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DEVELOPMENT OF FERMENTED SAUSAGE PRODUCTION TECHNOLOGY USING STARTER CULTURES OF MICROORGANISMS

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This article considers the influence of microorganisms on the composition of semi-smoked sausages and technological processes. Liquid propionic acid bacteria were introduced into beef and their biological value, organoleptic and physicochemical parameters were studied. The influence of the use of starter microorganisms on tenderness, juiciness, nutritional value of the finished product, formation of the necessary level of structure and ability to retain moisture was studied, and also it was established that due to the action of starter microorganisms organoleptic indicators increase. As a result of experimental samples and researches the optimised formulation and technology of fermented semi-smoked sausages have been created.

Physico-chemical, organoleptic indicators, moisture-holding capacity, active acidity pH of the finished product and raw materials were determined. The method of production of fermented semi-smoked sausages is characterised by the introduction of starter culture into the raw material after the stage of meat grinding, before salting. The optimal ratio of liquid propionibacterium Propionibacterium shermani and Lactobacillus acidophilus, L.casei, L.Plantarum (2 strains of propionibacterium shermani and Lactobacillus acidophilus) is 0.1%, fermentation time - 8 hours.

Based on the results of experimental studies, we consider the use of liquid propionic acid bacteria 'ProBioLiz' (2 strains of propionic acid bacteria Propionibacterium shermani and lactobacillus).

Keywords: 1st and 2nd category beef, starter microorganisms, propionic acid microorganism, fermented beef, fermented half-sausages, amino acid composition.

СТАРТЕР МИКРОАҒЗАЛАРЫМЕН ӨҢДЕЛГЕН ФЕРМЕНТТЕЛГЕН ЖАРТЫЛАЙ ЫСТАЛҒАН ШҰЖЫҚ ЖАСАУ ТЕХНОЛОГИЯСЫН ӘЗІРЛЕУ

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Бұл мақалада микроорганизмдердің жартылай ысталған шұжықтардың құрамына және технологиялық процестеріне әсері қарастырылады. Сиыр етіне сұйық пропион қышқылы бактериялары қосылып, олардың биологиялық құндылығы, органолептикалық және физика-химиялық көрсеткіштері зерттелді. Стартерлік микроорганизмдерді қолданудың дайын өнімнің нәзіктігіне, шырындылығына, тағамдық құндылығына, құрылымның қажетті деңгейінің қалыптасуына және ылғалды сақтау қабілетіне әсері зерттелді, сонымен қатар әрекетке байланысты стартердің органолептикалық көрсеткіштері жоғарылайды екендігі анықталды. Эксперименттік үлгілер мен зерттеулер нәтижесінде ферменттелген жартылай ысталған шұжықтардың оңтайландырылған рецептісі мен технологиясы жасалды.

Дайын өнім мен шикізаттың физикалық-химиялық, органолептикалық көрсеткіштері, ылғал ұстау қабілеті, белсенді қышқылдығының pH-ы анықталды. Ферменттелген жартылай ысталған шұжықтарды өндіру әдісі етті ұсақтау кезеңінен кейін, тұздаудың алдында шикізатқа стартерлі микроағзаны енгізумен сипатталады. «ProBioLys» сұйық пропион қышқылы бактериясын (Propionibacterium

shermani және lactobacilli Lactobacillus acidophilus, L.casei, L.Plantarum пропион қышқылының 2 штаммы) пайдаланудың оңтайлы арақатынасы 0,1%, ферменттеу уақыты 8 сағат.

Эксперименттік зерттеулердің нәтижелеріне сүйене отырып, ысталған ет өнімдерінде сұйық пропион қышқылы бактериясы «ProBioLys» (Propionibacterium shermani және lactobacilli Lactobacillus acidophilus, L.casei, L.Plantarum пропион қышқылының 2 штаммы) сұйық пропион қышқылы бактериясын қолдану орынды деп санаймыз.

