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The effect of probiotic culture on quality characteristics of sucuk

Probiyotik kültürün sucuğun kalite karakteristikleri üzerine etkisi

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Abstract

Objective: This study investigated the feasibility of incorporating the probiotic lactic acid bacteria strain *Lacticaseibacillus casei* 431 in sucuk production.

Material and methods: For this purpose, three groups of sucuk were prepared: Group 1 with a starter culture (*Latilactobacillus sakei* S15 + *Staphylococcus xylosus* GM92), Group 2 with the probiotic culture (*L. casei* 431), and Group 3 combining both starter and probiotic cultures. Then, the sucuk samples were subjected to physical, chemical, microbiological, and sensory analyses on the 0th, 30th, and 60th days of storage.

Results and conclusion: The results demonstrated that the microbial culture factor significantly influenced the sucuk samples' pH, L^* value, a^* value, thiobarbituric acid reactive substances (TBARS), odor, taste, and overall acceptability scores, as well as the counts of lactic acid bacteria, Micrococcus/Staphylococcus, yeastsmolds, and L. casei (P < 0.01). However, it had no significant impact (P > 0.05) on water activity, b^* value, color and texture parameters. The storage period notably affected the TBARS, L^* value, lactic acid bacteria (P < 0.05) and L. casei counts (P < 0.01), whereas it did not significantly alter other examined parameters.

Keywords: Lacticaseibacillus casei; probiotic sucuk; fermented sausage; starter culture

Öz

Amaç: Bu çalışmada, probiyotik laktik asit bakteri suşu *Lacticaseibacillus casei* 431'in sucuk üretiminde kullanılabilirliği arastırılmıstır.

Materyal ve yöntem: Bu maksatla çalışma kapsamında üç grup sucuk hazırlanmıştır: Grup 1: starter kültür (*Latilactobacillus sakei* S15 + *Staphylococcus xylosus* GM92); grup 2: probiyotik kültür (*L. casei* 431); grup 3: starter kültür + probiyotik kültür. Sucuk örneklerine üretimi müteakip depolamanın 0., 30. ve 60. günlerinde fiziksel, kimyasal, mikrobiyolojik ve duyusal analizler uygulanmıştır.

Tartışma ve sonuç: Araştırma sonuçlarına göre mikrobiyal kültür faktörü sucuk örneklerinin pH, L^* değeri, a^* değeri, tiyobarbitürik asit reaktif maddeler (TBARS), koku, tat ve genel kabul edilebilirlik değerleri ile laktik asit bakteri, Micrococcus/Staphylococcus, maya-küf ve L. casei sayıları üzerinde önemli düzeyde etkili olmuştur (P < 0.01). Buna karşın mikrobiyal kültür faktörü sucuk örneklerinin su aktivitesi, b^* değeri, renk ve tekstür parametreleri üzerinde önemli (P > 0.05) etkiye sahip değildir. Depolama periyodu ise örneklerin TBARS, L^* değeri, laktik asit bakteri (P < 0.05) ve L. casei sayıları (P < 0.01) üzerinde önemli etki sergilemiştir. Ancak depolama periyodunun incelenen diğer parametreler üzerinde önemli etkisi olmamıştır.

Anahtar kelimeler: Lacticaseibacillus casei; probiyotik sucuk; fermente sosis; starter kültür

1. Introduction

Probiotics are live microorganisms that have beneficial effects on host health when consumed in amounts Agricultural adequate (Food and Organization (FAO), 2006). These microorganisms can be directly added to food or offered as dietary supplements in the form of powder, capsules, or tablets (Cocconcelli and Fontana, 2008). The rapid increase in the trend towards health-focused and healthy living foods, which started with probiotic dairy products in the 1990s, has also increased the interest in developing probiotic meat products. Probiotics require the intake of adequate amounts of live microorganisms to confer their benefits, and fermented sausages are considered an optimal delivery matrix for such probiotics. (Bağdatlı and Kundakçı, 2013; Bis-Souza et al., 2019a; Khan et al., 2011; Kröckel, 2013; Pavli et al., 2020). The most commonly used probiotic microorganism strains belong to the genera Lactobacillus and Bifidobacterium. Additionally, microorganisms from the genera Enterococcus and Pediococcus are also used as probiotic cultures, albeit less frequently (Cavalheiro et al., 2015).

