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STUDY OF THE EFFECT OF PROBIOTICS ON THE SHELF LIFE OF CHILLED BROILER MEAT

^{1,2}N.ZH. BEGDILDAYEVA* ^{1,2,3}SH.N. AKHMETSADYKOVA , ²A.S. NURGAZINA , ⁴A.K. KUDAIBERGENOVA , ²N.N. AKHMETSADYKOV

(¹Almaty Technological University, Republic of Kazakhstan, 050012, Almaty, 100 Tole bi Str.

²LLP "Research and Production Enterprise "Antigen", Republic of
Kazakhstan, 040905, Abay, 4 Azerbayev Str.

³LLP "Kazakh Research Institute for Livestock and Fodder Production", Republic of Kazakhstan,
055552/A10M6C4, Almaty, 51 Zhandosov Str.

⁴Al-Farabi Kazakh National University, 050040, Republic of Kazakhstan, Almaty, 71 al-Farabi ave.)

Corresponding author e-mail: nbegdildayeva@gmail.com*

Poultry meat can be contaminated with a wide range of microorganisms, including those that can spoil the product during chill storage and certain foodborne pathogens. This study aims to investigate the effect of probiotics on the shelf life of shilled broiler meat. The object of the study was fresh broiler meat obtained from broiler chickens fed three different amounts of probiotics, which were isolated from shubat and koumiss. For the control group, birds were fed a basal diet (BD) and drinking water (DW). Chicks in the experimental groups were fed by BD, DW, and probiotics 0.25 mL, 0.5 mL, and 1.0 mL per bird (groups Pro1, Pro2, and Pro3, respectively), for 42 days. Following slaughter, the filets were packed aerobically and stored in temperature-controlled conditions at 4±2°C. Microbial examinations of meat quality included QMAFAnM, BGKP, pathogenic (Salmonella, Listeria monocytogenes), yeast, and mold. The sensory parameters were used to determine the meat samples' shelf life. Broiler filets had a sensory shelf life of 7 to 9 days. The novelty of this study lies in its focus on the use of probiotics in broiler meat, particularly in the context of Kazakhstan. While probiotics have been studied in a variety of food products, their effect on broiler meat in Kazakhstan has yet to be investigated. As per our results, we can recommend the application of the Pro2 level for the best shelf life, microbial quality, and sensory properties.

Keywords: meat quality, shelf life, poultry, probiotics, sensory index.

ИЗУЧЕНИЕ ВЛИЯНИЯ ПРОБИОТИКОВ НА СРОК ХРАНЕНИЯ ОХЛАЖДЕННОГО МЯСА БРОЙЛЕРОВ

 1,2 Н.Ж. БЕГДИЛДАЕВА*, 1,2,3 Ш.Н. АХМЕТСАДЫКОВА, 2 А.С. НУРГАЗИНА, 4 А.К. КУДАЙБЕРГЕНОВА, 2 Н.Н. АХМЕТСАДЫКОВ

(¹Алматинский Технологический Университет, Республика Казахстан, 050012, Алматы,ул. Толе би, 100 ²ТОО "Научно-производственное предприятие "Антиген",

Республика Казахстан 040905, п. Абай, ул. Азербаева, 4

³ТОО "Казахский научно-исследовательский институт животноводства и кормопроизводства ", Республика Казахстан, 055552/A10M6C4, Алматы, ул. Жандосова, 51 ⁴Казахский Национальный Университет им. Аль-Фараби, Республика Казахстан, 050040,

Алматы, пр. Аль-Фараби, 71) Электронная почта автора корреспондента: nbegdildayeva@gmail.com*

Мясо птицы содержит разнообразные микроорганизмы, среди которых могут быть как микробы, способные вызвать порчу продукта при хранении в холодильнике, так и конкретные патогены, вызывающие заболевания, передающиеся через пищу. Цель данного исследования - изучить влияние пробиотиков на