Түйін сөздер: 1 және 2 санаттағы сиыр еті, бастапқы микроорганизмдер, пропион қышқылы микроорганизмі, ферменттелген сиыр еті, ферменттелген жартылай шұжықтар, аминқышқыл құрамы.

РАЗРАБОТКА ТЕХНОЛОГИИ ПРОИЗВОДСТВА ФЕРМЕНТИРОВАННОЙ КОЛБАСЫ С ПРИМЕНЕНИЕМ СТАРТОВЫХ КУЛЬТУР МИКРООРГАНИЗМОВ

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В данной статье рассмотрено влияние микроорганизмов на состав полукопченых колбас и технологические процессы. В говядину вносили жидкие пропионокислые бактерии и изучали их биологическую ценность, органолептические и физико-химические показатели. Изучено влияние использования заквасочных микроорганизмов на нежность, сочность, пищевую ценность готового продукта, формирование необходимого уровня структуры и способность удерживать влагу, а также установлено, что за счет действия закваски повышаются органолептические показатели. В результате экспериментальных образцов и исследований создана оптимизированная рецептура и технология ферментированных полукопченых колбас.

Определены физико-химические, органолептические показатели, влагоудерживающая способность, pH активной кислотности готового продукта и сырья. Способ производства ферментированных полукопченых колбас характеризуется введением в сырье стартовую культуру после стадии измельчения мяса, перед посолом. Оптимальное соотношение использования жидкой пропионовокислой бактерии «ПроБиоЛиз» (2 штамма пропионовокислой бактерии Propionibacterium shermani и лактобациллы Lactobacillus acidophilus, L.casei, L.Plantarum) составляет 0,1%, время ферментации – 8 часов.

На основании результатов экспериментальных исследований мы считаем актуальным использование жидкой пропионовокислой бактерии «ПроБиоЛиз» (2 штамма пропионовокислой бактерии Propionibacterium shermani и лактобактерий Lactobacillus acidophilus, L.casei, L.Plantarum) в копченых мясных продуктах.

Ключевые слова: говядина 1-й и 2-й категории, стартовые микроорганизмы, пропионовокислый микроорганизм, ферментированная говядина, ферментированные полуколбасы, аминокилотный состав.

Introduction. The most important condition for increasing the range of sausage products is to reduce the cost of sausage products while maintaining standard quality. Currently, the best way to solve this problem is the introduction of new technologies that improve the biochemical changes occurring during the processing of meat raw materials. The use of microorganisms in the production of meat products helps to improve the quality of the finished product [1].

Currently, there are many large-scale theoretical and experimental materials on the use of sourdough starters in sausage production, including the study of probiotic microorganisms of scientific interest. Probiotic microorganisms include bifido and

propionic acid bacteria. Propionic acid bacteria grow at low temperatures, produce odorous, antimutagenic substances, vitamin B12, amino acids, and are highly active against pathogenic microflora.

Fermented sausages are produced at a constant temperature and, depending on the relative humidity, depending on the change in microbiological, biochemical and physical processes during ripening.

During the ripening of sausages, proteins are broken down into oligopeptides and free amino acids by the formation of histenzyme and aminopeptidase, which has a very good effect on the flavour of the finished product [2].

The aim of the work was to study the biochemical activity of propionic acid bacteria at the stage after treatment of raw materials with starter microorganisms at the stages of the technological process of production of semi-smoked sausages.

Propionic acid and bifidobacteria are very widely used in food production. Therefore, it is important to determine the odour-correcting and biochemical activity of the bacteria. In the works of Khanhalaev I.A. and I.S. Khamagaeva the influence of propionic acid and bifidobacteria on the finished sausage product was considered and it was proved that organoleptic parameters are significantly improved [3].

In their work, P. Nedelsheva and D. Zenkova proved that the addition of probiotic strain Lactobacillus Plantarum to the raw material provides mass and reduction of pathogenic flora in raw smoked sausage due to the necessary fermentation process [4].