Fermented sausages are generally classified into two categories as dry and semi-dry fermented sausages. Dry fermented sausages do not undergo any heat treatment, whereas the production process for semi-dry fermented sausages may include a heat treatment step (Caplice and Fitzgerald, 1999; Kumar et al., 2017). In these products, lactic acid bacteria and Gram-positive, catalase-positive cocci are technologically significant microorganisms constituting two important components of the product microbiota. Lactic acid bacteria contribute to product safety and the development of sensory attributes such as texture and color through acid production. Gram-positive, catalase-positive cocci influence color formation and stability through their nitrate reductase activities, delay oxidation with their catalase activities, and contribute to flavor formation through their proteolytic and lipolytic properties. Specific strains selected from both groups of microorganisms are used either individually or in combination as starter cultures in industrial production (Yalınkılıç, 2009; Karslıoğlu et al., 2014; Kaya and Kaban, 2019; Alvarez et al., 2023; Yılmaz Topcam et al., 2024).

Sucuk is a type of dry fermented sausage commonly produced in Türkiye, in which beef and/or sheep/buffalo meat, along with sheep tail fat and/or beef fat are used as raw materials. In this product, bovine small intestine or collagen casings are used as casing. Fermentation and drying are two critical stages in the production of sucuk (Kaban and Kaya, 2009; Yalınkılıç et al., 2012). The initial fermentation temperature and ripening period vary between 12 to 26 °C and 6 to 20 days, respectively (Kaban and Kaya, 2008, 2009; Sover et al., 2005; Yalınkılıç et al., 2012). Unlike semidry fermented sausages, which undergo heat treatment, the production process for dry fermented sausages does not involve any heat treatment. Therefore, sucuk provides a favorable environment for probiotic microorganisms.

In the production of probiotic meat products, strains exhibiting probiotic properties, isolated or from intestinal microbiota and/or identified fermented meat products, can be utilized. However, it is necessary to determine the resilience of these strains to processing conditions and storage environments (Kołozyn-Krajewska and Dolatowski, 2012; Neffe-Skocińska et al., 2016). To this end, studies have been conducted on the potential applications of probiotic microorganisms in both sucuk (Bağdatlı and Kundakçı, 2016; Ergönül and Kundakçı, 2011; Kaya and Aksu, 2005; Kozan and Sarıçoban, 2023; Öztürk Er, 2002; Tükel and Şengün, 2024; Ünal Turhan et al., 2019) and other non-heat-treated fermented sausages (Agüero et al., 2020; Arief et al., 2014; Bis-Souza et al., 2019b, Bis-Souza, 2020; Erkkilä et al., 2001; Holko et al., 2013; Klingberg and Budde, 2006; Klingberg et al., 2005). Moreover, each new probiotic strain, considered to have technological significance, may have varied effects on product quality characteristics, necessitating their experimental use in the product environment. As mentioned, the use of various probiotic strains in the production of sucuk has been examined. However, research is still needed to develop new strains with high potential for adaptation to the sucuk environment and to determine their effects on product quality characteristics. Thus, the aim of this study was to investigate the effects of simultaneous use of both starter culture (L. sakei S15 and S. xylosus GM92) and probiotic culture (L. casei 431) on the quality characteristics of sucuk.

2. Materials and methods

2.1. Materials

Beef meat and beef fat from three different beef carcasses, obtained from a local butcher, were used as raw materials. Spices used in the formulation were purchased from a local market. Sodium nitrite and all other chemicals used in the study were obtained from a medical supply company. The study was conducted with three replicates, using one carcass for each replicate. The strains Latilactobacillus sakei S15 (Kaya et al., 2015) and Staphylococcus xylosus GM92 (Kaban and Kaya, 2008) were used as starter cultures, while Lacticaseibacillus casei 431 (Christian Hansen, Denmark), a commercial probiotic culture strain, was used as the probiotic culture.

