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Determination of the optimal storage zone of functional beverages based on sprouted grain extracts using mathematical models

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ABSTRACT

Beverages based on sprouted cereals are an excellent basis for creating new types of functional foods, as they are rich in nutrients. Beverages made from sprouted grains aim to improve daily nutrition, prioritising food safety. The proper storage of these drinks depends on the processing techniques used, including chemical preservatives and the conditions under which they are stored. Thus, using a mathematical model, this study aimed to determine the optimal storage zone of functional beverages from sprouted raw materials with preservatives. The results of our study showed that the optimum storage temperature and citric acid content of wheat extract were 2.9% and +11°C; barley 2.4% and 18°C; triticale 2.2% and +11°C; sunflower 2.8% and +14°C; rapeseed 2.7% and +16°C; safflower 2.3% and +17°C; flax 2.6% and +17°C; soya 2.4% and +18°C; pea 2.3% and +18°C; chickpea 2.3% and +18°C, respectively. Overall, these outcomes theoretically support the processing of beverages from sprouted grains. Thus, for practical application, it is recommended to implement controlled storage environments with the recommended temperatures and ensure that citric acid is correctly dosed at the identified optimal levels to enhance the shelf life of beverages.

Keywords: beverage, mathematical model, storage, shelf life

INTRODUCTION

The need to enrich products with biologically active substances and dietary fibre is a major prerequisite for developing food products that meet the needs of consumers. Sprouting, also known as germination, is a natural process often used for various grains, legumes, and seeds, and it offers several benefits for nutrition, flavour, and digestion **[1]**. Sprouts of wheat and other cereals contain fibres, vitamins of the B group, antioxidants, and macro- and microelements, which have a positive effect on human health **[2]**. The germination process initiates various transformations as it revitalises the seed's metabolism. This results in the degradation of macronutrients and antinutritional substances while stimulating the synthesis of secondary metabolites that could provide health benefits **[3]**. Beverages are an excellent basis for creating new types of functional products. Currently, sprouted wheat, barley, rye, corn, buckwheat, triticale, rice, and sorghum are used to manufacture functional beverages **[4]**, **[5]**, **[6]**. In Kazakhstan, while cereal crops are primarily used for bread production, the population's modern shift towards a healthy lifestyle and demand for nutritious products has prompted manufacturers to diversify their grain-based offerings.

The developed beverages are intended to enrich the daily diet, so food safety issues are the main prerequisite for their use. Processing methods influence the effective storage of beverages, the presence of chemical preservatives, storage conditions, temperatures and the packaging materials. It is well known that the thermal method is widely used to increase the shelf life of food products. However, the existing heat treatment methods reduce the nutritional value of food products **[7]**. Food spoilage and high preservation of food products are serious economic problems for producers, indicating the possibility of preserving food safety indicators through natural preservative application **[8]**.

Previous studies analysed changes in amylase activity, carbohydrate and during germination of domestic cereal grain varieties such as triticale, wheat and barley **[9]**; and protein-protease complex of leguminous crops in Kazakhstan **[10]**; as well as the technology of beverages based on extracts from sprouted grains and seeds **[11]**. However, no studies have been conducted on the optimisation of storage conditions of functional beverages based on these raw materials. Thus, using a mathematical model, this study aimed to determine the optimal storage zone of functional beverages from sprouted raw materials with preservatives.

Scientific Hypothesis

Mathematical modelling has identified the optimal storage zone for functional beverages made from sprouted raw materials with preservatives. The model predicts that specific temperature and citric acid concentration will maximise the beverage's shelf life.

MATERIAL AND METHODOLOGY

Samples

The research objectives are turbid liquids, close to emulsions, with suspensions products of hydrolysis of starchy part of cereals (wheat, triticale, barley), legumes (soya, pea, chickpea) and oilseeds (sunflower, rapeseed, flax, safflower) (Figure 1). Further details about the conditions for sprouting grains and oilseeds and the microbiological assessment can be found in our earlier research **[12]**. Muslimov et al. (2023) **[13]** provide information on extraction technology.

Chemicals

Sodium hydroxide (purity ≥ 99.9%) (Topan, Kazakhstan), phenolphthalein (purity ≥ 98.0%), citric acid (purity ≥ 99%) (Abris+, Russia), distilled water (purity ≥ 99%) (Terranova, Kazakhstan). All chemicals were of analytical grade quality.

Instruments

Burettes, flasks, laboratory glass pipettes, glasses, glass funnels (Kazlabpribor, Kazakhstan), electric heating plate (Tomanalit, Kazakhstan), and stopwatch (LabTime, Kazakhstan).

Laboratory Methods

The shelf life of functional beverages was determined using extracts from sprouted grains and oilseeds according to the requirements of Methodical Instructions (MUK) 4.2.1847-04 **[14]**. Next, the acidity was determined following GOST 6687.4-86 **[15]**.

According to GOST 28188-2014, the suggested shelf life for soft drinks is as follows: when stored at temperatures between 0 and +18 °C, drinks made without preservatives should be consumed within 30 days of production. For those that contain preservatives or are pasteurised, the shelf life can extend to a maximum of 6 months. This research examined two beverage storage methods utilising extracts from sprouted cereal grains, legumes, and oilseeds: one method involved no preservatives, while the other included citric acid as a preservative. Experimental studies investigated the preservation of functional drinks for 6 months from 14.03.2023 to 10.09.2023 (180 days).

According to GOST 28188-2014 “Soft drinks. General technical conditions,” acidity is one of the main quality indicators of beverages based on plant raw materials. The core of the acid determination method involved titrating the research samples with an alkaline solution and storing them for varying durations. A minimum of two parallel measurements is required.

Description of the Experiment

Sample preparation: During 30 days, the functional beverages were stored at 18±2°C, with a humidity of not more than 75%.

Number of samples analyzed: 10
Number of repeated analyses: 30
Number of experiment replication: 3

Design of the experiment:

In the first stage, we investigated the acidity change during the storage of functional beverages based on the obtained extracts from sprouted grains. Then, we studied the process of storing objects using citric acid as a preservative. Methods of mathematical processing of statistical data were used to process the results of laboratory studies, after which a model in three-dimensional space was constructed. To determine the optimal zones of the storage process of functional beverages with the addition of preservatives, graphs with contours of the calculated response surface were plotted.

1) wheat 2) barley 3) triticale

4) sunflower 5) rapeseed 6) flax

7) safflower 8) soya 9) peas

10) chickpea

Figure 1 Extracts from different sprouted grains and seeds.

Statistical Analysis

Statistical processing of laboratory study results was carried out using mathematical methods. Calculations were performed using the Statgraphics Centurion software package (v19). Data were analysed by an analysis of variance (ANOVA) procedure. *P*-values are less than 0.05, indicating that they significantly differ from zero at the 95% confidence level.

When processing the experimental results and investigating the response functions, we use the second-order equation of the following form:

$$y = b_0 + \sum^{n} b_i\, x_i + \sum_{i<j}^{n} b_{ij}\, x_i x_j + \sum^{n} b_{ii} {x_i}^2 + \ldots,$$

Where: y is the estimated value of the optimisation criterion; x_1, x_2, ..., x_n - independent variables (factors).

The experiment utilised a central composite design (CCD 2^2+star) of the second order. This method was employed to develop a mathematical model for the storage process of functional beverages that incorporate preservatives. This experimental setup examined two factors (K=2), resulting in 13 experimental trials. Among these trials, 5 were conducted at the central point to provide a baseline, while 6 coefficients were used to formulate the regression equation. The analysis also revealed that there were 7 degrees of freedom available for estimating the residual variance, which is essential for assessing the accuracy and reliability of the model.

RESULTS AND DISCUSSION

Determination of the acidity with and without preservatives

A significant portion of food loss can be attributed to food degradation during storage, transportation, or processing **[16]**. Employing precise methodologies to forecast and compute the shelf life of products within particular environmental parameters can yield invaluable insights for optimising storage and distribution strategies pertinent to those products **[17]**, **[18]**.

Acidity significantly influences functional beverages' quality, flavour, and preservation. These beverages are often designed to provide health benefits beyond basic nutrition **[19]**. Table 1 presents the dynamics of acidity changes in beverages based on germinated grain extracts during storage. The data analysis indicates that the acidity decreased on average by 0.8-1.1 cm3 1 M NaOH solution/100 cm3 of the drink throughout the storage period.

Table 1 Changes in acidity during storage of functional beverages based on obtained extracts from sprouted grains.