In their workM. Laranjo and M.E. Potes suggested that propionic acid bacteria increase the safety of fermented meat products by rapidly acidifying the matrix and producing antimicrobial agents such as bacteriocins [5].

In his works S.E. Shokesheva, Y.M. Uzakov studied the effect of starter microorganisms on the acceleration of ripening of finished meat products and microstructure of finished sausage [6].

Materials and methods. Scientific and experimental work was carried out in the experimental shop for processing of meat products. As a material for experiments were taken experimental samples of semi-smoked sausage made on the basis of standard GOST 31785-2012. ST RK 1731-2007 Determination of organoleptic quality indicators of meat and meat products was carried out.

Acidity pH and water activity in sausage was measured using a digital pH-meter. The sample was chopped and mixed by passing it twice through a meat grinder. The prepared solution (1:10 ratio with distilled water) was determined from the mixture obtained after settling at 20°C for 30 minutes. The study was repeated 5 times. The total value is obtained from the method of variance calculation.

The ability to bind moisture was determined by the pressing method, based on the separation of water from the test sample during pressing, sorption of the separated moisture with filter paper and determination of the amount of water. The study was carried out by repeating 5 times by the area of the spot left on the filter paper. The total value is obtained based on the method of variance calculation.

Counting the number of mesophilic aerobic and facultative-anaerobic microorganisms in the prepared samples and the number of colonies in Petri dishes was carried out in accordance with ST GOST R 51448-2010 of the Republic of Kazakhstan. This study was carried out after the manufacture of samples and after expiry date.

Liquid propionic acid bacteria ‘ProBioLyz’ (2 strains of propionic acid bacteria Propionibacterium shermani and lactobacillus Lactobacillus acidophilus, L.casei, L.Plantarum) 0.08%, 0.1%, 0.15% per 100 kg were used as a starter.

The technological process was carried out in accordance with the technological instructions in compliance with the rules for meat industry enterprises.

The method of production of fermented semi-smoked sausages was characterised by the introduction of starter microorganisms into the raw material after the stage of meat grinding, before

salting. Beef semi-smoked sausage was chosen as the basic recipe (Table 1).

Table 1 - Recipe for fermented semi-smoked sausage

Raw materials	Beef sausage	100 кг/кг		
		Sample 1	Sample 2	Sample 3
beef 1 grade	85	60	55	50
beef 2 grade	-	25	30	35
Raw fat	10	10	10	10
Egg powder	5	5	5	5
Total quantity:	100,00	100,00	100,00	100,00
Spices:				
Table salt	2,5	2,5	2,5	2,5
Icing sugar	1,0	1,0	1,0	1,0
Black pepper	0,8	0,8	0,8	0,8
Sodium nitrite	0,045	0,040	0,030	0,025
Starter microorganism 'PROBIOLIS'	-	0,08	0,1	0,15

Results and discussion: According to the results of chemical analysis, it can be seen that the protein content and mineral composition of the new fermented sausage product is not significantly higher than that of the exemplary sausages (Table 2).

Table 2 - Chemical composition of fermented semi-smoked sausage product

Indicators	Half-smoked sausage			
	'Beef' (sample)	Fermented semi-smoked sausage (test sample 1)	Fermented semi-smoked sausage (test sample 2)	Fermented semi-smoked sausage (test sample 3)
Mass fraction of moisture, %	64,3±0,53	65,5±0,3	65,3±0,6	65,5±0,5
Mass fraction of protein, %	11,40±0,2	11,6±0,3	11,60±0,2	11,60±0,3
Mass fraction of fat, %	21,10±0,2	21,58±0,2	21,58±0,23	21,58±0,2
Acidity, pH	5,9±0,02	5,3±0,01	5,5±0,01	5,5±0,02
Mass fraction of residual sodium nitrite	0,06±0,0002	0,0025±0,0002	0,0033±0,0002	0,002±0,0001

It can also be seen that the mass fraction of moisture in the samples slightly increased and the acidity significantly decreased. And residual sodium nitrite in the tested sample was reduced compared to the control sample. The decrease in residual sodium nitrite in the tested samples was due to the action of starter microorganisms.