The chemicals and culture media used in this study are as follows: Sodium chloride (NaCl, Merck), Trichloroacetic Acid (TCA, 1.00807, Merck), Ethylenediaminetetraacetic Acid (EDTA, 1.08418, Merck), Propyl Gallate (P3130, Sigma-Aldrich), 2-Thiobarbituric Acid (TBA, T5500, Sigma-Aldrich), and Ethanol (1.00983, Merck). The culture media include De Man Rogosa Sharpe Agar (MRS, 1.10660, Merck), Mannitol Salt Phenol-Red Agar (MSA, 1.05404, Merck), Violet Red Bile Dextrose Agar (VRBD-Agar, 1.10275, Merck), Rose Bengal Chloramphenicol Agar (RBC-Agar, 1.00467, Merck), and MRS-IM Agar.

2.2. Sucuk production

In the production of sucuk, beef meat and fat were used in an 80:20 ratio as raw materials, and spice varieties were included in the formulation as specified by Yalınkılıç et al. (2012). Sucrose (4 g/kg of sucuk dough) was added as a source of sugar and 150 mg/kg of sodium nitrite as a curing agent. In the study, three groups of sucuk were prepared: Group 1 with a starter culture (Latilactobacillus sakei S15 + Staphylococcus xylosus GM92), Group 2 with the probiotic culture (Lacticaseibacillus casei 431), and Group 3 combining both starter and probiotic cultures (L. sakei S15 + S. xylosus GM92 + L. casei 431). A total of 9 sucuk doughs were prepared by making 3 production runs (replicate) for each experimental group. The sucuk doughs were prepared in a smallscale cutter (MADO, Germany) and stuffed into collagen casings (Naturin Darm, 38 mm) using a small-scale stuffer (MADO, Germany). After stuffing, the sucuk samples from each group were

allowed to rest for 4 hours in the production area before being transferred to the climate chamber (Reich, Germany) for the ripening stage, where air speed, relative humidity, and temperature could be automatically adjusted. The initial fermentation temperature was set at 24 ± 1 °C and was gradually reduced from the second day onward. The relative humidity was set at 92% and was gradually decreased over the following days. Air velocity was set at 0.5 m/s for the first three days and then reduced to 0.1 m/s in subsequent days. Ripening was terminated when the water activity (a_w) of the sucuk samples dropped below 0.90. The ripened sucuk samples were vacuum-packaged using a laboratory-type packaging machine (Multivac, Germany) in a packaging material made of Polyamide/Polyethylene with low oxygen, gas, and water vapor permeability. After packaging, the samples were stored at a cold temperature (4 \pm 1 °C) for 60 days, and samples from each group were subjected to microbiological, sensory, physicochemical analyses on days 0, 30, and 60.

2.3. Analyses

2.3.1. Microbiological analyses

For microbiological analyses, 25 g of samples were taken and homogenized in a sterile stomacher bag with 225 mL of sterile physiological saline for 2 minutes (Lab Stomacher Blander 400-BA 7021, Seward, England). The homogenized samples were then analyzed for lactic acid bacteria counts using de Man, Rogosa, and Sharpe Agar (MRS) (30°C 48 under anaerobic conditions), Micrococcus/Staphylococcus counts using Mannitol Salt Agar (MSA) (30°C for 48 h under aerobic conditions), Enterobacteriaceae counts using Violet Red Bile Dextrose Agar (VRBD) (30°C for 48 h under anaerobic conditions) (Baumgart et al., 1993), and yeast-mold counts using Rose Bengal Chloramphenicol Agar (RBC) (25°C for 5 days under aerobic conditions) (Gökalp et al., 2010). For determining the count of probiotic bacteria, MRS-IM Agar specified by manufacturer of the probiotic culture was used (Chr Hansen, Denmark, 2005). After inoculation, the plates were incubated at 20 °C for 6 days, followed by enumeration (Chr Hansen, Denmark, 2005). Additionally, verification tests were conducted on typical colonies (Baumgart et al., 1993).

2.3.2. pH and water activity (a_w)

Ten grams of samples were weighed and homogenized with 100 mL of distilled water using a homogenizer. The pH value of the homogenized samples was measured using a previously calibrated pH meter (Gökalp et al., 2010). The water activity (a_w) of the samples was measured using a water activity device (Novasina, Pfäffikon, Switzerland).

