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Research Article

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Development of Technology and Studying of Syrup with Acorus Calamus Rhizome Extract

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ABSTRACT

This article includes the description of obtaining of Acorus Calamus rhizome extract by the percolation method and the formulation of this extract in the syrup as a pharmaceutical dosage form. The percolation rate was 0.8 ml/min. The extraction of raw materials fraction with a particles size 0.25-3 mm has been investigated. It was found that the using of 50% ethanol as the extractant maximizes raw material exhaustion at a ratio of raw material: extractant 1 : 6. The results of the syrup development with the Acorus Calamus extract have been shown that the optimal base was sugar syrup. Such physical-chemical data as description, density, identification of the syrup have been determined.

Keywords: Acorus Calamus rhizome; Extraction; Percolation; Particles size; Extractant; Syrup

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INTRODUCTION

Today herbal medicines are medicines of the first choice in the treatment of diseases of different etiologies in many cases [1]. Unlike synthetic drugs, herbal medicines have a less possibility of the adverse reactions development along with the effectiveness. The steadily increasing bacterial resistance to existing antibiotics is a serious problem, and therefore there is a great need to search for new classes of antibacterial substances, especially from natural sources [2-4].

The perennial wild herbaceous plant *Acorus calamus L.* (of the family *Araceae*) is of interest among the variety of medicinal plants. Acorus Calamus rhizome extract contains α and β-asarones, sequesterpenes (calamusin A-H), norsequesterpine (calamusin I), polysaccharides (D-galacturonic acid with residues of galactose, arabinose, xylose, and rhamnose) and other biologically active substances. In folk medicine, drugs of Acorus calamus are recommended for the treatment of diseases of the central nervous system, the blood system, as an anti-inflammatory, analgesic, wound-healing, and sedatives. Acorus calamus rhizome extract promotes healing of festering wounds and ulcers, is used for mouthwashes in case of gum diseases. In officinal medicine *Acorus calamus* has found application as a part of complex products to excite the appetite and improve digestion in disorders of gastrointestinal tract functions [5-7]. The rhizome part of Acorus Calamus is found to possess the antibacterial activity against the bacterial strains of *Salmonella typhi, Pseudomonas aeruginosa, Klebsiella pneumoniae, Staphylococcus aureus, E. coli*. The extract of Acorus Calamus also showed the anti-fungal activity against the yeast strain of Candida Albicans and other fungi strains [8-10]. However, the use of Acorus calamus rhizome for the treatment of infectious inflammatory diseases of various organs in modern medicine, as well as the technology of drugs on the basis of this plant raw material is not well understood. Therefore the development of the technology of Acorus calamus extract for further medicine creation on its basis is relevant.

The aim of our research was to investigate parameters of the Acorus calamus rhizome extraction process, such as the required amount of extractant to complete depletion of the raw material and the yield of extractive substances by using the particular extractant, to develop composition, the scientifically and experimentally grounded technology of the syrup based on Acorus Calamus rhizome extract.

EXPERIMENTAL SECTION

The object of our research was Acorus Calamus dried rhizome (manufactured by the Private Joint-stock Company "Liktravy", Ukraine) and the syrup on its basis. The auxiliaries (ethanol, fructose, sucrose and sorbitol) met the requirements of State pharmacopoeia of Ukraine.

The moisture content of the initial plant raw material was determined (9.25%). Rhizome was crushed and sieved, the fraction with the particles size of 0.25-3 mm was used for the extraction. The extractant was 50% ethanol, which was chosen to enable extraction of both lipophilic substances (essential oil, terpenoids et al.) and hydrophilic substances (tannins, polysaccharides et al.).

The extract was obtained by the percolation. A sample (50±0.5 g) of the previously powdered Acorus Calamus rhizome was loaded in the laboratory extractor. The plant raw material was pressed with perforated disc for a maximum air displacement, filled with the extractant to "mirror" and left to steep for 24 hours. After steeping the actual percolation was performed with the rate of 0.8 ml/min while supplying fresh extractant. For studying the yield of extractives during the extraction some 50 ml extract portions were obtained (an equal amount in relation to the weight part of the loaded raw material). The yield of extractive substances for the each extraction portion was calculated after the evaporation of aliquots at 105 °C using the hygrometer «Sartorius» MA 150 (Germany) [11].

To obtain Acorus Calamus extract the first extract portion in an amount of 85% (43 ml) to the raw material mass was collected in a separate container (portion I). Subsequent portions of extract were combined. The combined extract was evaporated under vacuum at a temperature of 50-60 °C using a rotary evaporator ER-1M3 (JSC "Khimlaborpribor", Russia) to reach 7 ml residue (portion II – 15% relative to the mass of raw material loaded into the percolator). After cooling, the condensed residue was dissolved in the extraction portion I. The final extract was obtained in a ratio 1:1 relative to the raw material mass. The obtained extract was filtered.