Beverages based on extracts	**Storage time, days**					
	5	**10**	**15**	**20**	**25**	**30**
Wheat	2.4	2.3	2.1	2.0	1.7	1.5
Barley	2.5	2.3	2.1	1.9	1.8	1.6
Triticale	2.8	2.5	2.2	2.0	1.9	1.8
Sunflower	3.3	3.0	2.8	2.5	2.3	2.2
Rapeseed	3.2	3.1	2.9	2.6	2.4	2.3
Safflower	3.2	3.0	2.8	2.5	2.4	2.2
Falx	3.3	3.1	2.7	2.5	2.4	2.3
Soya	3.5	3.3	3.0	2.8	2.6	2.4
Peas	3.1	3.0	2.8	2.5	2.4	2.3
Chickpea	3.0	2.8	2.5	2.4	2.2	2.1

Meanwhile, citric acid is often added to beverages for its natural preservative properties. It helps prevent microbial growth and prolongs shelf life **[20]**. Table 2 exhibits the acidity changes by adding 1%, 2%, and 3% citric acid. Analysis of the presented data shows that with increasing storage time, the acidity values of the extracts decrease, which can be attributed to microbial metabolism and chemical reactions, such as the breakdown of certain compounds, which can lead to the production of alkaline byproducts, which can neutralise the acidity **[21]**. Generally, plant-based beverages tend to have a shorter shelf life than dairy products due to their high moisture content, processing conditions, and lower acidity levels **[22]**.

Table 2 Change of acidity with addition of preservatives.

Extracts	**Duration of storage, months**					
	1	**2**	**3**	**4**	**5**	**6**
Wheat	2.9	2.8	2.5	2.4	2.0	1.8
Barley	3.0	2.8	2.5	2.3	2.2	1.9
Triticale	3.4	3.0	2.6	2.4	2.3	2.2
Sunflower	4.0	3.6	3.4	3.0	2.8	2.6
Rapeseed	3.8	3.7	3.5	3.1	2.9	2.8
Safflower	3.8	3.6	3.4	3.0	2.9	2.6
Flax	4.0	3.7	3.2	3.0	2.9	2.8
Soybeans	4.2	4.0	3.6	3.4	3.1	2.9
Peas	3.7	3.6	3.4	3.0	2.9	2.8
Chickpea	3.6	3.4	3.0	2.9	2.6	2.5
with 2% citric acid						
Wheat	3.5	3.3	3.0	2.9	2.4	2.2
Barley	3.6	3.3	3.0	2.7	2.6	2.3
Triticale	4.0	3.6	3.2	2.9	2.7	2.6
Sunflower	4.8	4.3	4.0	3.6	3.3	3.2
Rapeseed	4.6	4.5	4.2	3.7	3.5	3.3
Safflower	4.6	4.3	4.0	3.6	3.5	3.2
Flax	4.8	4.5	3.9	3.6	3.5	3.3
Soybeans	5.0	4.8	4.3	4.0	3.7	3.5
Peas	4.5	4.3	4.0	3.6	3.5	3.3
Chickpea	4.3	4.0	3.6	3.5	3.2	3.0
with 3% citric acid						
Wheat	4.1	4.0	3.6	3.5	2.9	2.6
Barley	4.3	4.0	3.6	3.3	3.1	2.8
Triticale	4.8	4.3	3.8	3.5	3.3	3.1
Sunflower	5.7	5.2	4.8	4.3	4.0	3.8
Rapeseed	5.5	5.4	5.0	4.5	4.1	4.0
Safflower	5.5	5.2	4.8	4.3	4.1	3.8
Flax	5.7	5.4	4.7	4.3	4.1	4.0
Soybeans	6.0	5.7	5.2	4.8	4.5	4.1
Peas	5.4	5.2	4.8	4.3	4.1	4.0
Chickpea	5.2	4.8	2.5	4.1	3.8	3.6

Development of the mathematical model and evaluation of adequacy

Using optimization techniques, mathematical models can help identify the best combination of temperature and citric acid concentration that maximises the shelf-life or sensory qualities of the beverages. Previously, mathematical models were used to quantify functional beverage components, enabling a better understanding of how they interact and influence the final beverage product **[23]**, **[24]**. The interplay between acidity, storage duration, and temperature is vital in formulating and preserving functional beverages. Understanding these parameters is essential for manufacturers seeking to optimise flavour, enhance health benefits, and ensure product safety **[25]**.

Based on experimental studies of the storage process of functional drinks with the addition of preservatives, the following factors have been established: citric acid content x_1 (C, %) and temperatures x_2 (T, °C) influencing the optimisation criteria, including wheat extract acidity y_1, barley extract acidity y_2, triticale extract acidity y_3, sunflower extract acidity y_4, rapeseed extract acidity y_5, safflower extract acidity y_6, flax extract acidity y_7, soya extract acidity y_8, pea extract acidity y_9, chickpea extract acidity y_{10}. Next, we coded the intervals and levels of variation of input parameters, which are presented in Table 3. The planning matrix is presented in Table 4.

Table 3 Coding of intervals and levels of variation of input factors.

Factors		Variation levels					Variation intervals
Natural	**Encoded**	**-1.414**	**-1**	**0**	**+1**	**+1.414**	
Citric acid content, %	x_1	0.58	1	2	3	3.41	1
Storage temperature, °C	x_2	1.17	4	14	24	6.83	10

Table 4 Rotatable planning matrix of experimental studies of the storage process of functional beverages with the addition of preservatives.

Encoded values		Natural values		Experimental values									
x_1 2	x_2 3	C. % 4	T. °C 5	y_1 6	y_2 7	y_3 9	y_4 10	y_5 11	y_6 12	y_7 13	y_8 14	y_9 15	y_{10} 16
0	-1.414	2.0	0	3.43	3.05	3.83	4.56	5.15	3.72	5.42	4.17	3.62	3.46
-1.414	0	0.58	14	2.97	3.01	3.40	4.34	4.81	3.80	5.13	4.20	3.70	3.60
0	0	2.0	14	3.97	4.00	4.48	5.15	5.29	5.11	5.54	5.58	5.01	4.83
-1	-1	1.0	4	3.17	3.20	3.62	4.98	4.25	4.06	4.66	4.48	3.96	3.85
0	0	2.0	14	3.94	3.90	4.37	5.09	5.15	4.98	5.42	5.45	4.88	4.71
0	0	2.0	14	3.96	4.00	4.48	5.07	5.22	5.11	5.48	5.58	5.01	4.83
+1	-1	3.0	4	3.87	3.80	4.26	5.21	4.81	4.53	5.13	5.31	4.75	4.58
+1	+1	3.0	24	3.57	3.60	3.62	4.95	5.15	4.74	5.42	5.03	4.48	4.34
0	+1.414	2.0	28	2.95	4.30	3.40	4.75	5.00	4.90	5.42	6.00	5.40	5.20
0	0	2.0	14	3.94	4.10	4.58	5.21	5.22	5.04	5.48	5.72	5.14	4.95
-1	+1	1.0	24	3.22	3.10	3.51	4.46	4.74	3.93	5.07	4.34	3.83	3.72
+1.414	0	3.41	14	4.11	3.70	3.83	5.12	5.36	4.32	5.59	5.17	4.62	4.46
0	0	2.0	14	3.95	3.90	4.37	5.09	5.22	4.88	5.48	5.45	4.88	4.71

Note: x_1: citric acid content (C, %); x_2: temperatures (T, °C); y_1: wheat extract acidity; y_2: barley extract acidity; y_3: triticale extract acidity; y_4: sunflower extract acidity; y_5: rapeseed extract acidity; y_6: safflower extract acidity; y_7: flax extract acidity; y_8: soya extract acidity; y_9: pea extract acidity; y_{10}: chickpea extract acidity.

Tables 5-14 summarise the results of the analysis of variance for acidity.

Table 5 Analysis of variance (ANOVA) for wheat extract acidity.

Mean	Sum of squares	Degree of freedom	Mean square	F-value	*P*-value
x_1	0.885914	1	0.885914	77.24	0.0000
x_2	0.10784	1	0.10784	9.40	0.0182
x_1^2	0.232648	1	0.232648	20.28	0.0028
$x_1 x_2$	0.030625	1	0.030625	2.67	0.1463
x_2^2	0.890953	1	0.890953	77.68	0.0000
Lack of fit	0.0802885	7	0.0114698	-	-
Pure error	2.12689	12	-	-	-
Total (corr.)	0.885914	1	0.885914	77.24	0.0000

The ANOVA analyses the variability in the acidity of wheat extract by dividing it into distinct components associated with each factor (see Table 5). It subsequently assesses the statistical significance of these factors by comparing the root mean square (RMS) value to the estimated experimental error. The R^2 statistic indicates that the fitted model accounts for 96.2251% of the variability in the acidity of wheat extract, indicating that most of the differences in acidity can be attributed to the factors included in the model. This suggests that the model provides an excellent fit. The adjusted R^2 of 93.53% is ideal for comparing models with varying numbers of independent variables. The standard error was 0.1071, indicating the standard deviation of the residuals. The mean absolute error (MAE) of 0.0571 represents the average of the residuals. The Durbin-Watson (DW) statistic assesses whether there is a significant correlation among the residuals based on their order in the dataset, with a *P*-value exceeding 5%, suggesting no serial autocorrelation at the 5% significance level. The ANOVA results show that all factors are statistically significant ($P<0.05$) except for the interaction term x_1x_2 ($P>0.05$).

