# Sensory and Chemical Evaluation of Lactic Acid-Fermented Cabbage-Onion Juices

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The cabbage juices with various addition of onion juice (0.05~%, 0.1~%, and 0.2~%) were inoculated by *Lactobacillus plantarum* CCM 7039 and fermented during 168 h at 21 °C. The most of lactic acid was produced in cabbage juice with 0.1 % addition of onion juice  $(11.60~\text{g dm}^{-3}\text{ in the } 168~\text{h}$  fermentation). The most of acetic acid was produced in the cabbage juice with 0.2 % addition of onion juice  $(4.80~\text{g dm}^{-3}\text{ in the } 168~\text{h}$  fermentation). It was found that cabbage juice with 0.1 % addition of onion juice was the most acceptable for consumers.

Principal component analysis selected the most important variables for analytical determination (pH, total acidity, and content of lactic acid) and odour and taste evaluation of cabbage-onion juices (descriptors of odour and taste: cabbage, acid, sweet, spicy, and onion).

The vegetable juices processed by the lactic acid fermentation introduce change in the beverage assortment [1].

The lactic acid fermentation of vegetable, applied as a preservation method for the production of finished and half-finished products, is again being ranked as an important technology and is being further investigated because of the growing amount of raw materials processed in this way in the food industry. The main reasons for this interest are the nutritional, physiological, and hygienic aspects of the process and their corresponding implementation and production costs [2]. In a lot of countries consumption of the lactic acid-fermented vegetable juices increases [3]. Lactic acid-fermented vegetable juices are produced mainly from cabbage, red beet, carrot, celery and tomato [4].

The lactic acid-fermented vegetable juices preserve themselves high proportion of protective substances of the previous raw material. During fermentation, a great amount of another substances useful for health is produced by lactic acid bacteria and by other microorganisms. These produce organic acids, aldehydes, flavour substances, some antibiotics and others, that reduce the risk of civilization illnesses and so contribute to the importance of this group of food for health [5].

The use of statistical methods of experimental design represents an effective approach to identify the most significant factors regarding the quality of products. Multivariate statistical methods are an important tool for identifying of analytical and sensory attributes of products [6]. Principal component analysis

is a useful technique in exploratory data analysis and has been widely employed in research into dairy products [7]. This method reduces the number of original variables into a fewer number of unobservable variables (principal components) that are linear combinations of the original ones [8]. Through these statistical analysis data interpretation is simplified, relationships between different variables may be investigated and the most important factor of variability in data can be detected [7].

The combination of experimental design and multivariate methods should, therefore, be a powerful tool for improving the process of product development [6].

The purpose of this study was to prepare fermented cabbage juices with various addition of onion juice and from the analytical and sensory point of view to choose the most acceptable juice for the assessors and to determine optimal time of fermentation. The results of analytical determination and sensory evaluations were evaluated by multivariate statistic methods: principal component analysis (PCA) and cluster analysis (CA). The aim of the application of PCA was to determine the most important analytical and sensory variables for evaluation of cabbage-onion juices.

## **EXPERIMENTAL**

The measurement of pH was performed using a LABOR-pH-meter CG-834 SCHOTT, Germany. For calibration of pH-meter buffers at pH  $4.008 \pm 0.01$  and  $6.865 \pm 0.01$  (25 °C) were used. The total acidity [9] was determined by the visual titration with a 0.1 M-

NaOH using phenolphthalein indicator and expressed as lactic acid. Determination of reducing sugars [9] was performed according to Schoorl. The nonreacted Cu<sup>2+</sup> was determined after formation of Cu<sub>2</sub>O. The KI was oxidized by CuSO<sub>4</sub> to I<sub>2</sub> that was determined by titration with Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub> [1, 9].

Measurement of organic acids [10, 11] (lactic, acetic, citric, and L-ascorbic acid) was realized on the isotachophoretic analyzer ZKI 01 Villa Labeco (Spišská Nová Ves) with double-line recorder and conductivity detector. The electrolytic system used for identification and determination of organic acids had the following composition: concentration of leading electrolyte HCl  $1 \times 10^{-2}$  mol dm<sup>-3</sup>, counterion 6-aminocapronic acid, pH 4.25, additive 0.1 % methylhydroxyethylcellulose, concentration of terminating electrolyte capronic acid  $5 \times 10^{-3}$  mol dm<sup>-3</sup>. The samples were analyzed at a driving current of 250  $\mu A$  in the preseparation column. Quantitative analysis was performed by calibration [12]. At isotachophoretic analysis of juices, each sample was analyzed three times and each result of analysis represents average value from 3 determinations of the same sample. Relative standard deviation from determination of lactic acid varied from 0.2~% to 3.4~% and from determination of acetic acid between 1.7 % and 4.4 %.