срок хранения свежего мяса бройлеров. Объектом исследования было свежее мясо бройлеров, полученное от цыплят-бройлеров, которых кормили тремя различными дозировками пробиотика, выделенного из шубата и кумыса. В контрольной группе птиц кормили базовым рационом (БР) и питьевой водой (ПВ). Цыплят в экспериментальных группах кормили БР, ПВ и пробиотиками по 0,25 мл, 0,5 мл и 1,0 мл на птицу (группы Pro1, Pro2 и Pro3, соответственно) в течение 42 дней. После убоя филе упаковывали аэробно и хранили при температуре 4±2°С. Микробные исследования качества мяса включали КМАФАнМ, БГКП, патогенные (Salmonella, Listeria monocytogenes), дрожжи и плесени. Сенсорные параметры исполь-

зовались для определения срока годности образцов мяса. Срок годности филе бройлера по сенсорным показателям составил от 7 до 9 дней. Новизна данного исследования заключается в том, что оно посвящено использованию пробиотиков в мясе бройлеров, однако пробиотики были изучены в различных продуктах питания, несмотря на то, их влияние на мясо бройлеров в Казахстане еще не исследовалось. Согласно нашим результатам, мы можем рекомендовать применение уровня Pro2 для достижения наилучшего срока хранения, микробного качества и сенсорных свойств.

Ключевые слова: качество мяса, срок хранения, бройлер, пробиотики, сенсорный индекс.

ПРОБИОТИКТЕРДІҢ САЛҚЫНДАТЫЛҒАН БРОЙЛЕР ЕТІНІҢ САҚТАУ МЕРЗІМІНЕ ӘСЕРІН ЗЕРТТЕУ

 1,2 Н.Ж. БЕГДІЛДАЕВА*, 1,2,3 Ш.Н. АХМЕТСАДЫКОВА, 2 А.С. НУРГАЗИНА, 4 А.К. КУДАЙБЕРГЕНОВА, 2 Н.Н. АХМЕТСАДЫКОВ

(¹Алматы Технологиялық Университеті, Қазақстан Республикасы, 050012, Алматы, Төле би к-сі, 100 ²"Антиген" Ғылыми-өндірістік кәсіпорны" ЖШС, Қазақстан Республикасы, 040905, Абай ауылы, Әзірбаев к-сі, 4 ³"Қазақ мал шаруашылығы және жемшөп өндірісі ғылыми-зерттеу институты" ЖШС 055552 /

А10М6С4, Қазақстан Республикасы, Алматы, Жандосов көшесі, 51,

⁴Қазақ Ұлттық Университеті, әл-Фараби, Қазақстан Республикасы, 050040,
Алматы, әл-Фараби даңғылы, 71)

Автор-корреспонденттің электрондық поштасы: nbegdildayeva@gmail.com*

Кұс етінде әртүрлі микроорганизмдер болады, олардың арасында тоңазытқышта сақтаған кезде өнімнің бұзылуына әкелетін микробтар да, тамақ арқылы берілетін әртүрлі ауруларды тұдыратын арнайы қоздырғыштар да кездеседі. Бұл мақаланың мақсаты пробиотиктердің салқындатылған бройлер етінің сақтау мерзіміне әсерін зерттеу. Зерттеу нысаны ретінде шұбат пен қымыздан бөлініп алынған пробиотиктердің үш түрлі мөлшерімен қоректенген бройлер тауықтарынан алынған балғын бройлер еті алынды. Бақылау тобында құстарға негізгі диета (НД) және ауыз су (АС) берілді. Тәжірибе топтарындағы тауықтар 42 күн ішінде НД, АС және бір құсқа пробиотиктердің 0,25 мл, 0,5 мл және 1,0 мл (сәйкесінше Pro1, Pro2 және Pro3) мөлшерімен азықтандырылды. Союдан кейін филе аэробты түрде оралып, $4\pm2^{\circ}C$ температурада сақталды. Ет сапасын микробтық зерттеуге $MA\Phi A$ нM саны, колиморфты және колиформды бактериялар, патогендік (Salmonella, Listeria monocytogenes), ашытқы және зен санырауқұлақтары кірді. Ет үлгілерінің жарамдылық мерзімін анықтау үшін сенсорлық параметрлер қолданылды. Бройлер филесінің жарамдылық мерзімі сенсорлық көрсеткіштер бойынша 7-ден 9 күн аралығында болды. Бұл зерттеудің жаңалығы Қазақстанда бройлер етінде пробиотиктерді қолдануға арналған. Пробиотиктер әртүрлі тағамдарда зерттелгенімен, олардың Қазақстандағы бройлер етіне әсері әлі зерттелмеген. Зерттеу нәтижелеріне сәйкес, сақтау мерзіміне, жоғары микробтық сапаға және сенсорлық қасиеттерге қол жеткізу үшін Pro2 деңгейін қолдану ұсынылады.