The rapid change in the transformation and colour formation of sodium nitrite can be attributed to the decrease in acidity of pH. Due to the reduction of residual sodium nitrite, a saturated colour of the finished product can be obtained.

According to the results of the amino acid composition analysis, it can be seen that the protein of the sample is significantly richer in amino acids than the amino acids contained in the semi-smoked sausage 'Beef'. An increase in tryptophan, lysine, isoleucine, valine, leucine, threonine was observed in fermented semi-smoked sausage (Table 3).

Таблица 1 - Table 3 - Amino acid composition of semi-smoked sausage

Amino acids	Quantity, 1 mg per 100g		Amino acid value, %	
	Half-smoked sausage			
	'Beef' (sample)	Fermented sausage (trial)	Fermented sausage (trial)	Fermented sausage (trial)
	836,454	907,023	170	184
Valine	572,678	701,096	124	140
Isoleucine	1036,987	1336,084	112	132
Leucine	1025,999	1383,846	155	184
Lysine	367,654	411,613	88	84
Methionine	571,033	705,714	132	150
Threonine	169,031	187,023	112	128
Tryptophan	497,987	676,682	70	78
Phenylalaline	5077,823	6309,081	120	130

The amount of substituted amino acids does not tend to fluctuate strongly. That is why the content of amino acids in the sour sausage semi-smoked is higher than in the sample. From the above results, comparing the sample with the test sample, it is clear that the amount of essential amino acids methonine in the test sample is lower by 0.4 mg/g protein, in the test sample phenylalanine increased by 0.8 mg/g protein

Biological value of the finished product is determined by the composition of mineral substances and vitamins. An important indicator of the nutritional value of fats is the acid composition of fat. It increases the flavour and energy value of the finished product. Minerals and vitamins play an important role in the human body. The human body cannot synthesise vitamins, so they enter the human body through products of plant and animal origin. Deficiency of vitamins and minerals in food leads to metabolic disorders in the human body [7].

Table 4 shows the amount of minerals and vitamins in fermented semi-smoked sausage.

Table 4 - Mineral substances and vitamins in fermented semi-smoked sausages

Indicators, 100 mg	'Beef' (sample)	Fermented semi-smoked sausage (test sample 1)	Fermented semi-smoked sausage (test sample 2)	Fermented semi-smoked sausage (test sample 3)
Minerals:				
Calcium (Ca)	34,12±3,65	34,51±5	34,50±6,9	34,50±5,5
Magnesium (Mg)	5,61±1,07	5,73±1,1	5,70±1,14	5,73±1,2
iron (Fe)	1,77±0,3	1,85±0,5	1,85±0,37	1,87±0,43
Vitamins:				
Retinol (A)	0,018	0,031	0,027	0,03
Tocopherol (E)	0,027	0,039	0,031	0,04
Ascorbic acid (C)	0,41	0,42	0,43	0,42
Niacin PP	2,63	2,71	2,65	2,71
Thiamine (B1)	0,19	0,2	0,25	0,2
Riboflavin (B2)	0,2	0,22	0,21	0,22
Energy value, kcal (kJ)	244,7 (1023,8)	250,5 (1048,09)	250,5 (1048,09)	250,5 (1048,09)

According to biological sciences, fats, minerals are not only explained by substances and vitamins, the quality of their individual components is determined, as well as the presence of individual amino acids in protein and the presence of semi-unsaturated fatty acids in fats. The rate of digestion of protein in the intestinal structure or digestion of food by proteolytic enzymes is one of the important factors determining the biological value of food.