For evaluation of analytical and sensory results, the multivariate statistic methods PCA and CA were applied. The results were arranged into data matrix and analyzed by statistic program SGWIN (statgraphic for Windows), Version 3.0.

# Vegetable Juices

The fresh vegetables (cabbage and onion) were purchased in a local market in Slovakia. From the cabbage, the outer leaves were removed and the cabbage was chopped to small slices. The onion was skinned and cutted to slices. The juices were obtained by pressing and filtration of crushed vegetables. The cabbageonion juices were prepared by different addition of onion juice into cabbage juices. 0.05 % (samples A), 0.1 % (samples B), and 0.2 % (samples C) of onion juice was added into cabbage juices. After addition of onion juice, D-glucose and NaCl (to concentration 2 % and 0.5 % of these compounds in the juices) were added and juices were inoculated by Lactobacillus plantarum CCM 7039 (Faculty of Natural Sciences, Brno, Czech collection of microorganisms, Czech Republic) at concentration  $10^6$  CFU cm<sup>-3</sup>.

The initial concentration of microorganism was measured by plate count on pour plates (LS-agar, Imuna, Šarišské Michaľany). The pour plates were incubated for 48 h at 37 °C  $\pm$  0.1 °C and bacteria were counted (CFU cm<sup>-3</sup>).

The adjusted juices were placed into 250 cm<sup>-3</sup> sterile flasks (volume of juice in every flask was 150 cm<sup>3</sup>). Every flask represented a single sample. The

flasks were closed with sterile rubber stoppers to ensure anaerobic condition. The juices were fermented in thermostat at  $21\,^{\circ}\mathrm{C} \pm 0.1\,^{\circ}\mathrm{C}$  for 168 h. At individual time intervals, the single samples were refrigerated (at 24 h intervals) and the juices were taken for analytical determination (pH, total acidity, reducing sugars, organic acids) and sensory evaluation (colour, sediment, turbidity, appearance, odour, taste, acceptance of odour, acceptance of taste and flavour).

#### **Sensory Evaluation**

The samples were evaluated by 10 assessors. Before sensory evaluation, refrigerated samples were defrosted and heated to laboratory temperature and evaluated. The temperature of the evaluated samples was  $15-18\,^{\circ}\text{C}$ .

Turbidity and appearance were evaluated by a fivepoint intensity scale (1 – nonturbid, 5 – very strong turbid and 1 – nontypical, 5 – typical). The colour was evaluated by a six-point scale. For evaluation of odour and taste 100 mm graphical nonstructured abscissas with the description of extreme points were applied (minimal or maximal intensity of descriptors). For odour evaluation of cabbage-onion juices, the following descriptors were chosen: sweet, acid, cabbage, sharp, spicy, smelly, sweet-acid, onion, cabbage-onion. For taste evaluation, the following descriptors were chosen: sweet, acid, cabbage, salt, spicy, bitter, sharp, harmonic, sweet-acid, onion, and cabbage-onion. The evaluation of acceptance of odour, acceptance of taste and flavour was realized by 100 mm graphical nonstructured abscissas with the description of extreme points. Results of sensory evaluations represent average values from evaluation of 10 assessors. The assessors were before sensory analysis checked for primary sensory tests.

## RESULTS AND DISCUSSION

In the previous works we dealt with the lactic acid fermentation of cabbage [4] and cabbage-carrot [1] juices and their chemical determination and sensory evaluation. Several researchers [5, 13] described the positive healthy effects of onion. For this reason we fermented cabbage juices with various addition of onion juices (0.05 %, 0.1 %, 0.2 %).