Table 6 Analysis of variance (ANOVA) for barley extract acidity.

Mean	Sum of squares	Degree of freedom	Mean square	F-value	*P*-value
x_1	0.538619	1	0.538619	76.95	0.0009
x_2	0.269291	1	0.269291	38.47	0.0034
x_1^2	0.780695	1	0.780695	111.53	0.0005
$x_1 x_2$	0.0025	1	0.0025	0.36	0.5823
x_2^2	0.213044	1	0.213044	30.43	0.0053
Lack of fit	0.552585	3	0.184195	-	-
Pure error	0.028	4	0.007	-	-
Total (corr.)	2.29371	12	-	-	-

In Table 6, the R-squared value reveals that the model accounts for 94.69% of the variability in acidity, indicating a strong fit. The adjusted R^2 is 56.61%. The standard error of the estimate is 0.083666. The MAE is 0.167304, providing another measure of model accuracy. The ANOVA results demonstrate that all factors are statistically significant except for the interaction term x_1x_2, which has an insignificant *P*-value, implying that this particular interaction does not explain the variability in acidity.

Table 7 Analysis of variance (ANOVA) for triticale extract acidity.

Mean	Sum of squares	Degree of freedom	Mean square	F-value	*P*-value
x_1	0.230556	1	0.230556	21.76	0.0023
x_2	0.23056	1	0.23056	21.76	0.0023
x_1^2	1.03716	1	1.03716	97.89	0.0000
$x_1 x_2$	0.070225	1	0.070225	6.63	0.0368
x_2^2	1.03716	1	1.03716	97.89	0.0000
Lack of fit	0.0741665	7	0.0105952	-	-
Pure error	2.44049	12	-	-	-
Total (corr.)	0.230556	1	0.230556	21.76	0.0023

The R^2 statistic indicates that the above model explains 96.96% of the variability in acidity of triticale extract (Table 7). The adjusted R^2 is 94.7903%. The standard deviation of the residuals is 0.102933. The MAE is 0.0687698.

Table 8 Analysis of variance (ANOVA) for the acidity of sunflower extract.

Mean	Sum of squares	Degree of freedom	Mean square	F-value	*P*-value
x_1	0.415455	1	0.415455	125.14	0.0004
x_2	0.0326788	1	0.0326788	9.84	0.0349
x_1^2	0.1445	1	0.1445	43.52	0.0027
$x_1 x_2$	0.0169	1	0.0169	5.09	0.0870
x_2^2	0.22948	1	0.22948	69.12	0.0011
Lack of fit	0.241928	3	0.0806427	-	-
Pure error	0.01328	4	0.00332	-	-
Total (corr.)	1.05237	12	-	-	-

In Table 8, the R^2 statistic reveals that the fitted model accounts for 95.7492% of the variability in the acidity of sunflower extract. The adjusted R^2 statistic stands at 58.4272%. The standard error of estimation indicates that the standard deviation of the residuals is 0.0576194. Meanwhile, the MAE is 0.105824. The ANOVA results show that all factors are statistically significant except for x_1x_2 ($P > 0.05$).

Table 9 Analysis of variance (ANOVA) for the acidity of rapeseed extract.

Mean	Sum of squares	Degree of freedom	Mean square	F-value	*P*-value
x_1	0.381857	1	0.381857	155.86	0.0002
x_2	0.0477198	1	0.0477198	19.48	0.0116
x_1^2	0.163111	1	0.163111	66.58	0.0012
$x_1 x_2$	0.005625	1	0.005625	2.30	0.2043
x_2^2	0.173937	1	0.173937	70.99	0.0011
Lack of fit	0.374984	3	0.124995	-	-
Pure error	0.0098	4	0.00245	-	-
Total (corr.)	1.11817	12	-	-	-

In Table 9, the R^2 indicates that the above model explains 95.588% of the variability in acidity. The adjusted R^2 is 41.008%. The estimation's standard error shows that the residuals' standard deviation is 0.04975. The MAE is 0.11815. The results of the ANOVA indicate statistical significance of all factors except for x_1x_2, since the *P* value was insignificant ($P>0.05$).

Table 10 Analysis of variance (ANOVA) for acidity of safflower extract

Mean	Sum of squares	Degree of freedom	Mean square	F-value	*P*-value
x_1	0.507722	1	0.507722	53.84	0.0018
x_2	0.382273	1	0.382273	40.54	0.0031
x_1^2	1.40556	1	1.40556	149.05	0.0003
$x_1 x_2$	0.0289	1	0.0289	3.06	0.1549
x_2^2	0.732523	1	0.732523	77.68	0.0009
Lack of fit	0.3864	3	0.1288	-	-
Pure error	0.03772	4	0.00943	-	-
Total (corr.)	3.24883	12	-	-	-

Furthermore, the R^2 statistic shows that the fitted model explains 95.7492% of the variability in the acidity of safflower extract (Table 10). The adjusted R^2 statistic is 58.4272%. The standard error of estimation was 0.0576194. Additionally, the MAE is 0.105824, while the interaction term x_1x_2 was insignificant ($P>0.05$).

Table 11 Analysis of variance (ANOVA) for acidity of flax extract

Mean	Sum of squares	Degree of freedom e	Mean square	F-value	*P*-value
x_1	0.270309	1	0.270309	150.17	0.0003
x_2	0.0612497	1	0.0612497	34.03	0.0043
x_1^2	0.136348	1	0.136348	75.75	0.0010
$x_1 x_2$	0.0036	1	0.0036	2.00	0.2302
x_2^2	0.0841739	1	0.0841739	46.76	0.0024
Lack of fit	0.26964	3	0.0898798	-	-
Pure error	0.0072	4	0.0018	-	-
Total (corr.)	0.807908	12	-	-	-

The R^2 statistic indicates that the fitted model explains 95.7338% of the variability in acidity of flax extract (Table 11). The adjusted R^2 statistic is 41.2579%. The standard error of the estimation shows that the standard deviation of the residuals is 0.04264. The MAE is 0.107692. The results of the ANOVA indicate statistical significance of all factors except for x_1x_2, since the *P* value was insignificant ($P>0.05$).

Table 12 Analysis of variance (ANOVA) for acidity of soya extract.

Mean	Sum of squares	Degree of freedom	Mean square	F-value	*P*-value
x_1	1.0453	1	1.0453	82.76	0.0008
x_2	0.587531	1	0.587531	46.52	0.0024
x_1^2	1.4672	1	1.4672	116.17	0.0004
$x_1 x_2$	0.0049	1	0.0049	0.39	0.5671
x_2^2	0.467554	1	0.467554	37.02	0.0037
Lack of fit	1.15181	3	0.383937	-	-
Pure error	0.05052	4	0.01263	-	-
Total (corr.)	4.58851	12	-	-	-

The R² statistic shows that the model accounts for 93.7969% of the variation in acidity of soya extract (Table 12). The adjusted R^2 statistic is 55.0804%. The estimation's standard error shows that the residuals' standard deviation is 0.112383. The MAE is 0.237423. The results of the ANOVA indicate statistical significance of all factors except for x_1x_2, since the *P* value was insignificant (P>0.05).

Table 13 Analysis of variance (ANOVA) for acidity of pea extract.

Mean	Sum of squares	Degree of freedom	Mean square	F-value	*P*-value
x_1	0.939183	1	0.939183	79.39	0.0009
x_2	0.560367	1	0.560367	47.37	0.0023
x_1^2	1.29825	1	1.29825	109.74	0.0005
$x_1 x_2$	0.0049	1	0.0049	0.41	0.5549
x_2^2	0.459473	1	0.459473	38.84	0.0034
Lack of fit	1.07904	3	0.35968	-	-
Pure error	0.04732	4	0.01183	-	-
Total (corr.)	4.214	12	-	-	-

The R^2 statistic reveals that the fitted model accounts for 93.271% of the variability in acidity of pea extract (Table 13). The adjusted R-squared statistic is 54.1788%. The estimation's standard error shows that the residuals' standard deviation is 0.108766. The MAE is 0.229697. The results of the ANOVA indicate statistical significance of all factors, except for x_1x_2, since the *P* value was insignificant (P>0.05).