2.3.3. Color analysis

The color measurements of the sucuk samples were determined using a colorimeter (Chroma Meter CR-400, Japan). The color values (L^* , a^* , and b^*) of the samples were measured on the cutting surfaces of the samples.

2.3.4. TBARS analysis

The analysis of thiobarbituric acid reactive substances (TBARS) was performed using the method described by Lemon (1975), and the results are presented as mg MDA/kg.

2.3.5. Sensory analysis

Sucuk samples were evaluated by 25 trained panelists in each panel. Prior to the sensory analysis, participants were informed about the products and given preliminary instructions on how to taste the sucuk. The analysis was carried out by presenting the samples coded with 3-digit numbers to the panelists along with the sensory panel form. The number of participants in each replication was 25, with a total of 75 participants. Sensory evaluation parameters including color, texture, odor, taste, and overall acceptability were scored on a 1-9 point scale.

2.3.6. Statistical analyses

The study was carried out according to a randomized complete block experimental plan with 3 replications, based on the factors of treatment (starter culture, probiotic culture, starter culture + probiotic culture) and storage period (0, 30, and 60 days). The raw data were examined for normality of distribution and homogeneity of variances between groups, and then subjected to analysis of variance (ANOVA) using SPSS version 24 (SPSS Inc., Chicago, IL, USA). For the mean values that showed statistically significant differences (P < 0.05) according to the ANOVA, Duncan's multiple range test was also applied.

3. Results and discussion

3.1. pH

The effects of the microbial culture and storage period on the pH value of the sucuk are presented in Table 1. The pH value measured in all groups was below 5.0. This was probably due to the organic acids released during the metabolic activities of lactic acid bacteria used in sucuk production. Similar results have been obtained in studies utilizing starter cultures (Yalınkılıç et al., 2012) or probiotic cultures (Bağdatlı and Kundakcı, 2016). The average pH values in sucuk samples using either starter or probiotic cultures alone are higher compared to those where both were used together (P < 0.05). The combined use of starter and probiotic cultures has resulted in a greater reduction in product pH. The interaction of microbial culture and storage period did not significantly affect the average pH values of the sucuk samples over a 60-day storage period (P > 0.05). Contrary to our findings, a study using L. casei CRL-431 and Lactobacillus acidophilus as probiotic cultures in turkey meat-based probiotic sucuk found that the pH value decreased during the first two months of storage, with no change in the subsequent months (Ergönül, 2009). Another study found no variation in the pH of probiotic sucuk depending on the culture type, although a partial decrease was observed during storage (Ünal Turhan et al., 2017). However, in research using strains Lactobacillus acidophilus DSM 20079, Lacticaseibacillus casei 431, and L. acidophilus NCFM in probiotic sucuk, an increase in product pH values was observed in the later stages of storage. The same study also noted differences in product pH depending on the strain used (Kozan and Sarıçoban, 2023).

3.2. Water activity

The effects of the microbial culture and storage period on the water activity value of the sucuk are presented in Table 1. Water activity values in all samples were at least 0.90, indicating that the desired level of drying in the product was achieved. According to the results, neither microbial culture nor the storage process had a statistically significant effect on the water activity values of probiotic sucuk samples (P > 0.05). Similarly, a study on probiotic sucuk found that the microbial culture used did not significantly affect the product's water activity, and no significant changes were observed in the water activity values up to the 60th day of storage (Ünal Turhan et al., 2017). In

contrast, research conducted by Kozan and Sarıçoban (2023) determined that lactic acid bacteria strains used in the production of probiotic sucuk influenced the product's water activity, and the storage duration also negatively affected the water activity values. On the other hand, in the present study, the interaction of microbial culture with the storage period did not have a statistically significant impact on the average water activity values of the sucuk samples (P > 0.05).

3.3. Color

The effects of the microbial culture and storage period on the L^* , a^* , and b^* values of the sucuk are presented in Table 1. The microbial culture variable significantly influenced the L^* and a^* values of sucuk samples (P < 0.01) while it had no significant effect on the b^* value (P > 0.05). The storage period only significantly affected the L^* parameter of sucuk samples (P < 0.01), with no significant impact on other parameters (P > 0.05).

