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OPTIMIZATION OF THE PROCESS OF EXTRACTION OF THE ABOVE-GROUND PART PLANTS HAPLOPHYLLUM PERFORATUM JUSS

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Abstract

Studies are presented to study the optimization of the extraction process of the aerial part of the plant Haplophyllum perforatum Jussem. Rutaceae growing on the territory of the Republic of Uzbekistan using the Box-Wilson mathematical method, and optimal conditions for extraction from raw materials were identified, namely extraction with 96% alcohol at a temperature of 35 °C, extraction time – 6 hours.

Keywords: *Haplophyllum perforatum, aerial part, optimization, Box-Wilson method, insecticidal activity*

Introduction

In world practice, synthetic agents are widely used to protect plants from pests. Improper use and increased consumption of chemicals negatively affects human health and the environment. It is known that due to pests, the loss of agricultural products per year is 50–70%. Based on this, the search and implementation of environmentally friendly plant protection products is considered an urgent task.

Recently, interest has increased in plants that produce secondary compounds with repellent, insecticidal and acaricidal effects (Turaeva S.M., Kurbanova E.R., Mamarozikov U.B., Zakirova R.P., Khidirova N.K., 2022). This indicates the possibility of obtaining biological products from them as an alternative to synthetic pesticides (Mamarozikov U.B., Zakirova R.P., Khidyrova N.K. Rakhmatov A., Asatova S., 2017). Previously, we identified the insecticidal activity of the extract of the aerial parts of Haplophyllum perforatum Juss plants against aphids Aphis pomi, S. graminum, Macrosiphum euphorbiae (Mamarozikov U.B., Bobakulov Kh.M., Turaeva S.M., Zakirova R.P., Rakh-matov Kh.A., Abdullaev N.D., Khidyrova N.K., 2019).

This paper presents data on optimizing the extraction process of the aerial part of the plant Haplophyllum perforatum Juss. It is known that the extraction of natural compounds depends on many factors, each of which affects the yield of the final product to a greater or lesser extent.

Therefore, to assess the degree of their influence on extraction, as well as to determine the conditions for the maximum yield of dry extract from leaf plants, the method of mathematical planning of an experiment according to Box–Wilson was used (Hajibaev T.A., Khalilov R.M., Sagdullaev Sh. Sh., 2017; Ibragimov T.F., Khadjibaev T.A., Li A.V., Dzhanibekov A.A., 2015).

Method and Results

The optimization parameter was the yield of dry extract from the content in the raw material at the first contact of the phases. In all experiments, the amount of raw material and the isolation method were identical.

Based on a priori information (in this case, the results of one-factor experiments), we selected the factors that most influence extraction and established the following main levels and ranges of variation for them (Table 1):

X_1 – extraction temperature, °C;
X_2 – duration of the process, h;
X_3 – amount of ethyl alcohol,

Table 1. *Factors and ranges of variation*

Factor level	Factor		
	X_1	X_2	X_3
Upper	35	28	1:12
Average	30	25	1:10
Lower	25	22	1:8
Variation interval	5	3	2
Unit	°C	h	

Two levels of four factors are established, i.e. full factorial experiment of type 23. A matrix for planning experiments was compiled (**Table 2**) and the results of the experiments were recorded in it.

Table 2. *Experiment planning matrix and their results*

Experience No.	Factor code				Y_1	Y_2	$Y_{av.}$
	X_0	X_1	X_2	X_3			
1	+	+	–	–			6.512
2	+	+	–	+			6.615
3	+	+	+	–			6.550
4	+	+	+	+			6.619
5	+	–	–	–			5.560
6	+	–	–	+			5.618
7	+	–	+	–			5.673
8	+	–	+	+			5.810

Each of the 8 experiments was carried out in accordance with the compiled matrix, using selected levels of each factor, coded in the matrix with the signs «+» or «–» (upper and lower levels of variation, respectively).

For example, experiment № 1 was performed under the following conditions: plant leaves: extractant alcohol (1:8);

– extracted at a temperature of 35 °C, the duration of phase contact was 22 hours.

Experience № 5. plant leaves: alcohol extractant (1: 8):

– extracted at a temperature of 25 °C, the duration of phase contact was 22 hours and so on in all 8 experiments.

We present the experimental results in the form of a regression equation:

$$Y = b_0+b_1x_1+b_2x_2+b_3x_3;$$

where: b_0, b_1, b_2, b_3, are the regression coefficients of the complete quadratic equation.

Experimental Part

Postulating that the process under study at given intervals of variation of variables can be described by a linear dependence, and using the least squares method, we determined the coefficients:

$$b_i = \frac{\sum_{i=1}^{N}(X_{ij} \times Y_i)}{N} \qquad (1)$$

where: i is the number of experience (1,2...8); j-factor number (1,2...4); Xij – coded value of factors; N – is the number of experiments in the matrix.

Using formula 1, we calculated the values of the regression coefficients:

$b_0 = 6.125$; $b_1 = 0.45$; $b_2 = 0.048$; $b_3 = 0.05$;

Substituting the calculated values of the "b" coefficients into equation 2, we obtained:

$Y = 6.125 + 0.45X_1 + 0.048X_2 + 0.05X_3$

As a result, a mathematical model of the process was established, which is a first-order regression equation. To verify the correctness of the experiment and the adequacy of the resulting model, statistical processing of the data obtained was carried out (Table 3).