Table 14 Analysis of variance (ANOVA) for acidity of chickpea extract

Mean	Sum of squares	Degree of freedom	Mean square	F-value	*P*-value
x_1	0.823184	1	0.823184	81.67	0.0008
x_2	0.546392	1	0.546392	54.21	0.0018
x_1^2	1.1263	1	1.1263	111.74	0.0005
$x_1 x_2$	0.003025	1	0.003025	0.30	0.6130
x_2^2	0.443084	1	0.443084	43.96	0.0027
Lack of conformity	1.01048	3	0.336826	-	-
Pure error	0.04032	4	0.01008	-	-
Total (corr.)	3.83248	12	-	-	-

The R-squared statistic indicates that the fitted model explains 92.5818% of the variability in the acidity of chickpea extract (Table 14). The adjusted R-squared statistic is 52.9973%. The estimation's standard error shows that the residuals' standard deviation is 0.100399. The MAE is 0.219821. The results of the ANOVA indicate statistical significance of all factors except for x_1x_2 (P>0.05).

The interaction effects of the input factors are significant when the P-values from Tables 5-14 are compared with the corresponding regression coefficients in Table 15.

Table 15 Coefficients of regression equations of output parameters.

Coefficients	Optimisation criteria										
	y_1	y_2	y_3	y_4	y_5	y_6	y_7	y_8	y_9	y_{10}	y_{11}
b_0	1.771	1.451	2.725	1.682	4.005	3.647	2.018	4.13	2.011	1.598	1.617
b_1	1.188	1.634	1.562	1.899	0.713	0.884	1.931	0.786	2.247	2.12	1.969
b_2	0.106	-0.072	0.014	0.118	0.031	0.059	0.096	0.046	0.107	0.105	0.102
b_{11}	-0.183	-0.335	-0.377	-0.386	-0.144	-0.153	-0.45	-0.14	-0.459	-0.432	-0.402
b_{12}	-	-	0.015	-0.013	-	-	-	-	-	-	-
b_{22}	-0.004	-0.002	-	-0.004	-0.002	-0.002	-0.003	-0.001	-0.003	-0.003	-0.003

It follows from the analysis of variance that the influence of all remaining coefficients of the equation on the output parameter y is statistically significant (their corresponding values of significance levels P are less than 0.05). Consequently, the regression equation (mathematical model of the process) can be written in the following form:

$$y= b_0+ b_1x_1+ b_2x_2+ b_{12}x_1x_2+ b_{11}x_1^2+ b_{22}x_2^2$$

Thus, the regression equations for the process of storage of functional drinks with the addition of citric acid will be as follows:

$$y_1 = 1.771+1.188x_1+ 0.106x_2 - 0.183x_1^2- 0.004x_2^2$$

$$y_2= 1.451+1.634x_1-0.072x_2 - 0.335x_1^2- 0.002x_2^2$$

$$y_3= 1.682 +1.899x_1+ 0.118x_2-0.013x_1x_2- 0.386x_1^2- 0.004x_2^2$$

$$y_4= 4.005+0.713x_1+ 0.031x_2- 0.144x_1^2- 0.002x_2^2$$

$$y_5= 3.647+0.884x_1+ 0.059x_2- 0.153x_1^2+ 0.002x_2^2$$

$$y_6= 2.018 - 0.1574x_1+ 1.2616x_2- 0.001x_1^2+ 0.308x_2^2$$

$$y_7= 4.13+0.786x_1+ 0.046x_2- 0.14x_1^2- 0.001x_2^2$$

$$y_8= 2.011+2.247x_1+ 0.107x_2- 0.459x_1^2- 0.003x_2^2$$

$$y_9= 1.598+2.12x_1+ 0.105x_2- 0.432x_1^2- 0.003x_2^2$$

$$y_{10}= 1.617+1.969x_1+ 0.102x_2- 0.402x_1^2- 0.003x_2$$

The analysis of three-dimensional spatial models shows that the necessary values of the optimisation criterion y are achieved in the considered search area. This indicates that the variations of the initial factors in the planning of experiments are sufficiently taken into account. Figures 2-11 show graphical representations of dependence graphs.

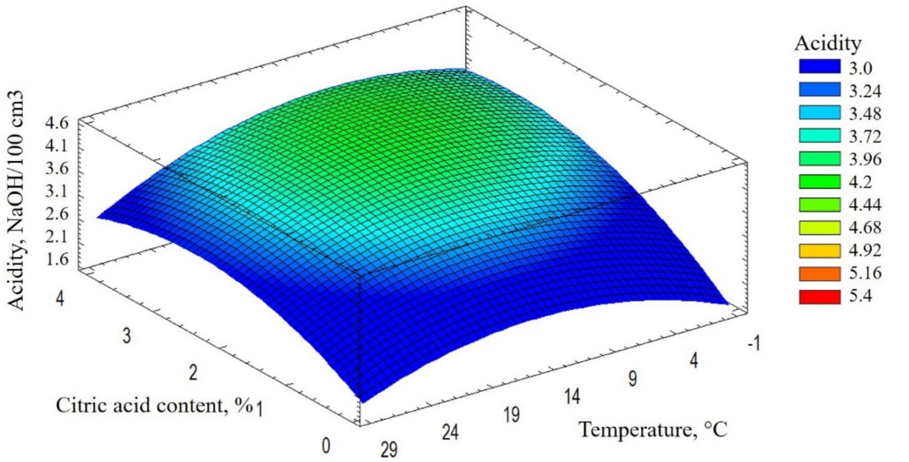

Figure 2 Three-dimensional space model characterising the dependence of $y_1=f(C, T)$ of citric acid content and storage temperature on wheat extract acidity.

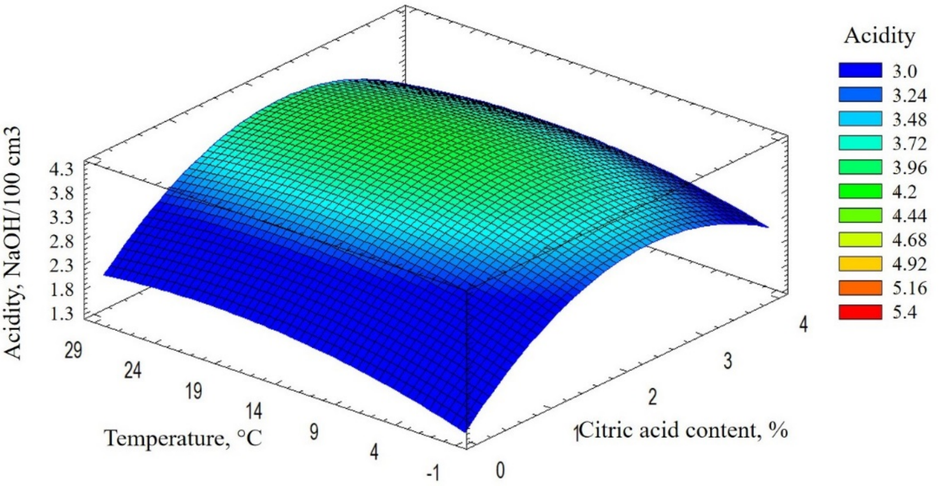

Figure 3 Three-dimensional space model characterising the dependence of $y_1=f(C, T)$ of citric acid content and storage temperature on barley extract acidity.

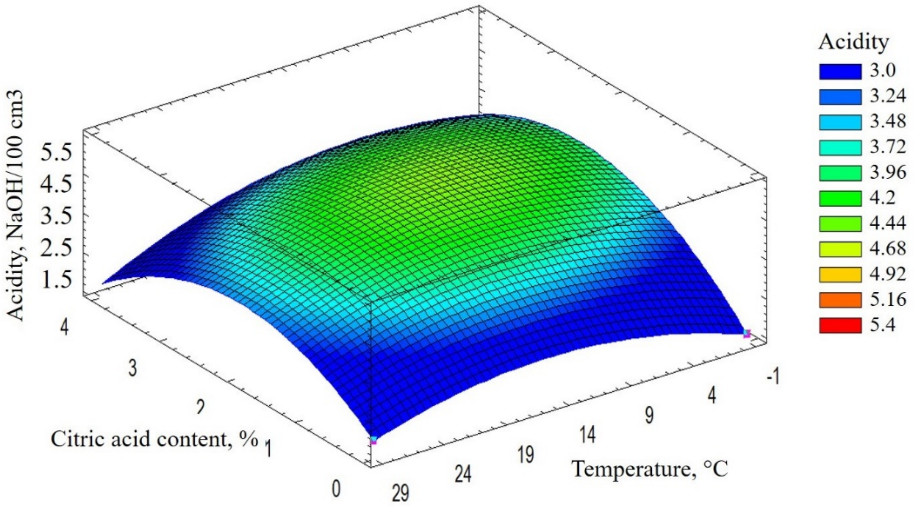

Figure 4 Three-dimensional space model characterising the dependence of $y_1=f(C, T)$ of citric acid content and storage temperature on triticale extract acidity.