The determination of relative density, ethanol content, dry residue of extract was carried out by the methods of State Pharmacopeia of Ukraine 2.0 [12].

The main active substances of Acorus Calamus extract are terpenoids and polysaccharides that make anti-inflammatory and antibacterial activity. That is why the effectiveness of extraction was considered to release these compounds.

The identification of polysaccharides in the obtained extract was carried out by using gravimetry: 1 g of liquid extract was dissolved in 10 ml of purified water. Polysaccharides were precipitated with threefold amount of 96% ethanol R [13].

The identification of terpenoids was carried out by Thin layer chromatography using «Sorbfil» PTLC-P-V-UV plate in a system of solvents ethyl acetate R – hexane R (10:90, V/V). Detection: the plate was sprayed with anisaldehyde solution R and was heated at a temperature of 105 °C to 110 °C for 8-10 min. The plate was viewed in daylight. The chromatogram obtained with the test solution showed several zones: gray, three purple zones and pink zone [14].

The syrups organoleptic evaluation was conducted by AI Tentsova method. The group of 20 people evaluated syrups with extracts and various sweeteners. Tasters had normal taste and did not take food and smoke 30 minutes before the experiment in the study. A single dose of 5 ml syrup was taken. The time interval between individual tastings was 15 minutes. Tasters necessarily rinsed oral cavity before and after the test. Samples of syrup were not swallowed. Each taster assessed the taste the five-point system: very nice – 5, nice – 4, good – 3, bad – 2, very bad – 1 [15,16]. The taste numeric index was inferred as mean value of all indicators. To ensure the reliability of the method, another group of 20 tasters conducted the organoleptic assessment of syrups, but with other points value in terms of the main taste on common classification: not bitter – 5; insignificantly bitter – 4; slightly bitter – 3; bitter – 2; very bitter – 1. The numeric index of main taste was inferred on the received data. The more main taste numeric index, the potential masking by sweetener is much greater. The results of the determination of two tasters groups were generalized and set out in table format.

The valuation taste panel (by IA Egorov) was used in addition to organoleptic evaluation of syrups. The method was to compiling taste formulas using letters and numeric index. The sensation of a taste was conventionally indicated by letters (Sw – sweet, B – bitter, S – salty, So – sour) and numeric index (1 – unsweetened, not bitter, unsalted, not sour; 2 – slightly sweet, slightly bitter, slightly salty, slightly sour; 3 – sweet, bitter, salty, sour; 4 – very sweet, too bitter, too salty, too sour [17,18].

RESULTS AND DISCUSSION

The resulting dependence of the yield of extractive substances from Acorus calamus rhizome on the amount of the extractant (50% ethanol) is shown in Figure 1.

As illustrated in figure, six times amount of the extractant 50% ethanol relative to the weight of raw material in the extractor should be used for the complete depletion of the plant raw material. In the process of percolation with a greater volume of extractant the extractives yield significantly decreased and was less than 0.6%.

The obtained extract was analyzed on some characteristics such as description, relative density, dry residue, and identification necessary to justify the further composition and technological researches of the syrup. The indicators of Acorus calamus extract are given in the table 1.

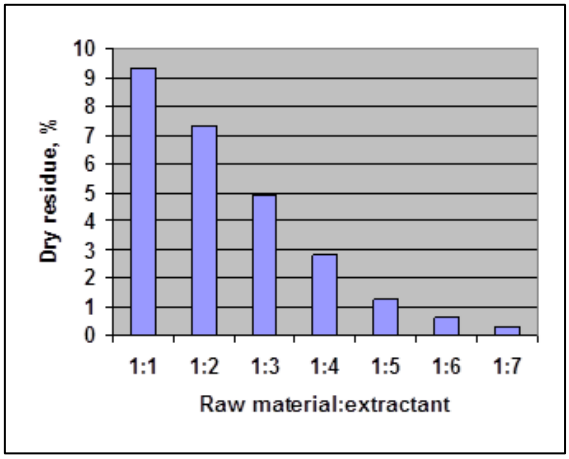

Figure 1: The rhizome extractives yield dependence on the extractant amount relative to the mass of the plant raw material

It was found that the obtained extract had a characteristic unpleasant bitter taste but nice smell. Taste confirmed the need to develop corrected form of extract. The studies showed the presence of terpenoids and polysaccharides in the extract.

Further researches were aimed on the development of the optimal taste composition and the basis for a syrup with the extract. Sugar syrup, sorbitol and fructose solutions were used as a sweetener system in the following proportions: saccharose and purified water – 64 : 36; sorbitol and purified water – 70 : 30; fructose and purified water – 70 : 30.

Table 1: The indicators of Acorus calamus liquid extract

Index	Characteristics
Description	A thick brown liquid with an intense characteristic smell and characteristic pungent and bitter taste
Dry Residue, %	5.1 ± 0.5
Relative Density, g/cm^3	0.974 ± 0.006
Ethanol content, % V/V	49 ± 1.14
Identification	The presence of terpenoids and polysaccharides

The technological flowshart of the syrup obtaining is presented in Figure 2. The technology was as follows. °Concentrated sweetener solutions were prepared by heating to 100 °C. The Acorus calamus extract was added to the cooled syrup.