Similarly, a research showed that the L^* values of turkey sucuk produced with different probiotic cultures varied depending on the type of culture used, and that an 8-month storage process had a significant effect on the product's L^* values. However, in contrast to our findings, the same study reported that the culture used did not significantly influence the a^* value. The researcher also observed that both the culture used and the period of storage had a significant effect on the b^* value (Ergönül, 2009). In sucuk samples produced using L. acidophilus DSM 20079, L. casei 431, and L. acidophilus NCFM strains, a regular decrease in L^* , a^* , and b^* values was observed throughout the storage period, and the cultures used did not have a significant impact on the product's L^* , a^* , and b^* values (Kozan and Sarıçoban, 2023). Additionally, in the present study, the interaction of microbial culture with the storage period had no statistically significant effect on the instrumental color parameters of the samples (P > 0.05) (Table 1).

Table 1. Overall effect of microbial culture and storage period on the pH, a_w, instrumental color and TBARS parameters of dryfermented probiotic sucuk.

	pН	$a_{ m w}$	L^*	a*	<i>b</i> *	TBARS (mg MDA/kg)	
Microbial Culture (MC)							
SC	$4{,}94{\pm}0{,}14^a$	$0,900\pm0,002^a$	$41,76\pm1,47^a$	$14,20\pm0,77^{b}$	$12,61\pm1,23^{a}$	$0,78\pm0,30^{a}$	
PC	$4,88\pm0,12^a$	$0,902\pm0,004^a$	39,71±0,80 ^b	16,29±0,93ª	$12,41\pm1,47^{a}$	$0,47\pm0,17^{c}$	
SC+PC	$4,78\pm0,07^{b}$	$0,899\pm0,005^a$	40,94±1,51 ^a	15,90±0,90°a	$13,05\pm0,84^{a}$	$0,63\pm0,28^{b}$	
Significance	**	NS	**	**	NS	**	
Storage Period (SP)							
Day 0	$4,83\pm0,12^a$	$0,902\pm0,005^a$	$41,72\pm1,48^a$	15,23±1,53 ^a	11,93±1,11 ^a	$0,36\pm0,08^{c}$	
Day 30	$4,86\pm0,13^a$	$0,901\pm0,003^a$	$40,\!82{\pm}1,\!63^{ab}$	15,42±1,36 ^a	13,35±0,75 ^a	$0,62\pm0,18^{b}$	
Day 60	$4,90\pm0,14^{a}$	$0,899\pm0,004^a$	$39,87\pm0,85b^a$	$15,74\pm0,84^a$	$12,79\pm1,30^{a}$	$0,91\pm0,21^{a}$	
Significance	NS	NS	**	NS	NS	**	
Interaction							
MCxSP	NS	NS	NS	NS	NS	*	

a-c: any two means in the same column having the same letters in the same section are not significantly different. *p < 0.05, **p < 0.01, NS: not significant. SC: starter culture, PC: probiotic culture, SC+PC: starter culture + probiotic culture.

3.4. TBARS

The effects of the microbial culture and storage period on the TBARS values of the sucuk samples are presented in Table 1. The microbial culture had a significant effect on the TBARS value of sucuk (P<0.01). The use of probiotic cultures resulted in lower TBARS values in the product compared to other groups (P < 0.05) (Table 1). Ergönül (2009)

found that turkey sucuk produced with *L. casei* CRL-431 had lower TBA values compared to those produced with *L. acidophilus* strain, with the highest average TBARS values observed in the group using starter cultures. Upon examining the storage period, it was determined that oxidation increased over time (P < 0.01), with the lowest TBARS values recorded on day 0 and the highest on day 60 of storage (P<0.05). Additionally, the

interaction of microbial culture with the storage period had a statistically significant effect on the TBARS values of the sausage samples (P < 0.05). The observed increase in TBARS values during storage is likely due to the accumulation of compounds released by oxidative reactions (Feiner, 2006). Parallel to our findings, a study on probiotic turkey sucuk noted an increase in TBA values over an eight-month storage period (Ergönül, 2009). However, in a study where three different probiotic lactic acid bacteria strains were used separately, Kozan and Sarıçoban (2023) observed a decrease in TBARS values during storage, and also noted variability in TBARS values depending on the bacterial strain used.