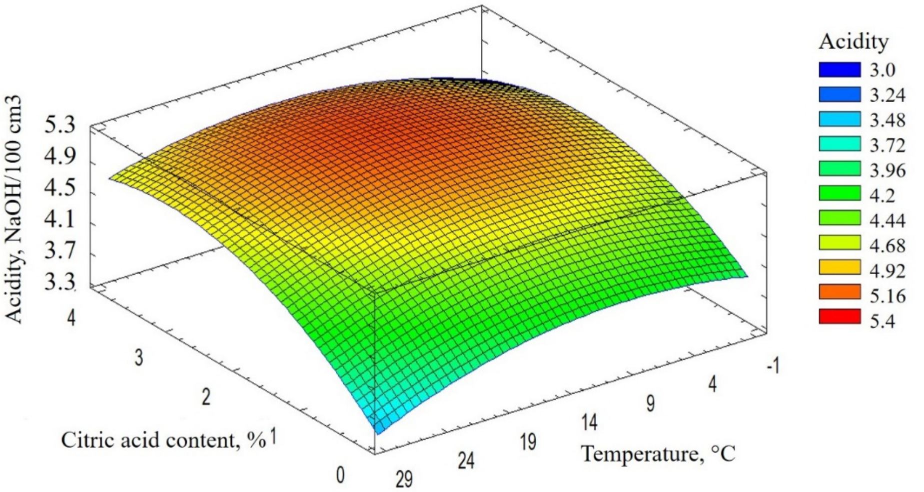

Figure 5 Three-dimensional space model characterising the dependence of $y_1=f(C, T)$ of citric acid content and storage temperature on sunflower extract acidity.

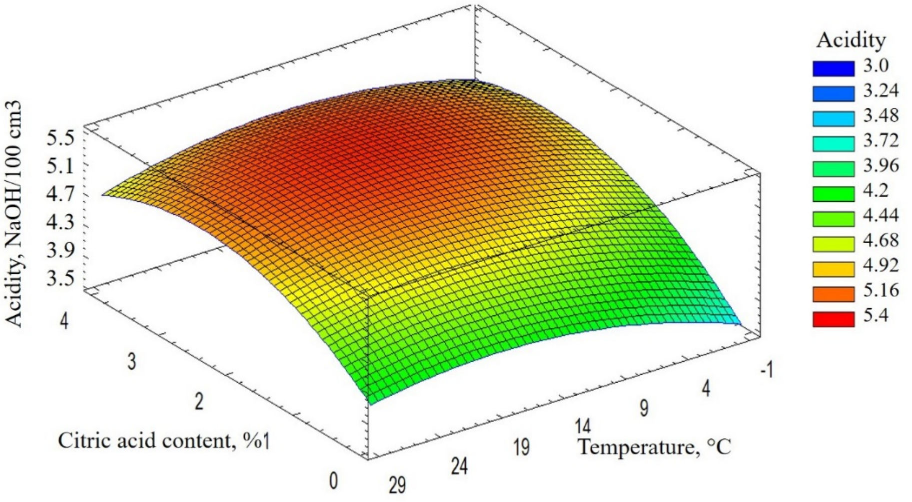

Figure 6 Three-dimensional space model characterising the dependence of $y_1=f(C, T)$ of citric acid content and storage temperature on rape extract acidity.

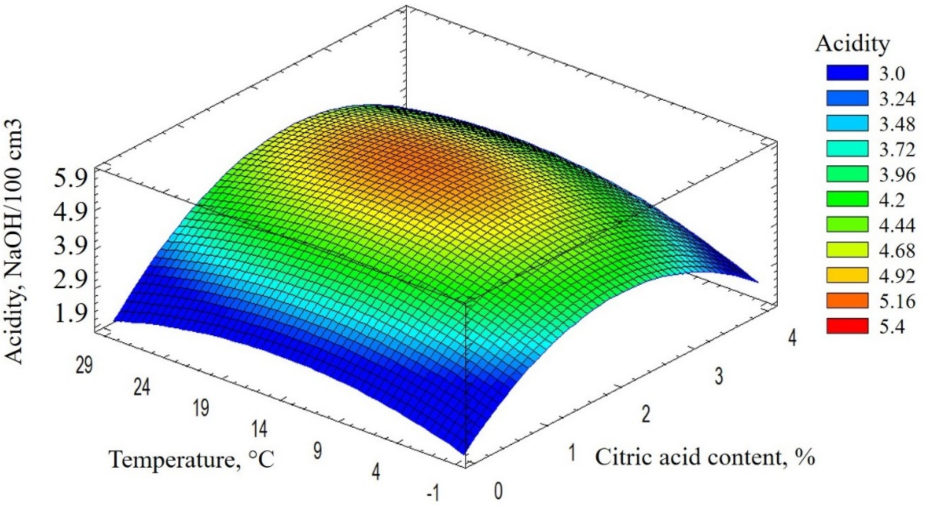

Figure 7 Three-dimensional space model characterising the dependence of $y_1=f(C, T)$ of citric acid content and storage temperature on safflower extract acidity.

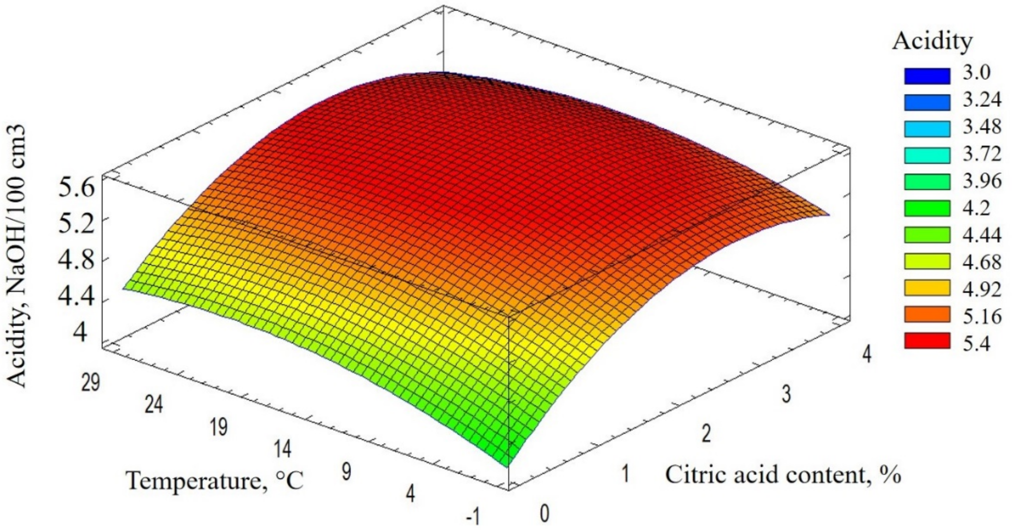

Figure 8 Three-dimensional space model characterising the dependence of $y_1=f(C, T)$ of citric acid content and storage temperature on flax extract acidity.

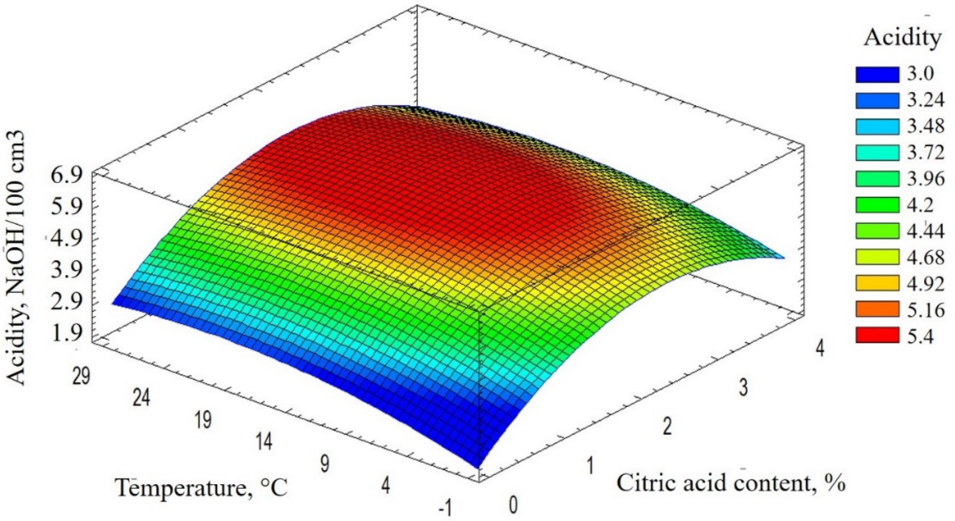

Figure 9 Three-dimensional model in space characterising the dependence of $y_1=f(C, T)$ citric acid content and storage temperature on soybean extract acidity.