Determination of analytical parameters: At the beginning of fermentation, the juices had the pH value between 6.10 and 6.15. The total acidity of juices was expressed as lactic acid and ranged from 1.50 g dm $^{-3}$  to 1.60 g dm $^{-3}$ . The content of reducing sugars expressed as glucose was between 63.81 g dm $^{-3}$  and 69.52 g dm $^{-3}$  and content of L-ascorbic acid ranged from 302.19 mg dm $^{-3}$  to 380.63 mg dm $^{-3}$ . The concentrations of organic acids were following: lactic acid from 0.24 g dm $^{-3}$  to 1.23 g dm $^{-3}$ , acetic acid from 0.26 g dm $^{-3}$  to 0.64 g dm $^{-3}$ , and citric acid from 3.04

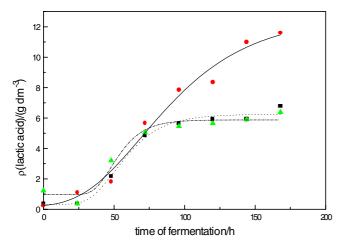


Fig. 1. Dependence between concentration of lactic acid and time of fermentation of cabbage-onion juices.  $\blacksquare$  0.05 %,  $\bullet$  0.1 %,  $\blacktriangle$  0.2 %.

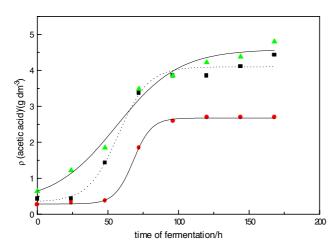


Fig. 2. Dependence between concentration of acetic acid and time of fermentation of cabbage-onion juices.  $\blacksquare$  0.05 %,  $\bullet$  0.1 %,  $\blacktriangle$  0.2 %.

**Table 1.** Parameters of Fitting Curves for Individual Lactic Acid-Fermented Cabbage-Onion Juices

Sample	Acid	$V_{\rm p}/({\rm g~dm^{-3}~h^{-1}})$	$C/(\mathrm{g~dm^{-3}})$	$M/\mathrm{h}$
A	Lactic	0.1208	5.9220	43.9240
	Acetic	0.0910	3.7176	46.1961
B	Lactic	0.1083	12.4334	54.1076
	Acetic	0.0771	2.4030	58.2534
C	Lactic	0.1454	4.9087	43.4836
	Acetic	0.0485	4.0503	37.4334

 ${\rm g} \ {\rm dm}^{-3} \ {\rm to} \ 3.49 \ {\rm g} \ {\rm dm}^{-3}.$ 

In course of fermentation, the pH value of juices and the content of reducing sugars decreased. The total acidity increased, lactic and acetic acids were produced by lactic acid bacteria and L-ascorbic and citric

acids were degraded. Degradation of L-ascorbic acid can be caused by chemical (minor content of oxygen or metals in raw material) or enzymatic oxidation (by enzymes naturally present in raw material). The citric acid can be degraded by several species of *Lactobacillus* genera to oxalacetic acid and acetic acid, by decarboxylation of oxalacetic acid, the pyruvic acid can be formed. The lactic acid and carbon dioxide are the end products of degradation of citric acid (reduction of pyruvic acid to lactic acid and carbon dioxide) [14].

At the end of fermentation, the juices had pH values between 3.60 (cabbage juices with 0.1 % addition of onion juice) and 3.85 (cabbage juices with 0.2 % addition of onion juice). The highest content increase of total acids and the highest content decrease of reducing sugars were recorded in the cabbage juice with 0.1 % addition of onion juice (increase about 7.7 times and decrease to the value  $45.0 \text{ g dm}^{-3}$ ).

The L-ascorbic acid is regarded as saving index of manufacture process [14]. According to Kopec [3], the lactic acid-fermented products contained from 20 % to 70 % of original content of L-ascorbic acid. At the end of fermentation, the cabbage-onion juices contained 48 % (cabbage juice with 0.05 % addition of onion juice), 53.4 % (cabbage juice with 0.1 % addition of onion juice), and 47.6 % (cabbage juice with 0.2 % addition of onion juice) from original amount of L-ascorbic acid contained in raw material. Content of reducing sugars at the end of fermentation ranged between 45.0 g dm $^{-3}$  and 57.0 g dm $^{-3}$ .

