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Optimization of the extraction process of *Pinus sylvestris L.* pine bark with monoethanolamine

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Abstract. The paper presents the results of optimizing the process of extraction of the bark of *Pinus sylvestris L.* Scotch pine. The input parameters were the duration of the process and the concentration of the monoethanolamine solution. The experimental data were processed using the STATGRAPHICS® Centurion software package. It has been established that the maximum extraction of extractive substances occurs when the duration of the extraction process is 5 hours with a concentration of monoethanolamine of 5%.

1 Introduction

The bark of a tree performs many different functions during the life of a plant. The thickness of the bark depends on the age and place of growth, as well as on environmental factors. The bark protects the tree from the weather, pests, and fires. The bark can be considered as a bioindicator due to its porosity and structure.

The Krasnoyarsk Territory is one of the leading regions in Russia in terms of timber harvesting. As a result of its processing, waste is generated that is not widely used. Part of the bark is utilized immediately in wood processing industries as an energy source by burning, which is not economically viable due to the high content of ash and humidity. Part of the bark is used in agriculture as a mulch material, is an effective way to retain water in the soil, as well as an alternative option for weed control. In addition, the bark can serve as an element of the design landscape [1].

Despite this, waste is a weak link in the complex processing of wood. Millions of tons of such waste has been in dumps for many years, creating a burden on the environment, and in the summer it is a fire hazard. Pollution of the environment with phenolic compounds formed as a result of decomposition can lead to a violation of the biological balance between the individual links of biogeocenoses and thereby cause great damage to the national economy. The content of valuable components in the bark allows it to be used as a raw material base for chemical processing.

So, for example, conifers are sources for obtaining biologically active substances, in particular polyphenols - dihydroquarcetin and dihydrokaempferol. A biologically active additive (BAA) Pinocalar, a natural antioxidant based on phenolic compounds, has been obtained, which has a number of properties, such as hepatoprotector, capillary protection, and antioxidant activity [2]. Phenolic compounds, especially of plant origin, have

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antimicrobial and antibiofilm activity, which are used against a wide range of microorganisms [3]. Pine bark extracts exhibit bacteriostatic activity against gram-positive and gram-negative strains [4]. There is evidence of the possibility of obtaining pectins from the bark of larch and pine, which are used in the food industry as a gelling agent, as well as in pharmaceutical preparations to remove radionuclides from the human body [5].

Since the bark differs from wood in a higher content of extractive substances, the extraction of extractive substances using various extractive agents is envisaged at the first stage of any chemical processing of the bark. During the extraction of the bark, various chemical compounds are released that can be used in all spheres of human life.

The most important component of the extractive substances of the bark are tannins - they are able to turn raw skin into tanned [6]. As a result of tanning, the spatial structure of collagen is strengthened due to the formation of mainly hydrogen cross-links between tannins and chains of the protein structure.

Tannins play an important role in the life of plants, take part in the final stages of biological oxidation, can be regulators of growth, development, reproduction, etc. In the national economy, tannins are used to control the viscosity of drilling fluids, are used as colloidal stabilizers - anticorrosive descaler in industrial installations. Tannins are also used in the production of ink, the refinement of low-grade ores, and oil refining. In addition, tanning extracts have found application in the production of plastics and binding materials for the preparation of plywood and furniture from sawdust and shavings. The bark of the tree is used to produce environmentally friendly insulating boards, which reduces the amount of free formaldehyde [7], and also as a filler in composites based on polylactic acid (PLA) [8].

The use of water as an extractant in the processing of the bark on an industrial scale does not allow the maximum extraction of the substances of the phenolic complex (the yield of the extract, as a rule, does not exceed 4% by weight, a.c. of the raw material). An increase in the efficiency of the extraction process can be carried out using an extractant with high selectivity with respect to substances of a phenolic nature, for example, the use of an aqueous solution of sodium hydroxide makes it possible to extract more than 40% of extractive substances from pine bark [9]. In addition, ampholytes, in particular, monoethanolamine, which has alkaline and alcohol properties, are good solvents [10–13].

The purpose of this study was to find the optimal conditions for the extraction of the bark of *Pinus sylvestris L.* Scots pine with an aqueous solution of monoethanolamine.

2 Research methods

The initial raw material was the bark of *Pinus sylvestris L.* Scotch pine, harvested in the Krasnoyarsk Territory. The bark was crushed in a disintegrator to a homogeneous composition with a particle size of 0.5-1 mm.

The air-dry bark was extracted with an aqueous solution of monoethanolamine at various concentrations (1, 3, and 5%). The choice of MEA is due to the fact that it is a typical ampholyte and, therefore, its presence contributes to the transition of substances of a different nature into the liquid phase. In addition, being an antioxidant and a weak lignin destroyer, MEA is able to prevent oxidative processes, as well as inhibit the condensation of polyphenolic compounds and ensure the preservation of the carbohydrate complex. When using MEA, the speed and completeness of the extraction of extractives increases, as it is a good swelling agent for plant materials. In addition to monoethanolamine, other ampholytes have the same properties.

Extractions were carried out at the boiling temperature, with a liquid modulus (core: extractant ratio) equal to 14, and the extraction duration varied in the range of 1–5 h.

The quantitative content of the total flavanoids in the extracts was determined using a UV 3000 spectrophotometer. A 2% alcoholic solution of aluminum chloride was added to the volume of an aliquot of the extract and adjusted to the mark with 95% ethyl alcohol, and the optical density was measured at a wavelength of 410 nm against the background of ethyl alcohol [15].