We investigated fermented semi-smoked sausages taking into account that the biological value of protein, which depends on the quantitative units of amino acids, is degraded by enzymes. In a sample of specifically fermented semi-smoked sausage, a hydrolysis stage due to starter microorganisms was observed.

In conclusion, having analysed the research data, we are convinced that the starter has an optimal effect on the biological parameters and food quality of the finished product. By treating meat raw material with propionic acid microorganism, it is possible to increase the species, physical and chemical composition, biological value of the finished product. Thus, we can conclude that propionic acid microorganisms developing in minced meat lead to a significant increase in the content of non-exchangeable amino acids in raw meat.

In order to evaluate the effect of fermented raw materials on the properties of the finished product, a study of such indicators as moisture-holding capacity, active acidity, and yield of the finished product was carried out (Table 5).

Table 5 - Physico-chemical parameters of a sample of semi-smoked sausage

Indicators	Finished product			
	Beef sausage	Sample 1 100 kg/kg	Sample 2 100 кг/кг	Sample 3 100 кг/кг
Active acidity, pH	5,9	5,5	5,3	5,5
Moisture retention capacity, by product weight, %	60	60,9	62.3	63,8
Finished product yield, %	103,5	104,4	105,8	106

Looking at the values in Table 5, it can be seen that the active acidity of the experimental sausages is lower than the pH value of the control sample. It is very important to evaluate the pH value (active acidity) of meat raw material for sausage production.

Analysing the obtained data, it can be said that the strains of selected microorganisms grow in the minced meat sample, which can be judged by the accumulation of lactic acid and decrease in the pH of the medium.

The increase in lactic acids during meat fermentation led to a decrease in pH (active acid), thereby increasing the resistance of raw materials to putrefactive microorganisms, increasing connective tissue collagen, and changing the odour and flavour of meat.

The decomposition of carbohydrates and organic acids after treatment of raw materials with the starter microorganism showed that the accumulation of lactic acid resulted in a significantly higher rate of acidification in treated batches than in the control, which is in agreement with the report of Lin and Zhang [8].

One of the most important indicators of sausage products is moisture retention and binding integrity. The importance of water in the production of meat and meat products is very important for the production and storage of products. Moisture affects the yield of the product due to the softness of the finished product, loss of mass during heat treatment.

The starter culture has a very high moisture

content, processing of raw material leads to changes in the internal structure of meat, our goal is to control the change in moisture content of prototypes after processing of raw material with culture.

The aim of the study was to determine the quality of the finished product, pH-activity, concentration of organic acids, microbiological quality and organoleptic suitability of semi-smoked sausage with the addition of liquid propionic acid bacteria starter 'ProBioLiz' (2 strains were observed propionic acid bacteria Propionibacterium shermani and lactobacilli Lactobacillus acidophilus, L.casei, L.Plantarum).

Taking into account the biological value, organoleptic and physico-chemical indicators of the finished product, it is recommended to use fermented beef for the production of semi-smoked sausage. The optimal ratio of using liquid propionic acid bacteria 'ProBioLiz' (2 strains of propionic acid bacteria Propionibacterium shermani and lactobacillus Lactobacillus acidophilus, L.casei, L.Plantarum) is 0.1%, fermentation time - 8 hours.

Table 5 shows that the control sample of non-fermented minced meat under study has lower moisture binding capacity than the fermented minced meat samples. From the data obtained, it can be seen that there is a tendency to increase the moisture-binding capacity of the sausage sample with the addition of experimental cultures.

The ability of raw materials to retain moisture is characterised by the ability of raw materials to retain moisture during thermal processing. This indicator ensures the yield of the finished product and is the most important technological indicator. Because the yield of the finished product increases with the increase of moisture holding capacity.

Based on the studies of some authors, such as Marcel Mati, M. Magal, J. Karovicova, Ladislav Staruch, it is proved that when meat products are treated with starter microorganisms, moisture retention increases by 0.96 % [9].