Негізгі сөздер: ет сапасы, сақтау мерзімі, құс еті, пробиотиктер, сенсорлық индекс.

Introduction

The food industry is always looking for new ways to extend the shelf life of fresh meat products while maintaining their microbial and sensory quality. Microbiological safety and poultry meat quality are equally important to producers, retailers, and consumers, and both involve microbial contaminants on the processed product. To some extent analogous to traditional poultry production, various factors can affect alternative production methods, and the addition of a suitable probiotic may enhance efficacy, decrease morbidity and mortality, diminish environmental contamination, and augment food safety. Probiotics, defined as live microorganisms that can provide a health benefit to the host when consumed in suffi-

cient amounts, have been shown in various studies to improve animal health and performance [1].

This research aims to look into the effect of probiotics on the shelf life of chilled broiler meat. In this study, the probiotics under investigation were sourced from koumiss and shubat, which are fermented drinks derived from mare's and camel's milk, respectively.

Shori (2017) [2] suggests that raw camel milk and its fermented products are potential sources of beneficial probiotic strains. Furthermore, camel milk and the microorganisms present in shubat are essential contributors to the sensory attributes, such as flavor, texture, and acidity, of these beverages. They also have therapeutic effects on digestion and antimicrobial properties [3].

Koumiss, a fermented drink derived from mare's milk, has been recognized as a potential source of probiotics. During the fermentation of koumiss, several microorganisms such as lactic acid bacteria, yeast, and acetic acid bacteria actively participate, thereby enhancing the microbial diversity and conferring health-promoting effects to the beverage. The presence of probiotic microorganisms, including *Lactobacillus* and *Bifidobacterium* species, has been identified in koumiss through scientific investigations, and these microorganisms have been shown to confer beneficial effects on the host's health [4].

The specific goals of this study are to determine the microbial quality of the meat samples, including QMAFAnM, BGKP, pathogenic (*Salmonella, Listeria monocytogenes*), yeast, and mold, as well as to evaluate the sensory parameters to determine the shelf life of the meat samples.

The study's hypothesis is that using probiotics in broiler feed will increase the shelf life and microbial quality of fresh broiler meat. The study will take a quantitative approach, with experimental design and statistical data analysis.

This study is significant because it adds to our understanding of the potential benefits of probiotics in fresh broiler meat in Kazakhstan. The findings of this study can help the food industry improve the shelf life and microbial quality of fresh meat products, as well as increase the nutritional value of animal feed.

Materials and Research Methods

Materials. Lacticaseibacillus paracasei B 5.2, Lacticaseibacillus paracasei subsp. paracasei SH1 (GenBank Accession No. OQ411023), Lactiplantibacillus plantarum K2 and Kazachstania unispora Y 2.2 (Genbank Accession No. OP984721) were used in this study as the potential probiotics microorganisms. The strains were obtained from the culture bank of LLP Research and Production Enterprise "Antigen" (Almaty, Kazakhstan) which was stored at -80°C.

A total of 240 one-day-old Ross 308 chicks were randomly allotted to four treatment groups of 3 replicates (20 birds per replicate). For the control group, birds were fed by basal diet and drinking water (DW). Chicks in the experimental groups were fed by BD, DW, and probiotics 0.25 mL, 0.5 mL, and 1.0 mL per bird (groups Pro1, Pro2, and Pro3, respectively), for 42 days.

For sensory and microbiological evaluations after slaughter, 40 samples were randomly

obtained and analyzed. These samples consisted of 10 filets from each group: 1 - control group, 2 - Pro1 experimental group, 3 - Pro2 experimental group, and 4 - Pro3 experimental group.