Figs. 1 and 2 represent changes of lactic and acetic acids in course of fermentation. At the end of fermentation, the juices contained following concentrations of organic acids: lactic acid from  $6.38~{\rm g~dm^{-3}}$  to  $11.60~{\rm g~dm^{-3}}$ , acetic acid between  $2.69~{\rm g~dm^{-3}}$  and  $4.80~{\rm g~dm^{-3}}$ , and citric acid from  $1.07~{\rm g~dm^{-3}}$  to  $1.61~{\rm g}$  $\mathrm{dm}^{-3}$ . The most of lactic acid was produced in cabbage juice with addition of 0.1 % onion juice ( $\rho =$  $11.60 \text{ g dm}^{-3}$  in the 168 h of fermentation). In this juice the highest utilization of reducing sugars by lactic acid bacteria (reduction from  $63.80 \mathrm{~g~dm^{-3}}$  to 45.0g dm<sup>-3</sup>) was recorded. These results are identical with the data obtained in course of lactic acid fermentation of cabbage-garlic juices (with the same additions of garlic juice) inoculated with the same Lactobacillus strain [15]. It was found that Lactobacillus plantarum CCM 7039 produced more of lactic acid than Lactobacillus plantarum 92H ( $\rho = 8.31 \text{ g dm}^{-3}$ ) that was applied for preparation of lactic acid-fermented cabbage juice prepared in the previous work [16]. The most of acetic acid was produced in cabbage juice with 0.2 % addition of onion juice ( $\rho = 4.80 \text{ g dm}^{-3}$  in the 168 h of fermentation). It was observed that production of acetic acid by Lactobacillus plantarum CCM 7039 was approximately to the same level as production of acetic acid during fermentation of pure cabbage juice inoculated by Lactobacillus plantarum 92H ( $\rho =$ 4.70 g dm<sup>-3</sup>) presented in the previous work [16]. De-

pendences in Figs. 1 and 2 were expressed by fitting curve running in program Origin 3.5 (Gompertz fitting curve) [17]

$$y = A + C \exp(-\exp(-B(x - M)))$$

where A is initial content of lactic or acetic acid (g dm<sup>-3</sup>), C is maximal content of lactic or acetic acid (g dm<sup>-3</sup>), B is tangent of curve in point M (growth rate in exponential phase), and M is time of maximal rate increase of product (h). Parameters of fitting curves for individual juices are presented in Table 1. The specific product formation rate  $V_P$  (g dm<sup>-3</sup> h<sup>-1</sup>) was calculated according to the equation:  $V_P = BC/e$  (e = 2.71828, Euler's number).

After fermentation, the juices were stored in the refrigerator at temperature  $9^{\circ}\text{C} \pm 0.1^{\circ}\text{C}$ . Karovičová et al. [16] demonstrated that this temperature inhibited growth of lactic acid bacteria. The juices prepared by lactic acid fermentation can be filled into bottles and pasteurized or aseptically filled into bottles after previous filtration [18].

Sensory evaluation: During fermentation, the colour, sediment, appearance, turbidity, odour, taste, acceptance of odour, acceptance of taste and flavour of juices were evaluated. The aim of sensory evaluation was to specify the most acceptable juice for consumers. For this reason, the harmonic taste was the most important sensory parameter. The harmonic taste introduces taste by which the other taste descriptors in the optimal proportions are characterized.

It was found that the cabbage juice with 0.1 %addition of onion juice had the highest intensity of harmonic taste in the 72 h of fermentation (64.4 %from scale), it was very strong turbid, had light-yellow colour, and typical appearance. The cabbage juice with 0.2 % addition of onion juice had the highest intensity of harmonic taste in the 48 h of fermentation (57.3 % from scale) it was soft turbid, had light-yellow colour with brown shade and typical appearance. The cabbage juice with 0.05 % addition of onion juice had the highest intensity of harmonic taste in the 72 h of fermentation (48 % from scale) it was turbid, had yellow-brown colour with green-brown shade and soft typical appearance. In these hours of fermentation, the juices had pH values 4.0, 3.9, and 3.5, respectively and the content of lactic acid (the main product of lactic acid fermentation) was 5.65 g dm<sup>-3</sup>, 3.21 g dm<sup>-3</sup>, and  $4.86 \text{ g dm}^{-3}$ .

Figs. 3 and 4 show the graphic diagram of sensory profile of juices in hours with the highest intensity of harmonic taste. From these diagrams differences in intensity of odour and taste descriptors in various samples can be determined. The graphic diagrams of sensory profile of juices showed that juices were distinguished mainly by intensity of sweet-acid, onion, and cabbage-onion odour and taste. Different intensity of onion and cabbage-onion odour and taste of

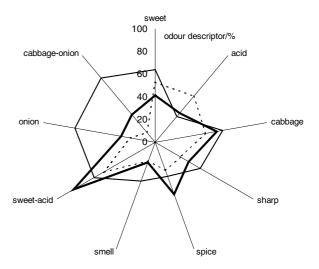


Fig. 3. Graphic diagram of sensory profile of odour for lactic acid-fermented cabbage-onion juices. --0.05% (72 h), -0.1% (72 h), -0.2% (48 h).

