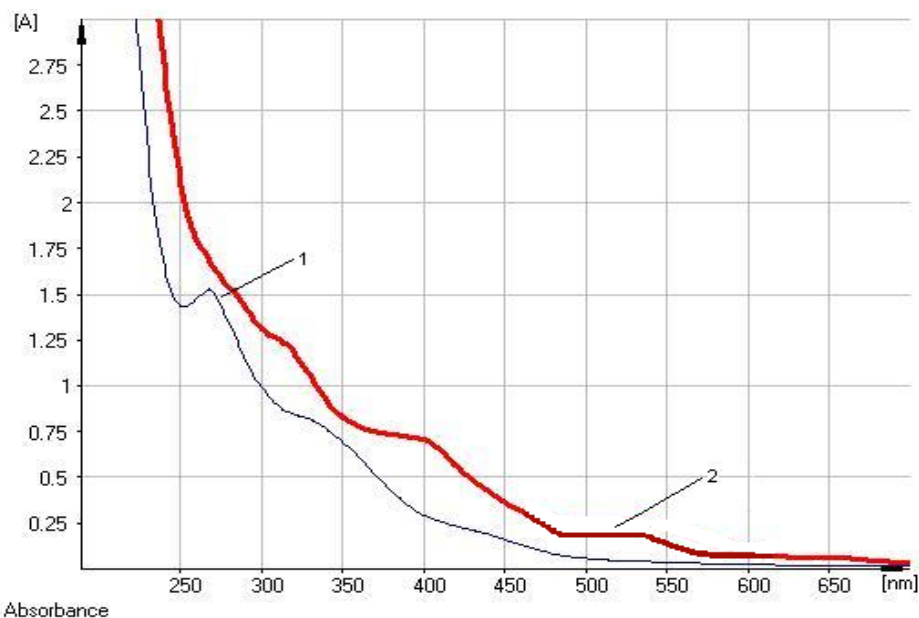


Fig 1: Electronic spectra of the starting solution (1) and an alkaline ammoniacal solution (2) of the hydroalcoholic extract of "the Siberian swallow".

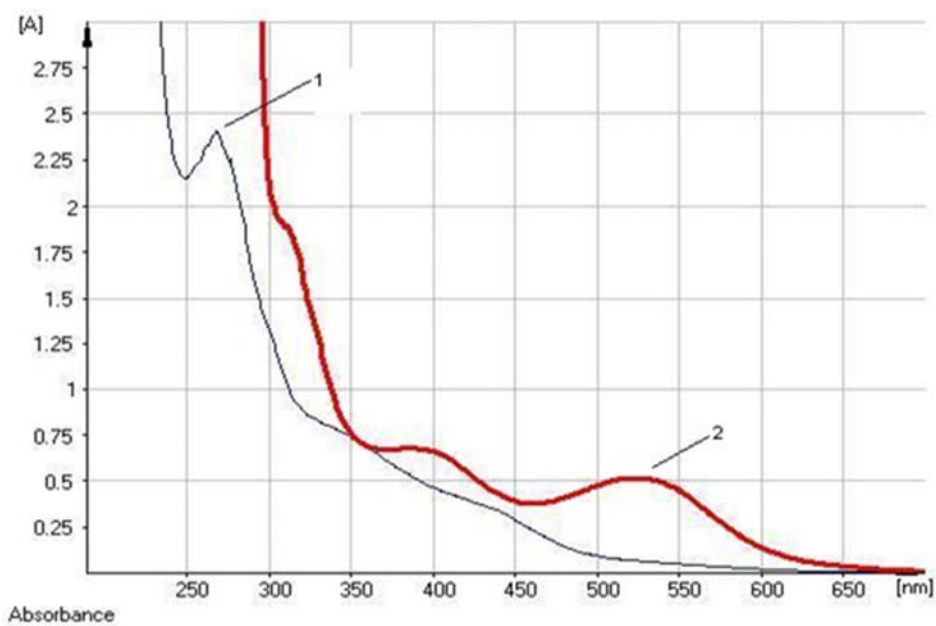


Fig 2: Electronic spectra of the starting solution (1) and an alkaline ammoniacal solution (2) of the hydroalcoholic extract of bark of *Frangulae alnus*.