Methods. Microbiological analysis. According to the Health Regulations «Sanitary and epidemiological requirements for food production facilities» (dated April 28, 2021, No. KR DSM-36), broiler chicken carcasses must undergo at least three sanitary and microbiological control points during both control and experimental batches. The first control point immediately after production and serves as a background check. The second and third control points occur on the second and third days. Subsequent control points are conducted every two days until the microbial indicators meet the requirements of the sanitary rules.

Of the standard microbiological methods available for analyzing food products, the following will be applicable:

-State Standard 10444.15-94. Food products. Methods for determination of the quantity of mesophilic aerobes and facultative anaerobes;

-State Standard 52816 -2007. Food products. Methods for detection and quantity determination of coliforms:

-State Standard 30726-2001. Food-stuffs. Methods for detection and determination of Escherichia coli;

-State Standard 52814-2007 (ISO 6579:2002). Food products. Method for the detection of Salmonella;

-State Standard 29185 -91. Food products. Methods for detection and quantity determination of sulfite-reducing clostridium;

-State Standard 51921 – 2002. Food products. Methods for detection and determination of Listeria monocytogenes bacteria;

-State Standard 10444.12-88. Food products. Method for determination of yeast and mold.

Sensory Investigation. A trained sensory group evaluated the sensory investigations 24 hours and 192 hours after slaughter using a three-point rating system, with 3 indicating freshness and high quality and 1 indicating unacceptability. Color, odor, and texture were among the characteristics assessed for each sample. The sensory index (SI) was computed as a weighted average value using the following equation (1):

$$SI = \frac{2 \times O + 2 \times C + T}{5} \tag{1}$$

where SI is Sensory Index, O is odor, C is color and T is texture.

According to the scheme, the product degrades when SI reaches 1.8. SI was built as a time

series and fitted to a linear model. As a result, the shelf life of each sample was calculated as follows using Equation 2:

$$SL = \frac{1.8 - a}{b} \tag{2}$$

where SL is shelf life, a is the intercept of the linear model, and b is the slope of the linear model.

Samples exhibiting an atypical spoilage process (no degradation in color or texture) were not included in the statistical analyses of sensory characteristics. Outliers were defined as samples with a shelf life of fewer than 100 hours and greater than 300 hours, and they were removed from the data set [5].

Literature review

Kazakhstan aims to quadruple domestic poultry meat production to 740,000 tonnes per year by 2027, this includes 100.5 tonnes of fresh chicken meat production [6]. Consequently, there are more demands for quality, which is the most important thing in the modern poultry processing industry.

The indiscriminate use of antibiotics in poultry farms has heightened public concern about the residual contamination of poultry products. The ban on the sale of antibiotic-based growth promoters on markets in the European Union and many other countries has pushed poultry producers to find suitable alternatives [7].

Probiotics are considered environmentally friendly feed additives and promising non-traditional alternatives to chemotherapy in poultry [8]. Furthermore, any animal must maintain a certain amount of beneficial microbiota in the gastro-intestinal tract at all times in order to maintain proper microbial balance [9].

Poultry meat is more perishable than meat from other animals, such as beef or pork. Freshness measurement is thus critical in poultry processing plants to ensure meat quality [10]. The storage and processing of poultry meat directly impact its functional and palatability properties. Meat quality control is required to improve the sensory characteristics and functional properties of meat samples, reduce economic losses, and increase poultry industry efficiency [11]. Physical and chemical methods have traditionally been used to assess meat quality. The current trend in meat quality monitoring is to move quality measurements from the laboratory to the processing line for quick and early detection of qualityrelated parameters. Because of its physicochemical properties, raw poultry meat is especially vulnerable to microbiological spoilage [12]. Thus, poultry meat can respond significantly to physicochemical changes caused by dietary changes. There has been little research into the connection between feed composition and meat spoilage.

There have also been few studies on the effect of feed composition on quality loss and shelf life. Most studies focus solely on typical quality parameters following the slaughtering process, with no consideration given to the effect on freshness parameters during storage [13,14]. To our knowledge, there have been limited investigations on how probiotic food additives affect the usual sensory and microbiological parameters during storage. The purpose of this research paper is to investigate the effect of probiotics on the shelf life of broiler meat.