Table 3. *Statistical analysis*

Y_1	Y_2	Y_{av}	D	D	S_i^2	Y_{cal}	D	(D
6.522	6.502	6.512	0.01	0.0001	0.0002	6.48	0.032	0.001
6.605	6.625	6.615	0.01	0.0001	0.0002	6.58	–0.035	0.001
6.590	6.510	6.550	0.04	0.0016	0.0032	6.57	–0.02	0.0004
6.649	6.589	6.619	0.03	0.0009	0.00018	6.67	–0.051	0.003
5.570	5.55	5.560	0.01	0.0001	0.0002	5.58	0.02	0.0004
5.608	5.628	5.618	0.01	0.0001	0.0002	5.68	–0.062	0.0038
5.690	5.655	5.673	0.017	0.00029	0.00058	5.67	0.003	0.0009
5.828	5.792	5.810	0.018	0.00032	0.00064	5.77	0.04	0.0016
Sum					0.00540		–	0.0121

To determine the variation in the values of repeated experiments, we used the variance calculated by the formula:

$$S_i^2 = \frac{\sum_{q=1}^{n}(Y_q - Y_{cp})}{n-1} \qquad (2.3)$$

where: Y_q is the result of a separate experiment; Y_{cp} is its arithmetic mean value; $(n - -1)$ – the number of degrees of freedom equal to the number of repeated experiments minus one.

For two repeated experiments, formula 2.3 took on the following form:

$$S_i^2 = \frac{2\Delta Y^2}{1} \qquad (2.4)$$

Homogeneity of variance was carried out using the Cochran criterion:

$$G_{экс} = \frac{S_{\max}^2}{\sum_{i=1}^{N} S_i^2} \le G_{kp} \qquad (2.5)$$

$G_{кр} = 0.6798$ [129]
$G_{экс} = 0.12$
$0.12 < 0.6798$

The result obtained corresponds to the conditions of formula 5. The dispersion is homogeneous.

To check the adequacy of the resulting model, the variance of adequacy was first determined.

$$S_{ad}^2 = \frac{\sum_{i=1}^{N}(\Delta Y_i')^2}{f} \qquad (2.6)$$

Then Yras was found; (Table 2)

Based on the results obtained, we find – Y_i' using the formula

$$\Delta Y_i' = Y_{cp} - Y_{pac}; \qquad (2.7)$$

After this, the reproducibility variance was determined using the formula:

$$S_Y^2 = \frac{\sum_{i=1}^{N}\sum_{q=1}^{n}(Y_{iq} - Y)^2}{N(n-1)} \qquad (2.8)$$

where: i =1,2, ..., N
q =1,2,..., n

For two repeated experiments, formula 8 took the form:

$$S_Y^2 = \frac{2\sum_{i=1}^{n}\left(Y_{iq} - Y\right)^2}{N} = \frac{\sum_{i=1}^{N} S_i^2}{N} \qquad (2.9)$$

$$S_Y^2 = \frac{0.054}{8} = 0.007$$

We found the variance of adequacy:

$$S_{ad}^2 = \frac{n\sum(Y_{cp} - Y_{pac})^2}{N - q} \qquad (2.10)$$

where: $q = K + 1$;

K – number of regression coefficients.

$$S_{ad}^2 = \frac{0.0242}{4} = 0.0061$$

The adequacy of the model was checked using the Fisher criterion:

$$F_{экс} = \frac{S_{ad}^2}{S_Y^2} = \frac{0.0061}{0.007} = 0.87$$

F_{tab} (2.8) = 4,5 for $f_1 = 2, f_2 = 8$

In this case, $F_{ex} < F_{tab}$; 0.87 < 4.5; therefore, the model is adequate.

To check the significance of the coefficients (regression), you need to: find the variance of the regression coefficients S^2_{bi} using the formula:

$$S_{bi}^2 = \frac{S_y^2}{N} = \frac{0.007}{8} = 0.0009$$

Then construct a confidence interval; $\Delta b_i = tS_{bi.}$

Here: t is the table value of the Student's test for the number of degrees
freedoms with which S_{2y} was determined at the selected significance level (usually 0.05);

S_{bi} – square error of the regression coefficient.

$$S_{bi} = \pm\sqrt{S_{bi}^2} = \sqrt{0.0009} = 0.03$$

$$\Delta t_{кр} = 3.182$$

$$\Delta bi = t\,x\,S_{bi} = 3.182 \times 0.03 = 0.095$$

The coefficient is significant if its absolute value is greater than the confidence interval (Table 3).

Table 4. *Significance of coefficients*

b_i – values	Symbol	D	Condition values	Results
B_0=+6.125	>	0.095	pleasure	The coefficient is significant
b_1=+0.45	>	0.095	pleasure	The coefficient is significant
b_2=+0.048	>	0.095	pleasure	The coefficient is insignificant
b_3=+0.05	>	0.095	pleasure	The coefficient is insignificant

Conclusion

As can be seen from (table 4), factor X_1, turned out to be significant, which is understandable.

One of the tasks of optimizing the extraction process using the method of mathematical experimental planning is to quantify the contribution of each of the selected factors to the extraction result.

It was found that the main influence on the process of extraction of the above-ground part of the plant is exerted by factor X_1 – temperature.

A steep ascent was not carried out, since in experiment № 4 the yield obtained was quite acceptable at the first contact of the phases.

Thus, the research carried out using the method of mathematical experimental planning revealed the optimal conditions for extraction from raw materials at the first phase contact, namely extraction with 96% alcohol at a temperature of 35 °C, extraction time – 6 hours.

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