After 2 hours of culturing the sample with 0.1% starter in the finished product, the lactic acid content increased by 5.5% compared to the control. After 6 hours, the difference was 15.3%, indicating a faster accumulation of lactic acid in the sample. This allows to increase moisture retention properties, due to which the content of active acid decreases.

Microbiological indicators of sausages, namely the number of mesophilic aerobic and facultative-anaerobic microorganisms, were taken into account in two stages - after sausages preparation and after the expiry of storage period. The results were processed in accordance with TR TS TS 034/2013 'On the safety of meat and meat products' [10].

Table 6 - Microbiological parameters of semi-smoked sausage after smoking

On request	CU TR 034/2013	Beef sausage	Sample 1 100 kg/kg	Sample 2 100 кг/кг	Sample 3 100 кг/кг
NoMAaAM CFU*/g, not more	Not allowed	unavailable	unavailable	unavailable	$1*10^2$, не менее
Yeast CFU*/g, not more	100	10	20	50	120

Table 7 - Microbiological indices of semi-smoked sausage after expiry date of sausage products

On request	CU TR 034/2013	Beef sausage	Sample 1 100 kg/kg	Sample 2 100 кг/кг	Sample 3 100 кг/кг
NoMAaAM CFU*/g, not more	$1*10^4$	$1*10^8$	$1*10^5$	$1*10^4$	$1*10^2$
Yeast CFU*/g, not more	100	30	45	80	150

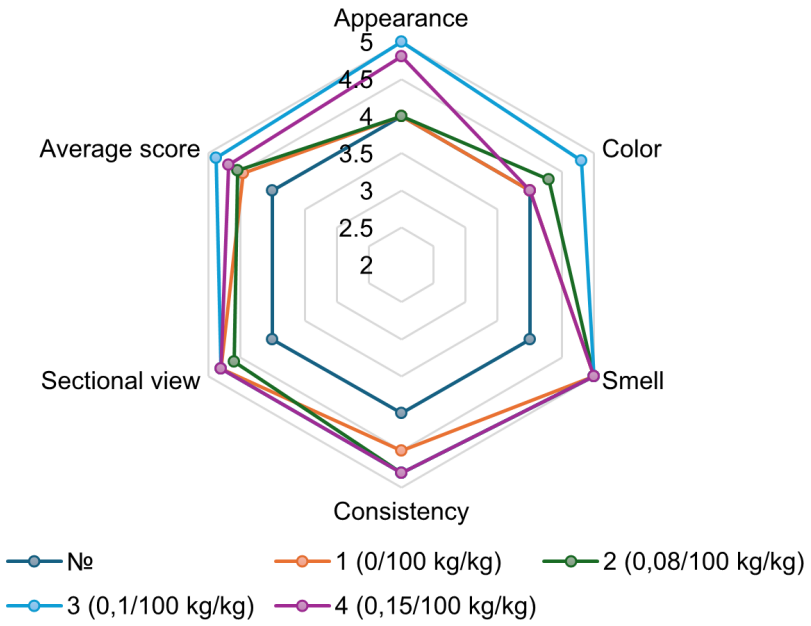
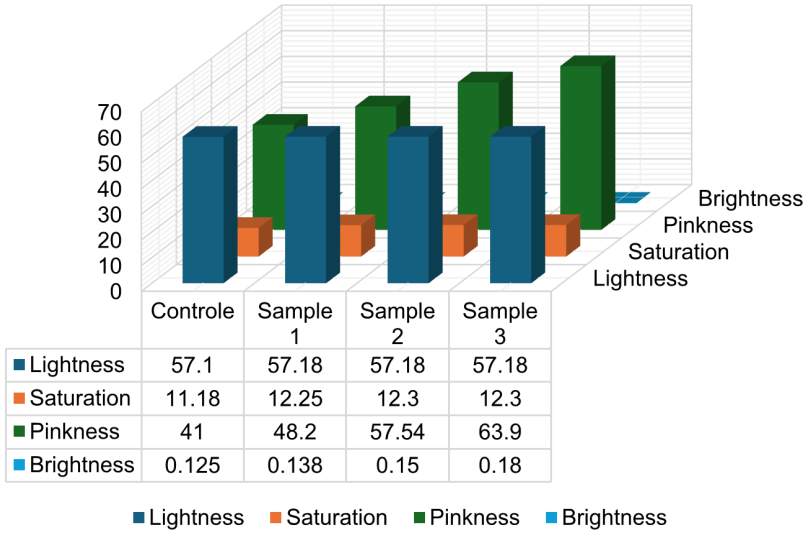