3.5. Microbiological properties

The effects of microbial culture and storage period on microbiologically significant groups in sucuk samples are presented in Table 2. The lactic acid bacteria count was affected by microbial culture (P<0.01). The highest average number of lactic acid bacteria was found in the group using starter culture, with significant differences between this group and all others (P < 0.05). The lowest average number of lactic acid bacteria was observed in the group using only the probiotic culture (P<0.05). Similar results were observed for the counts of *Micrococcus/Staphylococcus* (P < 0.01) (Table 2). However, the interaction of microbial culture and storage period had a statistically significant effect on the Micrococcus/Staphylococcus values of the sucuk samples (P < 0.01). A difference at the P < 0.05 level was found between the group using only the probiotic culture and the group using both starter and probiotic cultures in terms of L. casei counts. Similar to our findings, Bağdatlı and Kundakcı (2016) found lactic acid bacteria and L. casei CRL-431 counts to be above 8 log and 6 log, respectively, in their study on probiotic sucuk. This study shows that the count of L. casei 431 in probiotic sucuk samples exceeded the 6 log cfu/g level as indicated by Kolozyn-Krajewska and Dolatowski (2009), demonstrating that the L. casei 431 strain has sufficient adaptability for probiotic sucuk production (Bağdatlı and Kundakcı, 2016). The count of yeasts/molds in all groups was below the 3 log level, with the lowest average value found in the group produced with the starter culture (P < 0.05). In contrast to our findings, studies on probiotic sucuk produced from beef (Bağdatlı and Kundakcı, 2016) and turkey meat (Ergönül, 2009) using the *L. casei* CRL-431 strain reported yeast/mold counts below detectable limits. In the present study, Enterobacteriaceae counts remained below the detectable limit (<2 log) in all groups, likely due to the reduction in water activity below 0.90 and the observed decrease in pH.

The storage period had a significant (P < 0.05)effect on the count of lactic acid bacteria and a very significant (P < 0.01) effect on the count of L. casei, with both microbial parameters experiencing significant declines during storage. Neither Micrococcus/Staphylococcus nor veast/mold counts showed significant changes during the storage period (P > 0.05) (Table 2). Data from the study indicate that the count of L. casei remained above 7 log cfu/g during storage, demonstrating the high survivability of this strain throughout the storage period. This indicates that the L. casei 431 strain used in the study is suitable for probiotic sucuk production. Similarly, Ünal Turhan et al. (2017) observed a decline in both lactic acid bacteria and Lactobacillus rhamnosus counts during the storage period in a study that used microencapsulated probiotic L. rhamnosus. Another study reported an increase in lactic acid bacteria during storage in probiotic sucuk and also noted that yeast/mold counts reached 4 log in the samples (Kozan and Sarıçoban, 2023).

3.6. Sensory parameters

The effects of the microbial culture and storage period on the sensory parameters of the sucuk samples are presented in Table 2. The storage period did not have a statistically significant effect on the sensory characteristics of the product (P > 0.05). However, the microbial culture factor had a highly significant impact (P < 0.01) on odor, taste, and overall acceptability parameters, but was not significant for color and texture parameters (P > 0.05). Average values for color and texture across all groups were above 7 points. However, the lowest average values in groups where the microbial culture factor was significant were found in samples using only the probiotic culture. The use of probiotic culture had a negative impact on product sensory parameters such as odor, taste, and overall acceptability. In contrast, a study using L. casei CRL-431 in sucuk production reported that this strain improved the sensory characteristics of the product (Bağdatlı and Kundakcı, 2016). The highest average values for odor and overall acceptability were found in the group using both starter and probiotic cultures. Additionally, the interaction of microbial culture and storage period had statistically significant effect on texture (P<0.01) and taste (P<0.05) parameters of the samples. Ergönül (2009) found that the microbial culture used in the production of probiotic turkey sucuk significantly affected the product's odor value and observed a decline in odor value over the storage period, contrary to our findings. The researcher also stated, contrary to our results, that color and texture parameters were significantly affected by the microbial culture used and the duration of storage. In a study on sucuk production using L plantarum and microencapsulated