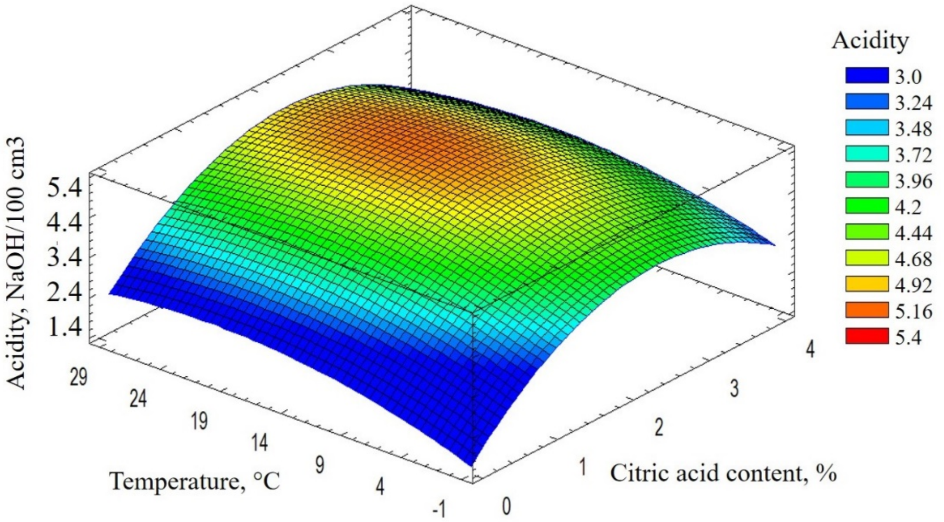

Figure 10 Three-dimensional space model characterising the dependence of $y_1=f(C, T)$ of citric acid content and storage temperature on pea extract acidity.

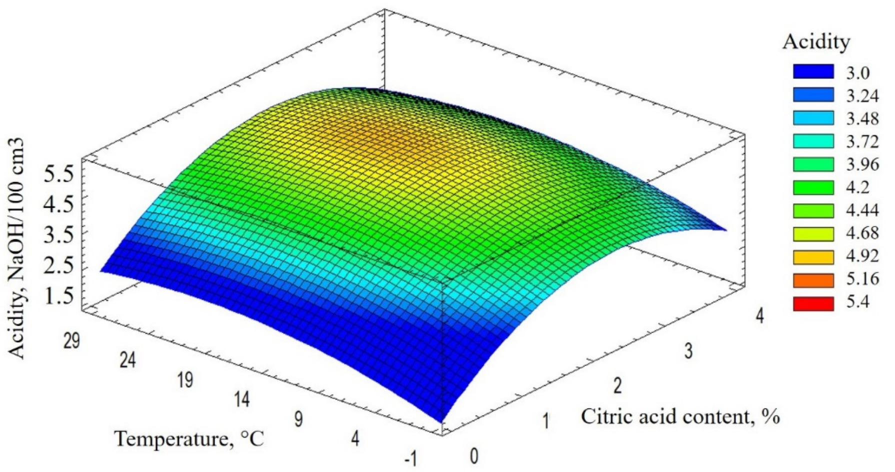

Figure 11 Three-dimensional space model characterising the dependence of $y_1=f(C, T)$ of citric acid content and storage temperature on chickpea extract acidity.

To determine the optimum zones for storing functional beverages with the addition of a preservative, graphs with contours of the calculated response surface were plotted, which are presented in Figures 12-21.

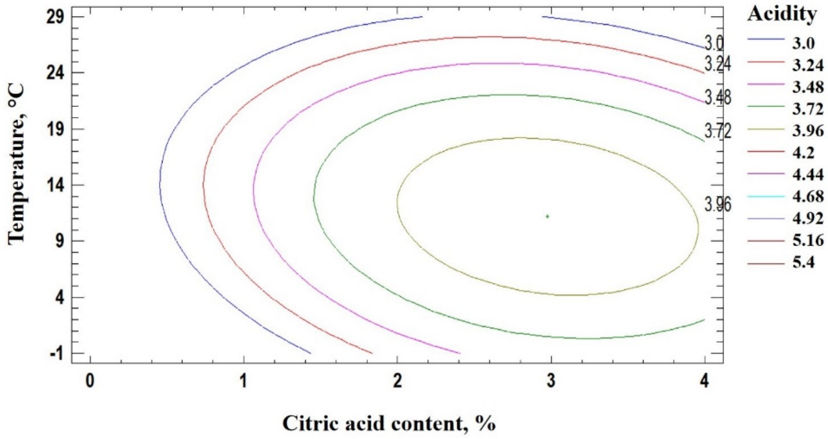

Figure 12 Contours of the calculated response surface characterising the dependence of citric acid content and storage temperature on wheat extract acidity.

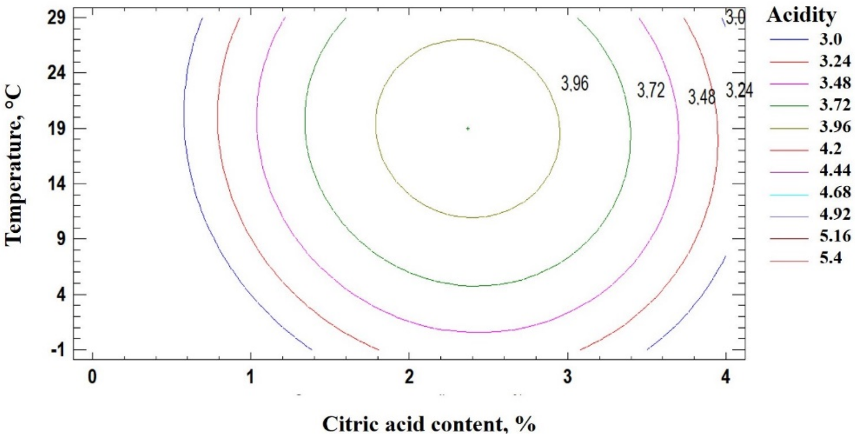

Figure 13 Contours of the calculated response surface characterising the dependence of citric acid content and storage temperature on barley extract acidity.

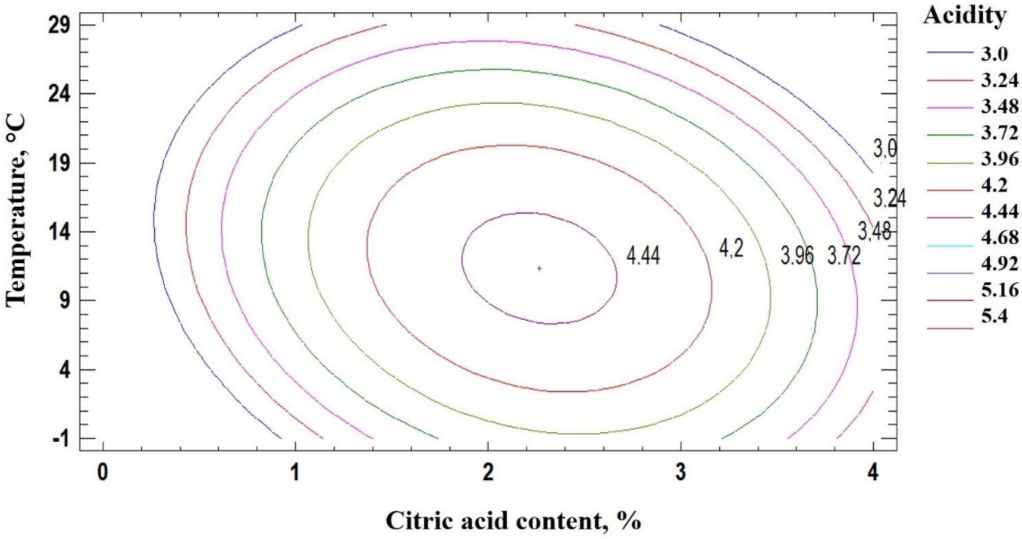

Figure 14 Contours of the calculated response surface characterising the dependence of citric acid content and storage temperature on triticale extract acidity.