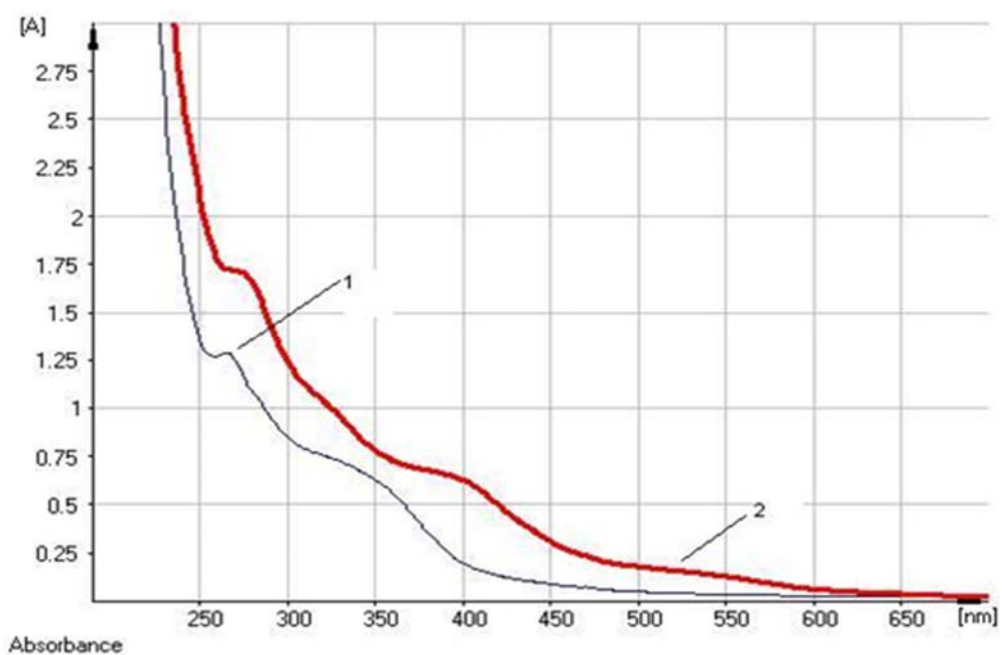


Fig 3: Electronic spectra of the starting solution (1) and an alkaline ammoniacal solution (2) of the hydroalcoholic extract of *Senna* leaves.

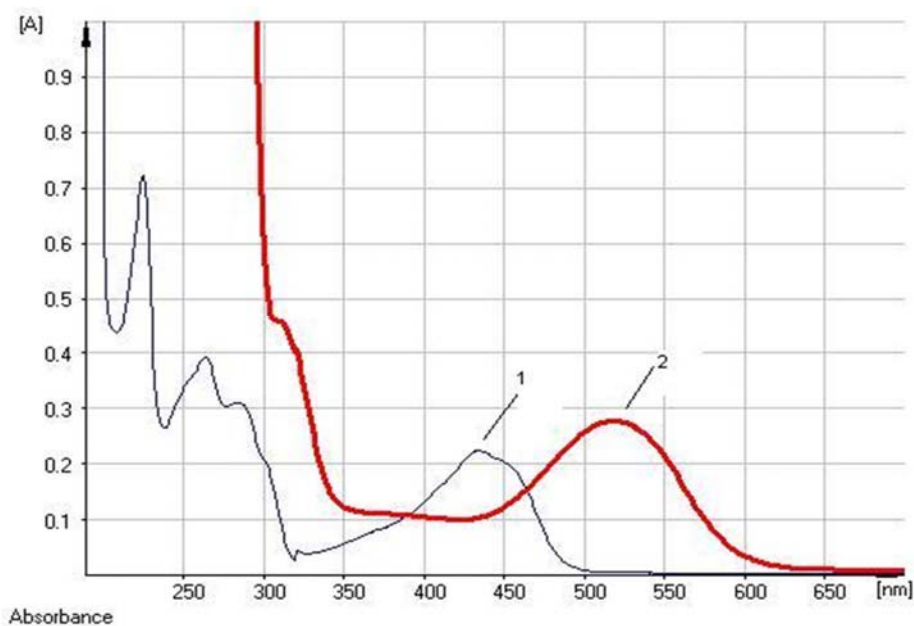


Fig 4: Electronic spectra of frangulin A feed solution (1) and an alkaline ammoniacal solution of frangulin A (2).

To develop methods of quantitative determination of the amount of anthracen derivatives us the optimal conditions for the extraction of anthracen derivatives from the species "the Siberian

swallow": extragent 40% ethyl alcohol; the ratio of "raw materials-extracting" -1:50; the extraction time -extraction on a boiling water bath (moderate boiling) within 90 min (Table. 1).

Table 1: Dependence of the completeness of extraction amount of anthracen derivatives collection "the Siberian swallow".