probiotic *L. rhamnosus*, it was determined that the microbial culture had no significant impact on the product's slice color, texture, pleasant odor, and overall acceptability scores (Ünal et al., 2017). Another study on sucuk production using microencapsulated probiotic *L. rhamnosus* also found no significant differences in slice color, texture, pleasant odor, and overall acceptability scores among the groups during the first 60 days of storage, but a significant decline in sensory scores was observed by the sixth month of storage. In the same study, it was determined that the varieties of cultures used did not have a significant impact on the sensory characteristics of the product (Ünal Turhan et al., 2017).

Table 2. Overall effect of microbial culture and storage period on the sensory and microbiological parameters of dry-fermented probiotic sucuk.

	Color	Texture	Odor	Taste	Overall acceptability	Lactic acid bacteria (log CFU/g)	Micrococcus / Staphylococcus (log CFU/g)	Mold- Yeast (log CFU/g)	Lacticaseibacillus casei (log CFU/g)
Microbial Culture (MC)									
SC	$7,10\pm0,36^{a}$	7,20±0,32°7	7,22±0,39 ^t	⁵ 7,17±0,64 ^a	$7,04\pm0,34^{b}$	8,70±0,13 ^a	$6,28\pm0,23^{a}$	$2,21\pm0,21^{b}$	ND
PC	7,20±0,31ª	7,10±0,21ª6	6,30±0,27°	6,11±0,29b	6,11±0,21°	7,56±0,17°	5,26±0,65 ^b	2,77±0,45a	$7,60\pm0,16^{a}$
SC+PC	7,34±0,22ª	7,24±0,19 ^a 7	7,56±0,41°	a7,31±0,22a	7,38±0,20 ^a	8,55±0,20 ^b	5,52±0,25 ^b	2,87±0,43ª	7,46±0,11 ^b
Significance	NS	NS	**	**	**	**	**	**	**
Storage Period (SP)									
Day 0	$7,14\pm0,32^{a}$	7,16±0,18 ^a 7	7,10±0,61°	a6,77±0,56a	$6,79\pm0,59^{a}$	8,37±0,55a	$5,87\pm0,47^{a}$	$2,82\pm0,64^{a}$	$7,63\pm0,13^{a}$
Day 30	$7,20\pm0,36^{a}$	7,11±0,28a6	5,84±0,55	a6,74±0,67a	$6,89\pm0,62^{a}$	8,27±0,54ab	5,63±0,54 ^a	2,63±0,32a	$7,51\pm0,16^{b}$
Day 60	$7,30\pm0,25^{a}$	7,28±0,26°7	7,13±0,79°	a7,08±0,81a	$6,86\pm0,65^{a}$	8,17±0,57 ^b	$5,57\pm0,77^{a}$	2,40±0,32a	$7,45\pm0,14^{b}$
Significance	NS	NS	NS	NS	NS	*	NS	NS	**
Interactions									
MCxSP	NS	**	NS	*	NS	NS	**	NS	NS

a-c: any two means in the same column having the same letters in the same section are not significantly different. *p < 0.05, **p < 0.01, NS: not significant. SC: starter culture, PC: probiotic culture, SC+PC: starter culture + probiotic culture.

4. Conclusion

Considering the results obtained in this study, sucuk, a traditional dry fermented sausage, has proven to be a suitable matrix for the probiotic *L. casei* 431 during both production and storage. The findings demonstrated that *L. casei* 431 can successfully survive and maintain its viability within the sucuk matrix throughout fermentation and storage, reinforcing its potential as a probiotic carrier food. However, it was observed that *L. casei* 431 negatively affected certain sensory properties of sucuk, particularly when used alone. Therefore,

it is recommended to combine this strain with starter cultures to optimize both sensory quality and product acceptance. Future research could explore additional probiotic strains to further enhance the sensory and functional characteristics of sucuk, paving the way for innovative fermented meat products with added health benefits.

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Conflict of interest

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