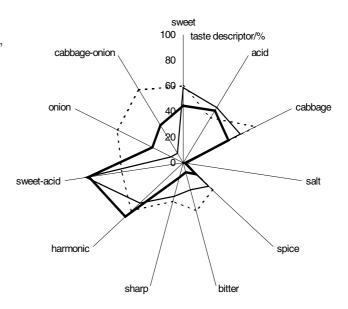


Fig. 4. Graphic diagram of sensory profile of taste for lactic acid-fermented cabbage-onion juices. --0.05~% (72 h), -0.1~% (72 h), -0.2~% (48 h).

juices was caused by various additions of onion juice into the cabbage juices.

Table 2 shows results from acceptance evaluation of odour, taste, and flavour in hours with the highest intensity of harmonic taste. Cabbage juices with 0.05~% and 0.1~% addition of onion juice had the highest intensity of these parameters in the 72 h of fermentation and cabbage juice with 0.2~% addition of onion juice in the 48 h of fermentation. The highest acceptance of odour, taste, and flavour was recorded in the cabbage juice with 0.1~% addition of onion juice (90.7 %, 93.2 %, and 91 % from scale).

The results of analytical determination and sensory evaluation were processed by multivariate statis-

58

Table 2. Acceptance of Odour, Acceptance of Taste, and Flavour Evaluation with the Highest Intensity of Harmonic Taste

Sample	Fermentation time with the highest intensity of harmonic taste	Acceptance of odour	Acceptance of taste	Flavour	
Sample	h	% from scale	% from scale	% from scale	
A	72	84.3	80.2	78.0	
B	72	90.7	93.2	91.0	
C	48	65.7	62.3	63.8	

**Table 3.** Component Loadings Expressed by the First Principal Component for Analytical Variables

Analytical variable	Component loadings expressed by the first principal component
Acetic acid Ascorbic acid Citric acid Lactic acid pH Reducing sugars Total acidity	0.3517 $-0.3359$ $-0.3922$ $0.3808$ $-0.4038$ $-0.3649$ $0.4044$

tic methods. The PCA was used for reduction of information of a large number of variables into a smaller set while losing only a small amount of information [19]. The purpose of CA was to join data into clusters in order to increase their withingroup homogeneity as well as the differences among the clusters and the individual groups [20].

Evaluation of analytical measurements: PCA (standardized input data) reduced seven original analytical parameters to one principal component that accounted for 81.45~% from the total variance. Table 3 presents component loadings expressed by the first principal component for analytical variables. The most notable variables were: pH, total acidity, and lactic acid.

At CA analysis, the distance between objects was measured as squared Euclidean distance and for dividing of juices into the groups, the group average method was used. CA divided samples into five groups that were characterized by similar analytical properties. In Fig. 5 five clusters of samples in the axes of two selected variables (pH and lactic acid, the most explained variables by PCA) are shown. Distribution of juices into clusters was as follows: Cluster 1 (samples A0, B0, C0, A24, C24) was characterized by the highest pH value (6.10—6.25) and the highest total acidity  $(1.50-1.60 \text{ g dm}^{-3})$ , cluster 2 (samples B24) and B48) was characterized by similar total acidity, cluster 3 (samples A48 and C48) was characterized by identical content of L-ascorbic acid (279.13 mg dm<sup>-3</sup>), cluster 4 (samples B144 and B168) was characterized by identical content of acetic acid  $(2.69 \text{ g dm}^{-3})$ , identical pH value (3.60), and the highest concentration of

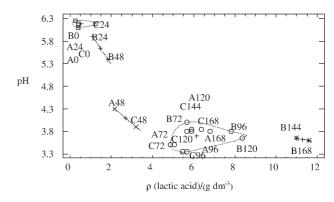


Fig. 5. Plotting of clusters of odour descriptors for lactic acidfermented cabbage-onion juices in coordinates of two selected variables. □ Cluster 1, × cluster 2, ○ cluster 3, + cluster 4, \* cluster 5, + centroids.