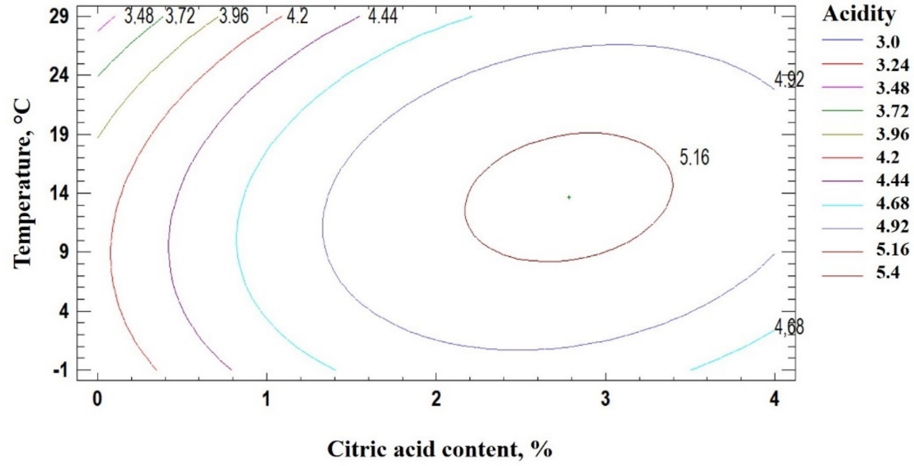

Figure 15 Contours of the calculated response surface characterising the dependence of citric acid content and storage temperature on sunflower extract acidity.

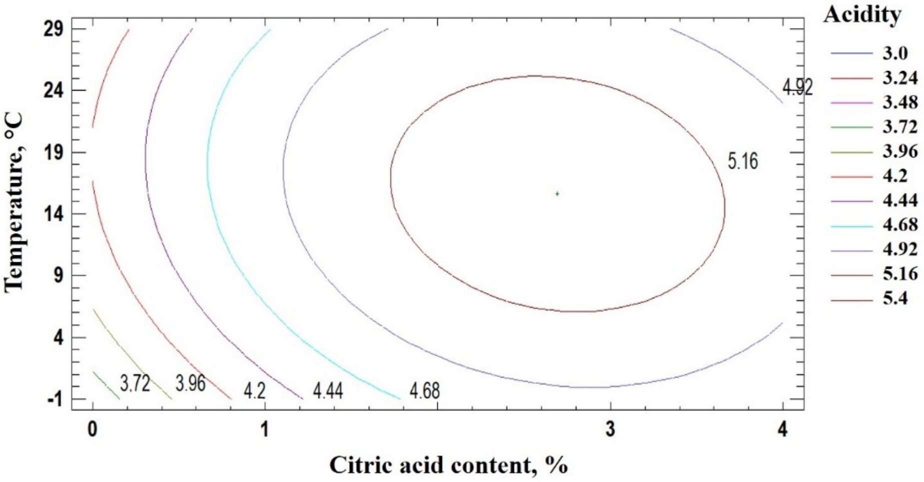

Figure 16 Contours of the calculated response surface characterising the dependence of citric acid content and storage temperature on rapeseed extract acidity.

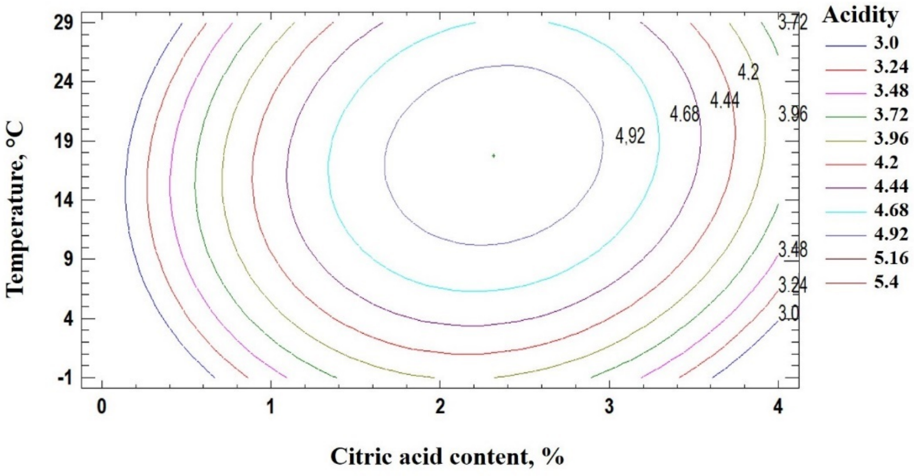

Figure 17 Contours of the calculated response surface characterising the dependence of citric acid content and storage temperature on the acidity of the safflower extract acidity.

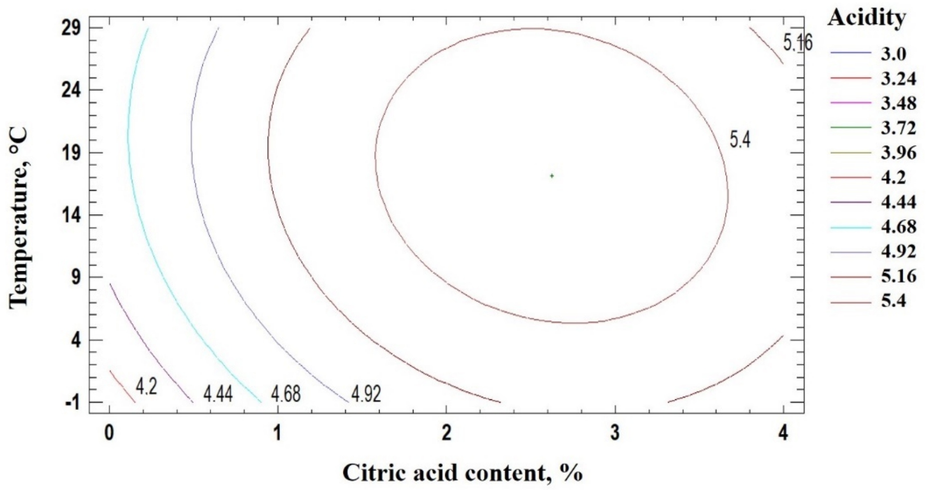

Figure 18 Contours of the calculated response surface characterising the dependence of citric acid content and storage temperature on flax extract acidity.

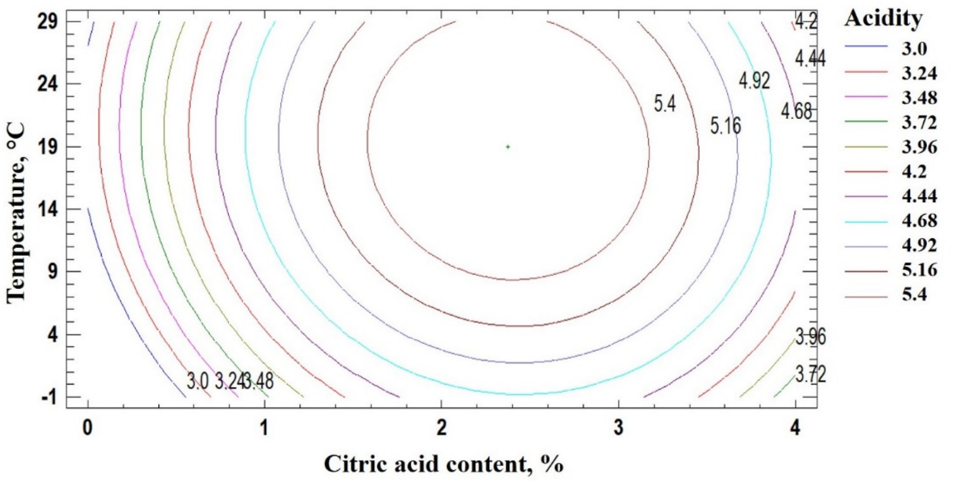

Figure 19 Contours of the calculated response surface characterising the dependence of citric acid content and storage temperature on soybean extract acidity.