Results and their discussion

The effects of probiotics on microbiological parameters are shown in Table 1. The study findings indicated that the QMAFAnM index of the experimental group on the second, third, fifth, and seventh days met the standards specified in the Health Regulations, with the index value being $1,27\pm0,06\times10^2,\ 2,97\pm0,04\ \times10^2,\ 5,21\pm0,16\ \times10^2,$ and $9,86\pm0,11\ \times10^2\ CFU/g$, respectively. No other microbial contaminants were detected during the investigation. However, on the eighth day, the QMAFAnM index of the bird carcass flushes increased to a level that did not comply with the Health Regulations, with a value of $2,18\pm0,06\ \times10^3\ CFU/g$, and the presence of yeast and mold fungi was also noted.

Upon analyzing the flushes of broiler chicken carcasses from Pro1 and Pro3 groups, the QMAFAnM values were found to be 2,86±0,13 x10², 2,94±0,04 x10² on the third day, 4,95±0,07 x10², 5,16±0,01 x10²CFU/g on the fifth day, and 9,62±0,08 x10², 9,71±0,07 x10²CFU/g on the seventh day, respectively, which were within the acceptable range. No other sanitary and microbiological contamination was observed. However, on the ninth day, laboratory investigations demonstrated an elevation in QMAFAnM to 2,23±0,04 x10³, 1,94±0,05 x10³ CFU/g in the flushes of broiler chicken carcasses, along with the identification of BGKP, *L.monocytogenes*, yeast, and mold fungi.

Table 1. Microbial parameters of poultry meat stored at a temperature of 4 ± 2 °C, n = 40

	QMAFAnM, CFU/g, no more	The weight of the product (g)			Yeast,	Mold,	Sulfite -	
Para- meters		is prohibited						
		BG	Pathogenic	L.monoc	CFU/g, no	CFU/g, no	reducing	E.coli
meters	er erg, no more	KP	, including	ytogenes	more	more	clostridia	
			salmonella	yrogenes				_
Admis-	4.03	0.4	2.5	2.5	In-	In-	In-	In-
sible	$1*10^3$	0,1	25	25	admissible	admissible	admissible	admissi
norm	Miana	1.: -1			h1 1 /	- Ct 1 1-t	-)	ble
Microbial contamination during storage, background (after slaughter) Control 1,27±0,06x10 ²								
Pro1	$1,34\pm0,11 \times 10^2$	-	-	-	-	-	-	-
Pro1	1,34±0,11 X10	-	-	-	-	-	-	-
Pro2	$1,24\pm0,09 \text{ x} 10^2$	-	-	-	-	-	-	-
Pro 3	$1,31\pm0,02 \text{ x} 10^2$	-	-	ı	-	-	-	-
Microbial contamination during storage, 3rd day								
Control	$2,97\pm0,04 \text{ x} 10^2$	-	=	-	-	-	-	-
Pro1	$2,86\pm0,13 \text{ x}10^2$	-	=	-	-	=	-	-
Pro2	$2,62\pm0,08 \text{ x} 10^2$	-	=	-	-	-	-	-
Pro 3	$2,94\pm0,04 \text{ x} 10^2$	ı	-	ı	=	=	-	-
Microbial contamination during storage, 5th day								
Control	$5,21\pm0,16 \times 10^2$	-	-	ı	-	-	-	-
Pro1	$4,95\pm0,07 \text{ x} 10^2$	-	-	-	-	-	-	-
Pro2	$4,67\pm0,09 \text{ x}10^2$	-	-	-	-	-	-	-
Pro 3	$5,16\pm0,01 \text{ x} 10^2$	-	-	-	-	-	-	-
Microbial contamination during storage, 7th day								
Control	$9,86\pm0,11 \text{ x}10^2$	-	-	-	-	-	-	-
Pro1	$8,19\pm0,05 \text{ x}10^2$	-	-	-	-	-	-	-
Pro2	$7,49\pm0,08 \text{ x} 10^2$	-	-	-	-	-	-	-
Pro 3	$8,97\pm0,14 \text{ x}10^2$	-	-	-	-	-	-	-
Microbial contamination during storage, 8th day								
Control	$2,18\pm0,06 \text{ x}10^3$	-	-	-	+	+	-	-
Pro1	$9,62\pm0,08 \text{ x} 10^2$	-	-	-	-	-	-	-
Pro2	$8,47\pm0,15 \text{ x}10^2$	-	=	-	-	=	-	-
Pro 3	$9,71\pm0,07 \text{ x}10^2$	-	=	-	-	-	-	-
Microbial contamination during storage, 9th day								
Control	$3,07\pm0,09 \text{ x}10^3$	-	-	-	+	+	-	-
Pro1	$2,23\pm0,04 \times 10^3$	-	-	-	-	-	-	-
Pro2	$9,61\pm0,06 \times 10^{2}$	-	-	-	-	-	-	-
Pro 3	$1,94\pm0,05 \text{ x} 10^3$	-	-		-	-	-	-
Microbial contamination during storage, 10th day								
Control	$4,12\pm0,07 \times 10^3$	-	-	+	+	+	-	-
Pro1	$3,85\pm0,04 \times 10^3$	-	-	+	+	+	-	-
Pro2	$1,56\pm0,06 \times 10^3$	-	-	-	+	+	-	-
Pro 3 2,82±0,11 x10 ³ - - + + - -								
«-» - absence of microbial growth; «+»-presence of microbial growth								