The extract dose in the syrup was selected according to the literature data [19,20] taking into account the syrup dosage with a teaspoon and syrup density. The dose was 3 ml of extract per 100 ml of syrup.

In the study of medicines for oral use their taste characteristics were determined. The evaluation results of organoleptic properties of Acorus Calamus syrup are shown in Table 2. In the study, sucrose syrup received the highest rating organoleptic properties. Sucrose corrects the bitter taste of the extract. The obtained syrup was transparent viscous solution with sweet taste, yellowy-brown colour and aromatic smell.

Table 2: The results of the evaluation of syrups organoleptic properties

Name flavouring compositions	Numeric index value (by Tentsova method)	The valuation taste panel (by Egorov)	
	The sense of taste/the sense of main taste	The taste formula	The general taste
Composition with sucrose	4.5/5	Sw3	Sweet
Composition with fructose	3.7/4.6	B2Sw3	Slightly bitter, sweet
Composition with sorbitol	4.0/4.4	B2Sw2	Slightly bitter, slightly sweet

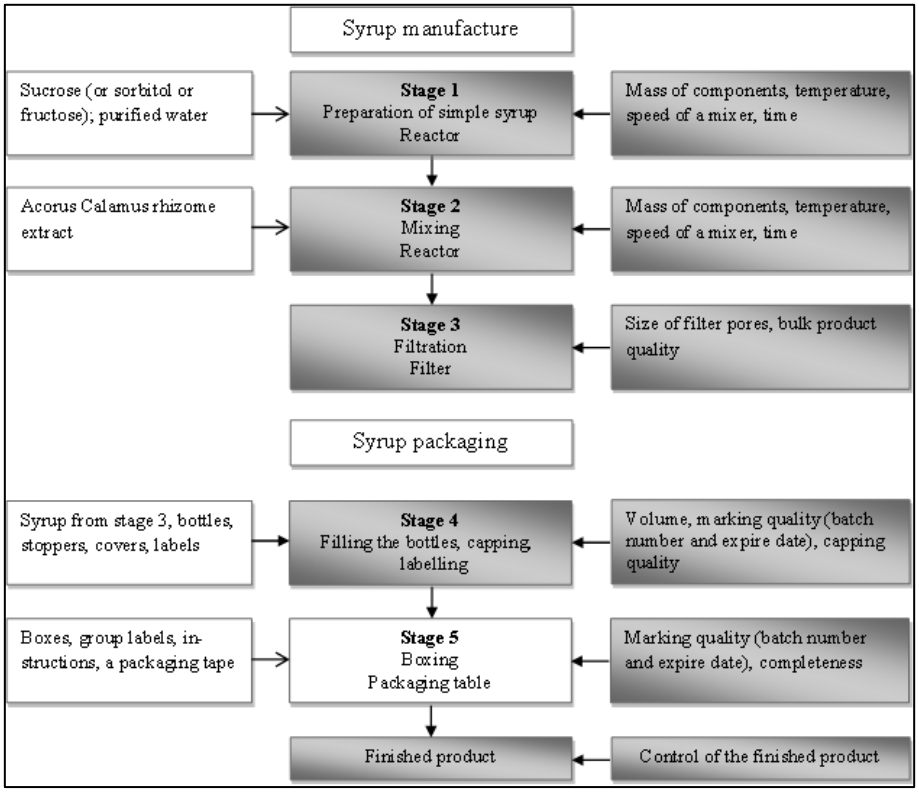

Figure 2: The flowchart of technological process of syrup

Thus, the experimental reasonable composition of Acorus Calamus syrup was offered: sucrose – 64.0 g; purified water –36.0 ml; Acorus Calamus rhizome extract – 3 ml.

The estimation of the obtained syrup quality was on the organoleptic and physical-chemical properties: description, density, identification. The results are presented in Table 3.

The further investigation of the stability of Acorus Calamus syrup confirmed the right choice of excipients since there were no changes of quality during the shelf-life

Table 3: The indexes of the syrup quality

Index	Characteristic
Description	The transparent viscous solution with sweet taste, yellowy-brown colour and aromatic smell
Density, g/ml	1.3 ± 0.003
Identification	The Presence of terpenoids and polysaccharides

CONCLUSION

As a result of technological researches the rational parameters or the extraction process of Acorus Calamus rhizome with 50% ethanol by the percolation have been determined. The quality indexes of the obtained liquid extract have been studied. The optimum base for syrup with Acorus Calamus extract (sucrose syrup) has been selected. Organoleptic parameters and some physical-chemical indexes (description, density, identification) that can be included in the reference documentation for syrup have been determined.

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