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Research Article

Mechanical and Microbiological Properties of Natural Casings Using in Meat Products

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Abstract

The aim of this study is to investigate microbiological and mechanical specifications of the 21 natural casings were provided from various retail markets, slaughter houses and meat processing plants in Turkey that is ready to process. The ash content of the casings were varied between 0.61-3.07% ($P<0.05$). Water vapor permeability (WVP) of the casings was determined between 1.02-4.37 mg/cm^{2}h ($P<0.05$). Elongation and tensile strength of the casings were determined between 2.5-18.21% and 5.88-44.08 N/mm^2 respectively ($P<0.05$). The most stable sample expressed as breaking force was reported as Casing7 with 39N while the weakest samples were defined as Casing11 and Casing19 ($P<0.05$). Contamination with *Escherichia coli* was determined in nine samples. Sixteen samples were contaminated with coagulase positive *Staphylococci* where Casing1 and Casing14 had the highest contamination values as 5.77 log cfu/g and 5.81 log cfu/g respectively ($P<0.05$). Casing14 had the highest total aerobic mesophilic bacteria recorded as 6.49 log cfu/g while Casing6 and Casing1 were recorded as 6.32 log cfu/g and 6.34 log cfu/g respectively ($P<0.05$). *Salmonella* spp. was detected on Casing13 and Casing19.

Keywords: *Natural casing, Water vapor permeability, Mechanical properties, Tensile strength, Elongation break, Microbiological safety*

Et Ürünlerinde Kullanılan Doğal Kılıfların Mekanik ve Mikrobiyolojik Özellikleri

Öz

Bu çalışmanın amacı Türkiye'nin çeşitli bölgelerindeki perakende satış mağazalarından, kesimhanelerden ve et işleme tesislerinden tedarik edilen kullanıma hazır 21 doğal kılıfın mikrobiyolojik ve mekanik özelliklerinin araştırılmasıdır. Kılıfların kül içeriği %0.61-3.07 arasında tespit edilmiştir ($P<0.05$). Örneklerin su buhar geçirgenliği (WVP) ise 1.02-4.37 mg/cm^{2}h ($P<0.05$) olarak kaydedilmiştir. Uzama ve kopma dayanımları sırasıyla %2.5-18.21 ve 5.88-44.08 N/mm^2 ($P<0.05$) olarak ölçülmüştür. Kopma dayanımı testine en dayanıklı örnek 39N ile Casing7 olarak tespit edilirken, en dayanıksız örneğin Casing11 ve Casing19 ($P<0.05$) olduğu gözlemlenmiştir. Dokuz örneğin *Escherichia coli* ile kontamine olduğu belirlenmiştir. Onaltı kılıfın koagulaz pozitif *Staphylococci* ile kontaminasyonu tanımlanırken Casing1 ve Casing14 5.77 log kob/g ve 5.81 log kob/g ile en yüksek değerlere sahip örnekler olduğu belirlenmiştir ($P<0.05$). Casing14'de 6.49 log kob/g toplam aerobik mezofilik bakteri tespit edilmişken Casing6'da ve Casing1'de sırasıyla 6.32 log kob/g ve 6.34 log kob/g değerlerinde kontaminasyon belirlenmiştir ($P<0.05$). *Salmonella* spp. Casing13 ve Casing19'da tespit edilmiştir.

Anahtar sözcükler: *Doğal kılıf, Su buharı geçirgenliği, Mekanik özellikler, Kopma dayanımı, Uzama yüzdesi, Mikrobiyal güvenlik*

INTRODUCTION

Meat and meat products is one of the cornerstones of the healthy diet with the rich content of high biological value proteins and micro essential nutrients [1-3]. Although source and type of the meat products may be varied depending on the culture, customer preferences, dietary habits and life style, Food and Agriculture Organization

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(FAO) estimates 37 kg meat consumption in 2030 in developing countries [4].