To build a mathematical model, check its adequacy and determine the significance of each technological parameter taken into account, the influence of the concentration of monoethanolamine and the duration of the extraction process was studied. The planning of the experiment was based on the plan of the second order - the plan on the cube (plan Ko-2), which has good statistical characteristics with a small number of experimental points. The main factors and levels of their variation are presented in Table 1. The following optimization parameters were chosen: Y_1 – yield of extractive substances, %; Y_2 – yield of total flavonoids, %. The regression equations and the values of the factors of the extraction process were obtained using the STATGRAPHICS® Centurion software package [16]

Table 1. The main variables of the experiment and the levels of their variation for the extraction of *Pinus sylvestris* L. Scots pine.

Plan characteristics	Variable factors	
	MEA, %, X_1 concentration	Durability, h, X_2
Main level, X (0)	3.0	3.0
Variation step	2.0	2.0
Upper level, X (+1)	5.0	5.0
Lower level, X (-1)	1.0	1.0

3 Research results

The plan and results of the factorial experiment are shown in Table 2. For the reliability of the results obtained, the experiment was carried out in duplicate.

Table 2. Plan and results of a factorial experiment to optimize the extraction of *Pinus sylvestris* L. Scots pine bark.

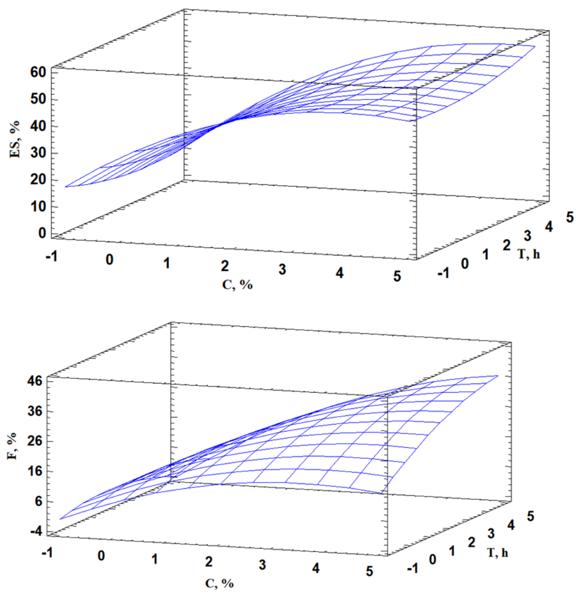
Experiment factors		Yield of extractives, Y_1, %	The output of the sum of flavanoids, Y_2, %
MEA concentration, X_1, %	Durability, X_2, h		
5	5	54.76	39.78
		56.15	33.45
3	5	55.22	29.75
		55.78	30.38
1	5	37.72	15.23
		36.74	17.52
5	3	50.59	34.35
		49.45	31.09
3	3	50.69	28.47
		47.89	29.08
1	3	35.35	19.26
		37.34	17.38
5	1	51.35	25.90
		48.44	25.09
3	1	42.89	22.19
		47.68	25.91

1	1	36.37	15.52
		37.43	17.52

As a result of mathematical processing of experimental data, regression equations were obtained, which have the following form:

$$У_1 = 27{,}2166 + 11{,}3383X_1 - 1{,}87783X_2 - 1{,}42979X_1^2 + 0{,}32725X_1X_2 + 0{,}373021X_2^2$$

$$У_2 = 8{,}65103 + 6{,}43815X_1 + 1{,}93456X_2 - 0{,}816833X_1^2 + 0{,}695312X_1X_2 - 0{,}432083X_2^2$$



ES is the yield of extractives, %; F is the total yield of flavonoids, %.
T - duration, h; C – MEA concentration, %;

Fig. 1. Response surfaces obtained from the extraction of *Pinus sylvestris* L. Scots pine bark.

As the results of the regression analysis show, the concentration of monoethanolamine and the duration of extraction have a significant role in the process of extracting extractive substances from pine bark. Statistical significance is affected by MEA concentration as well as pair interaction with duration. Thus, an increase in the concentration of monoethanolamine leads to an increase in both the yield of extractive substances and the content of the total flavonoids in them. It should be noted that an increase in the duration of the extraction process with a 1% MEA solution for more than 1 hour does not significantly affect the yield of extractives and the amount of flavanodes, while the good quality (the ratio of phenolic substances to the yield of extractives) is about 42%. Increasing the concentration of MEA to 3% makes it possible to increase the yield of extractives from 42 to 55%, while the quality of the extract increases to 55%. The use of 5% MEA also increases the yield of extractives up to 56%, while the good quality is 62-65%.

Since MEA is a good swelling agent, a further increase in concentration makes it difficult to separate the reaction mass, which is an undesirable factor when implementing the method under production conditions. On the basis of experimental studies and statistical processing of the results, optimal conditions were found: MEA concentration - 5%; extraction duration - 5 hours; hydromodulus - 14. Under optimal conditions, the extract obtained has a good quality of 65%, and allows it to be used for further processing, which consists in adjusting the pH and giving a marketable appearance (solution concentration or drying). Unlike aqueous extracts, monoethanolamine extract retains its properties for a long time, since monoethanolamine has good stabilizing properties, so it can be released in liquid form, which significantly reduces the cost of the extract. After extraction processing, a solid residue remains, which is cellolignin and can be used for further processing. One option is to use it as a substrate for cultivating micro- and macromycetes [17, 18]. Mushrooms are a promising producer of biologically active substances that have a powerful source of enzymes.

4 Conclusion

Thus, the results of the study showed that the yield of extractives and the amount of flavanoids are significantly affected by the duration of the process and the concentration of monoethanolamine in the solution. The optimal conditions for the extraction process with an aqueous solution of monoethanolamine have been established: the duration is 5 hours, the concentration of MEA is 5%, the hydromodulus is 14, which allow extracting more than 50% of extractive substances from the bark with a content of more than 65% of substances of a flavanoid nature in them, and use the solid residue in as a substrate for the cultivation of micro- and macromycetes.

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