When evaluating the sanitary and microbiological parameters of Group Pro2, the QMAFAnM index increased from 1,24±0,09 x10² to 9,61±0,06 x10²CFU/g on the third and ninth days, respectively. However, during laboratory assessments on the tenth day, the QMAFAnM index was found to exceed the limits prescribed by the Health Regulations, amounting to 1,56±0,06 x10³ CFU/g, in addition, bacteria from

the *Escherichia coli* group were isolated on the tenth day. Therefore, the results of the investigation suggest that Pro3 is more practical for future research aimed at extending the shelf life of chilled poultry filets, as preliminary data suggest that it can maintain surface microbial contamination of broiler chicken filets within the range required by the Health Regulations. Research indicates that probiotic bacteria have the ability to

generate various antimicrobial substances, which possess antibacterial and antifungal propertiesThe sensory evaluations conducted at the onset of the storage period indicated that the supplemented groups exhibited a superior sensory quality of meat samples with regards to odor, color, and texture, as reflected by mean values above 2.7 on the

assessment scale where 3 represents the highest quality. No significant differences were observed between the treatment groups after a storage period of 24 hours (Figure 1). However, the control group showed lower scores for odor and color parameters compared to the supplemented groups, leading to a lower sensory index.

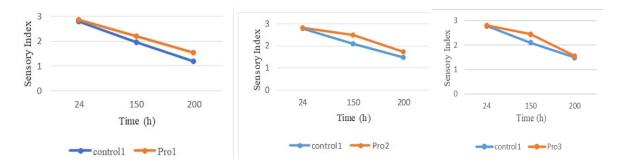


Figure 1. Determination of the sensory index and shelf life for each treatment group (n-40)

According to Bruckner et al. (2012) [15], when stored at 4°C, commercially produced fresh poultry has a sensory shelf life of 6 to 8 days. However, the shelf life was slightly extended in the current study, most likely due to the use of noncommercial slaughtering methods. When compared to broiler meat production in high-throughput commercial facilities, the microbial load, air composition, and processing equipment used in the current study resulted in comparable low initial microbial counts on the meat [16], resulting in lower initial microbial counts on the meat.

Conclusions

The present study investigated the impact of probiotics on the sensory attributes and shelf life of fresh broiler meat. No significant differences between the Pro1 and Pro3 treatment groups were observed in meat quality characteristics. However, the addition of Pro2 led to a more compact fillet and an extended shelf life compared to the control group. Despite all treatment groups exhibiting acceptable microbial levels, the control group had a higher microbial load at the end of the storage period, indicating less microbiological degradation in the probiotic-supplemented groups. Further research is necessary to establish a precise correlation between microbial growth and shelf life.

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