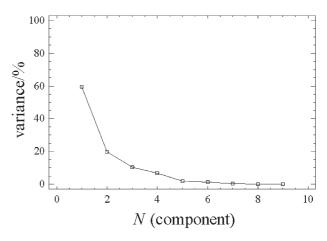


Fig. 6. Dependence between number of principal components and variance for odour descriptors of lactic acidfermented cabbage-onion juices.

lactic acid, cluster 5 (other samples) was characterized by high concentration of lactic acid and low pH values.

Odour evaluation: Fig. 6 shows dependence between numbers of principal components and percentage of explained variance. PCA extracted from original odour descriptors two independent components that explained 79.14 % from total variance. The first principal component (PC1) accounted for 59.89 % and the second principal component (PC2) accounted for 19.25 % from total variance of original data. Table 4 presents component loadings expressed by the

**Table 4.** Component Loadings Expressed by the First Two Principal Components for Odour Descriptors

Odour dogarintor	Component loadings expressed by the		
Odour descriptor	first principal component	second principal component	
Acid	0.41258	-0.0597	
Cabbage	-0.4252	0.0402	
Cabbage-onion	-0.2959	0.5112	
Onion	-0.3459	0.4295	
Sharp	0.1986	0.5139	
Smelly	0.2803	0.3638	
Spicy	0.3771	0.2729	
Sweet	-0.4132	-0.0134	
Sweet-acid	-0.1038	-0.2793	

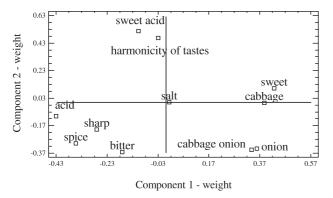


Fig. 7. Loadings of variables (taste descriptors) in coordinates PC1, PC2, and PC3.

first two principal components for odour descriptors. PC1 explained mostly the cabbage, acid, sweet, spicy, and onion odours and PC2 cabbage-onion, sharp, and onion odours. In case, when only the variables that were mostly explained by PC1 were retained the re-

distribution of original data was reached. In this instance was extracted one principal component that accounts for  $84.86\,\%$  from total variance of original data. From the obtained data it follows that using these variables is sufficient for odour evaluation of cabbageonion juices.

Taste evaluation: PCA reduced original taste descriptors to three independent components that explained 82.40 % from total variance of original data: PC1 43.4 %, PC2 20.30 %, and the third principal component (PC3) 18.70 %. Table 5 presents component loadings expressed by the first three principal components for taste descriptors. Fig. 7 shows component loadings in coordinates PC1 and PC2 for taste descriptors of cabbage-onion juices. PC1 describes mostly acid, sweet, onion, spicy, and cabbage tastes and PC2 the harmonic taste and sweet-acid taste. In case, when only the variables were retained that were mostly explained, redistribution of original data was reached. In this instance two principal components were extracted that accounts for 91.10 % from total variance of original data (PC1 73.30 % and PC2 17.80 %). From the obtained data it results that using these variables is sufficient for taste evaluation of cabbage-onion juices.

#### CONCLUSION

The consumption of lactic acid-fermented food should be increased in population. An important factor is also the supply of lactic acid-fermented vegetable juices with new quality characteristics, without application of preserving agents [3].

It was found that the cabbage juice with addition of 0.1 % of onion juice was the most acceptable from the analytical (recorded the highest concentration of lactic acid that is considered as a critical preservation factor for lactic fermentation) and sensory (the highest intensity of harmonic taste, acceptance of odour,

Table 5. Component Loadings Expressed by the First Three Principal Components for Taste Descriptors

T	Component loadings expressed by the		
Taste descriptor	first principal component	second principal component	third principal component
Sweet-acid	0.0929	0.5366	-0.2893
Onion	-0.3547	-0.3419	-0.1325
Cabbage-onion	-0.3353	-0.3472	-0.1282
Sweet	-0.4283	0.1068	-0.1396
Acid	0.4316	-0.1046	0.1266
Cabbage	-0.3887	0.0191	-0.2967
Salt	-0.0381	-0.0314	0.4187
Spicy	0.3551	-0.2682	-0.2237
Bitter	0.1762	-0.3052	-0.5273
Sharp	0.2712	-0.1511	-0.3864
Harmonic	-0.0008	0.5128	-0.3257

taste, and flavour) points of view. For reaching of optimal analytical and sensory attributes of end product it is required to stop fermentation in the 72 h of the process.

Application of principal component analysis selected the most important variables for analytical determination (pH, total acidity, and content of lactic acid) and odour and taste evaluation of cabbage-onion juices.

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