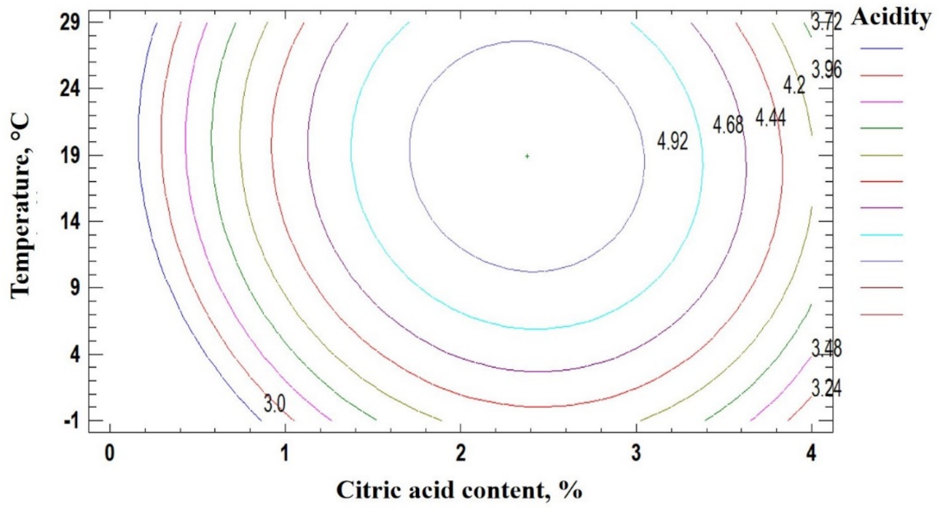

Figure 20 Contours of the calculated response surface characterising the dependence of citric acid content and storage temperature on the acidity of the pea extract.

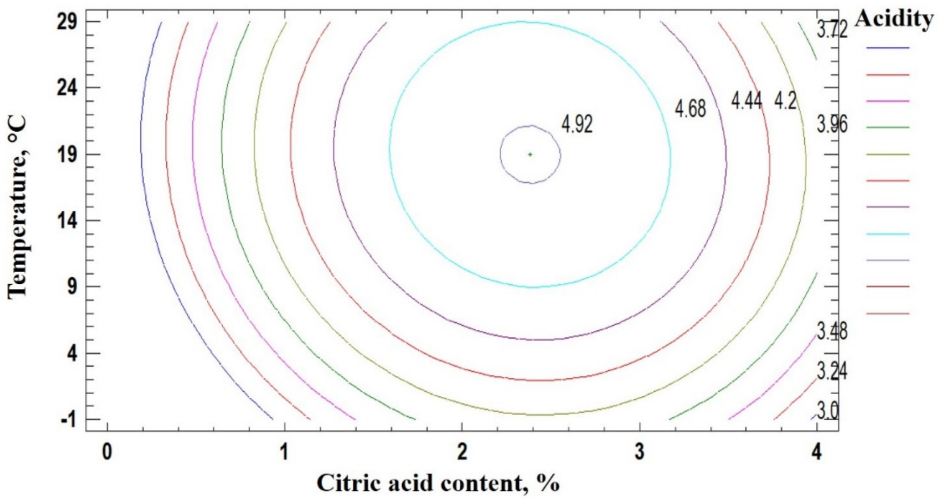

Figure 21 Contains the calculated response surface characterising the dependence of citric acid content and storage temperature on chickpea extract acidity.

It was established that the optimum zone of storage of research objects with the use of preservatives for wheat extract, which is achieved when the content of citric acid and storage temperature was 2.9% and +11°C; for barley extract was 2.4% and 18°C; for triticale, the extract was 2.2% and +11°C; for sunflower, the extract was 2.8% and +14°C; for rapeseed, the extract was 2.7% and +16°C; for safflower, the extract was 2.3% and +17°C; for flax, extract was 2.6% and +17°C; for soya, extract was 2.4% and +18°C; for pea, extract was 2.3% and +18°C; for chickpea extract was 2.3% and +18°C, respectively.

Response surface methodology (RSM) is an essential statistical and mathematical technique for optimising processes and understanding the relationships between several explanatory variables and one or more response variables **[26]**. In studying the shelf life of beverages, RSM offers advantages. Although many studies are using mathematical models analysing different beverages **[27]**, **[28]**, **[29]**; grains **[30]**, **[31]**, **[32]**, **[33]** and food products **[34]**, **[35]** from different aspects, there remains a notable deficiency in scholarly inquiry focused specifically on the optimal storage conditions for non-alcoholic functional beverages based on germinated grain extracts. For example, it was noted that soybeans harvested with a moisture content of 23%, dried at 80 °C, and stored at temperatures below 23 °C preserved their oil content (25.89%), crude protein (35.69%), and lipid acidity (5.54 mL), as demonstrated through mathematical modelling and multivariate analysis **[36]**.

Furthermore, the regression equation developed has been chosen as the mathematical model for predicting the shelf life of the functionally enriched sugarcane juice based on the independent variables **[37]**. A binary logistic regression model has been created to predict the growth of spoilage microorganisms in craft beer, demonstrating strong goodness of fit and accurate predictions in prior research **[38]**. Additionally, there has been a concise overview of the application of mathematical models for estimating the shelf life of coffee when stored under accelerated conditions **[28]**. Storage studies showed that the germinated brown rice beverage has beneficial nutritional properties, including high levels of total phenolic content, γ-oryzanol, niacin and γ-Aminobutyric acid **[39]**. The beverage with three parts jujube concentrate, two parts water, and 0.1% citric acid demonstrated better sensory and microbial quality than the other samples **[40]**. It was observed that the functional multigrain probiotic drink can be kept in refrigeration (4°C) for a maximum of 2 weeks without any decline in quality **[41]**. These findings indicate that the optimal formulation can significantly enhance sensory and microbial quality attributes, extending the drink's usability.

In addition, the results of the ANOVA analysis indicate that the interaction terms between citric acid content and temperature (x_1x_2) are not statistically significant across several scenarios. While the non-significant interaction terms between citric acid content and temperature suggest a consistent independent effect of each variable, it is crucial to acknowledge the current model's limitations. One possible limitation is that the model may oversimplify the complex biological or chemical interactions occurring in the beverage matrix, for example, additional factors such as pH, sugar content, or the presence of other acids may influence the relationship between temperature, citric acid content, and sensory characteristics of beverages. In addition, the ranges of citric acid and temperature investigated may not reflect potential nonlinear effects or other interactions that may be relevant at different levels or combinations of these variables. Given these considerations, including more independent variables (e.g. pH or presence of other flavour compounds) in the analysis could provide a more complete model that takes into account interactions not assessed in this study. Also, more advanced statistical techniques, such as mixed-effects models, could account for individual variations and allow for a more thorough examination of interactions **[42]**. Additionally, we plan to enhance our model's reliability by conducting further experiments to assess the statistical significance of citric acid content and temperature interactions in different beverage types. By expanding our research to include external data, we hope to validate our existing model and provide more robust conclusions in our upcoming studies.

Current storage technologies may not adequately maintain the optimal conditions required for preserving products, particularly in less industrialized markets where traditional storage facilities often struggle to control humidity and temperature precisely **[43]**. This challenge is compounded by the significant financial investments that innovative storage techniques may require, creating barriers for smaller companies in the functional beverage industry. However, the findings from the present study offer promising advancements specifically tailored to the functional beverage sector, particularly concerning the storage of beverages derived from sprouted grains and oilseeds. Manufacturers can create products with longer shelf lives without compromising flavour or nutritional value by identifying optimal storage parameters such as the concentration of citric acid as a preservative and appropriate temperature ranges. This research supports the development of new functional beverages that emphasise the nutritious properties and stability of sprouted grains and highlights the economic benefits of effective storage parameters. Besides, implementing these optimised storage practices can reduce waste due to spoilage, lower logistics costs, and ultimately increase inventory turnover rates. Therefore, the insights gathered from this study could pave the way for smaller brands to enhance their competitive edge while fostering sustainable growth within the functional beverage market.

CONCLUSION

Collectively, a mathematical model has been developed in this study, which makes it possible to determine the optimal parameters of the process of storage of research objects. The study outcomes revealed the optimum zone of storage of functional beverages with the use of citric acid as a preservative for wheat (2.9% and +11°C), barley (2.4% and 18°C), triticale (2.2% and +11°C), sunflower (2.8% and +14°C), rapeseed (2.7% and +16°C); safflower (2.3% and +17°C), flax (2.6% and +17°C), soya (2.4% and +18°C), pea (2.3% and +18°C), chickpea (2.3% and +18°C) extracts. These results provide theoretical support for the storage of sprouted grain beverages. Thus, for practical application, it is recommended to implement controlled storage environments with the recommended temperatures and ensure that citric acid is correctly dosed at the identified optimal levels to enhance the shelf life of beverages. Additionally, monitoring microbial activity and sensory properties during storage can help ensure product quality and safety over time.

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