№	Extractant	The ratio of raw material: extractant	Extraction time, min	Contents of total anthracenderivatives calculated on frangulin A and absolutely dry raw material (in%)
1.	30% ethanol	1:50	60	3,40 ± 0,06
2.	40% ethanol	1:50	60	3,45 ± 0,05
3.	50% ethanol	1:50	60	3,48 ± 0,04
4.	60% ethanol	1:50	60	3,50 ± 0,05
5.	70% ethanol	1:50	60	3,57 ± 0,07
6.	80% ethanol	1:50	60	3,58 ± 0,08
7.	40% ethanol	1:50	30	3,60 ± 0,06
8.	40% ethanol	1:50	90	3,63± 0,05
9.	40% ethanol	1:50	120	3,56 ± 0,06
10.	40% ethanol	1:30	90	3,54 ± 0,08
11.	40% ethanol	1:100	90	3,53 ± 0,05
12.	40% ethanol	1:200	90	3,51 ± 0,06

A technique of quantitative definition of the total anthracen derivatives in "the Siberian swallow". Analytical sample species is crushed to the size of the particles passing through a sieve with apertures in diameter of about 1 mm. 1 g chopped species (precise linkage) is placed in a flask with a grinding capacity of 100 ml, add 50 ml of 40% ethyl alcohol. Closed the flask and weigh on calibrated scale accurate to $\pm 0,01$ g. Flask attached to reverse refrigerator and heated on a boiling water bath (moderate boiling) within 90 minutes. Then the flask close with the same tube, weighed again and fill in the missing extragent to the original mass. Removing filtered through paper filter («red» band) and cool for 30 minutes. Tested solution is prepared as follows: 1 ml of obtained extract placed in a volumetric flask with a capacity of 50 ml and bring the volume of the solution to the mark alkali-ammoniac solution (test solution). The test solution A placed in a flask with a capacity of 50 ml and heated for 15 min in a boiling water bath with reverse refrigerator. After cooling measure the optical density of the solution on the spectrophotometer at a wavelength of 524 nm. The reference solution is purified water.

Note: preparation of the solution fragulin A - standard sample. About 0.02 g (precise linkage) fragulin A placed in a volumetric flask with a capacity of 50 ml, dissolved in 30 ml of 70% ethyl alcohol when heated in a water bath. After cooling the contents of the flask to room temperature bring the volume of the 70% solution of ethyl alcohol up to the mark (solution A fragulin A). 1 ml solution A of fragulin A placed in a volumetric flask 25 ml and bring the volume of the solution to the mark alkali-ammoniac solution (test solution B). Solution B is placed in a flask with a capacity of 50 ml and heated for 15

min in a boiling water bath with reverse refrigerator. After cooling measure the optical density of a solution on the spectrophotometer at a wavelength of 524 nm. The reference solution is purified water. Content amount of anthracen derivatives in "the Siberian swallow" in terms on fragulin A and absolutely dry raw materials in percent (X) is calculated by the formula:

$$X = \frac{D * m_0 * 50 * 1 * 50 * 100 * 100}{D_0 * m * 50 * 1 * 25 * (100 - W)}$$

Where D is optical density of the test solution; D_0 - optical density of the solution nor fragulin A; m - the mass of raw material, g; m_0 - the mass of the working standard sample fragulin A, g; W - loss of mass on drying in percent. A simplified calculation formula as an alternative:

$$X = \frac{D * 50 * 50 * 100}{m * 180 * (100 - W)}$$

Where D - optical density of the test solution; W - loss of mass on drying in percentage; 180 - specific absorption of the working standard sample fragulin A.

Metrological characteristics of the methodology of quantitative measurement of the amount of anthracen derivatives in "the Siberian swallow" presented in table 2. The results of statistical processing of experiments show that the error of a single determine the amount of anthracen derivatives in practical with confidence probability of 95% is $\pm 4,25$ % (Table 2).

Table 2: Metrological characteristics of the methods of quantitative determination of the amount of anthracen derivatives in "the Siberian swallow":

f	\bar{X}	S	$P, \%$	$t(P, f)$	ΔX	$E, \%$
10	3,63	0,0692	95	2,23	$\pm 0,154$	$\pm 4,25$

Using the developed methods we analyzed a number of sample practical (Table 3) and determined that the content of the amount of anthracen derivatives varies from 3,50% to 3,63%,

which can be recommended as a lower limit for raw materials this plant the content of the amount of anthracen derivatives not less than 3,0 per cent.

Table 3: The total content of anthracen derivatives in various samples of "the Siberian swallow":

S. №	Characteristics of the sample materials	Contents of total anthracen derivatives calculated on frangulin A and absolutely dry raw material (in%)
1.	ОАО "Krasnogorskleksredstva" (Moscow region, 2011)	3,50 ±0,10
2.	ОАО "Krasnogorskleksredstva" (Moscow region, 2012)	3,58 ±0,12
3.	ОАО "Krasnogorskleksredstva" (Moscow region, 2013)	3,63 ±0,11

4. Conclusions

1. Based on the results of chemical researches the bark of *Frangula alnus* developed methodological approaches to the standardization of "the Siberian swallow", consisting in the determination of anthracen derivatives and the using of techniques of the analysis of standard sample of frangulin A.

2. The method of quantitative determination of the total anthracen derivatives in terms frangulin A practical using 40% ethyl alcohol as a solvent and UV-spectroscopy at the analytical wavelength 524 nm.

3. The research results allow to recommend a lower limit on the content of the total anthracen derivatives in practical not less than 3,0 per cent.

5. References

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