Figure 1 - Organoleptic evaluation of semi-smoked sausages using propionic acid bacteria



	Controle	Sample 1	Sample 2	Sample 3
Lightness	57.1	57.18	57.18	57.18
Saturation	11.18	12.25	12.3	12.3
Pinkness	41	48.2	57.54	63.9
Brightness	0.125	0.138	0.15	0.18

Figure 2 - The result of the color characteristics of semi-smoked sausages

According to the results of Table 6, due to the primary treatment with microorganisms at the stage of raw material preparation, we reduced the growth of the total number of bacteria in the meat raw material, and due to the introduction of activated starter microorganisms we obtained a finished product with the maximum amount of the necessary microflora.

This cannot be achieved by traditional introduction of starter microorganisms. This can be observed by comparing the indices of mesophilic aerobic and facultative-anaerobic microorganisms between the product sample and samples #1, #2, #3. It is observed that the microflora of sample No. 3 has increased and if the health improvement process is not carried out properly, the microbiological indices will increase more than normal.

This results in shortening the shelf life of the finished product. If we consider the research after the expiry date based on Table 7, it can be seen that the microbiological indices of the product sample are very high. It can be seen that the finished product

has not reached its shelf life. According to the results, it can be concluded that model #2 is the most optimal.

The consistency of the finished samples was characterized by a more dense, monolithic structure compared to the control, which is consistent with the results of microstructural studies (Fig. 1). The color of sample No. 3 was homogeneous, red. In the control, a looser consistency was noted, the color was less intense. Thus, in sample No. 4, sour taste, pungent odor, dark red color was detected. This may be due to the presence of lactic acid microorganisms in the experimental sample, which play an important role in the formation of the flavor of semi-smoked sausages. The flavor in sample No. 3, made with propionic acid bacteria, differs from the rest by a pronounced taste and aroma.

The data of organoleptic evaluation agree with the data of the results of color characteristics of sausages and structural and mechanical indicators (Fig. 2).

The performed differential analysis showed that treatment of raw materials with propionic acid bacteria makes it possible to reduce the lightness index and increase the rose part of the spectrum, which forms higher levels of saturation and brightness of color of finished products.

Conclusion. The quality of finished product, pH-activity, concentration of organic acids, microbiological quality and organoleptic properties of semi-smoked sausage treated with liquid propionic acid bacteria 'ProBioLiz' (2 strains of propionic acid bacteria Propionibacterium shermani and lactobacillus Lactobacillus acidophilus, L.casei, L.Plantarum) were studied.

The optimal ratio of using liquid propionic acid bacteria 'ProBioLiz' (2 strains of propionic acid bacteria Propionibacterium shermani and lactobacillus Lactobacillus acidophilus, L.casei, L.Plantarum) is 0.1%, fermentation time - 8 hours.

Considering the works of the above-mentioned researchers and the results obtained in the course of research, the technology of semi-smoked sausage with the use of starter cultures was developed.

On the basis of the obtained data it is established that the use of propionic acid microorganisms in the processing of meat products leads to the disintegration of fibrous weights.

Technological scheme of production of traditional semi-smoked sausages does not require special technical changes, so the new type of product can be produced at any meat processing enterprise.

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