The first record on traditional meat product filled to a special casing that is called as sausage in general term was stated in Babylon by Sumerians around 4000 BC [5,6]. Definition of Làchàng, traditional Chinese sausage made by goat and lamb meat, was first indicated in 589 BC. Blood sausage in Greek history was mentioned in Odyssey Book in 550 BC [6]. Moreover, sucuk is the traditional fermented meat product filled into the casings in Turkish culture. Sucuk can be produced from beef and water buffalo meat and it may contain beef fat, sheep tail fat, salt, sugar, nitrite, nitrate and/or nitrite/nitrate and various spices [7,8] and this mixture is stuffed into a casing where fermentation is carried out until a semi or dry product is obtained [9]. Processing steps and conditions and specifications of the sausages may be differing culture to culture by the time and various sausage type meat products are available in the markets nowadays.

Casing is one of the critical parts of the traditional and common sausages by shaping, supporting the size and stability, maturating characteristic taste and odor, promoting chemical, microbiological and structural properties during the production, fermentation and storage steps [10-15]. Casings can be mainly categorized by to two groups as natural and artificial ones [15-17]. Natural casings used for sausage industry are derived from intestines of animals such as cattle, sheep, goats, pigs and very rarely deer and horse and process is followed by cleaning, washing, scraping, treating with solutions such as additives and preservatives and drying [6,12-14,18,19]. Water vapor and gas permeability, promotion on microflora of fermentation process and supporting traditional taste are some of the characteristics of natural casings to make a demand [14]. However, they have some disadvantages such as microbial risk, cost on production, unstandardized structure and insufficient mechanical strength during process [5,10,17]. Although artificial casings do not hold any of these disadvantages of natural casings, low permeability for both gas and water vapor does not meet the requirements of traditional fermented sausage type products. Moreover, low costs, stability on shelf life and uniform physical properties increased the interest of artificial casings last decades [5,12,17,18].

The aim of this study is to investigate microbiological and mechanical specifications of the natural cattle casings achieved from the market that is ready to process.

MATERIAL and METHODS

Materials

Casings: Twenty one casings were provided from various retail markets, slaughter houses and meat processing plants randomly from Afyonkarahisar, Antalya, Gaziantep, Konya and Mersin provinces in Turkey. Intestines were achieved by cattle, cleaned and dried properly and became ready to fill sucuk, traditional Turkish meat product. Dried casings were supplied and stored in clean plastic bags at 4°C for the analysis.

Methods

Analyses were carried at Kazlıçeşme R&D Center and Accredited Test Laboratories, (Istanbul, Turkey) based on the international standards.

Ash Content (%): Three-five grams casing was weighted into porcelain crucible. Two mL paraffin (liquid petrolatum) was added into the crucible to prevent overflow. Crucibles were placed into the stove (Nabertherm, Germany) which was heated at 550°C for 3 h. Samples were burned at 550°C for 16 h and, then, moved desiccator immediately and weighted after 3 h. Percentage of ash content (W_{TA}) is calculated as the formulation indicated below [20]:

$$W_{TA} = (m_2 - m_1) * \frac{100}{m_0}$$

Where, m_o is the sample weight (g), m_1 is the crucible weight (g), m_2 is the crucible and ash weight after burning (g).

Water Vapor Permeability: Water vapor permeability of the casings was determined based on TS EN ISO 14268 [21] standards. Initially, silica gel was added to the jar until half of it was almost full. Samples were cut into the pieces where the diameter is 4 cm and placed into the neck of the jar and sealed with the cover tightly where the 3 cm of the sample was in contact with the air between the jar and the environment. Jar with the sample and silica gel was weighted and placed into machine (Hilab, Portugal). After running the fan for 8-16 h at 23°C and 51% moisture environment, jar was weighted again. Water vapor permeability in mg/(cm^2.h) (W_{VP}) is calculated as the formulation indicated below:

$$W_{VP} = \frac{m}{At} = \frac{m}{\pi r^2 t}$$

Where, m is the weight difference (First weight - Second weight) (mg), r is the diameter of the sample in contact with air (cm), t is the analyze period (h).

Permanent Elongation (E_s): TS EN ISO 17236 [22] is the standard used for determination of permanent elongation. Sample was cut into the strips and analyzed equipment (Shimadzu, Japan) set based on the sample dimensions. Pieces placed in the jaw of the analyzer in a vertical position. Analyzer run until 20.0N±0.5N, hold 10±1 sec and moved quickly to the initial position. Permanent elongation (E_s) as percentage is calculated as below:

$$E_s = \frac{(L_1 - L_{0)}}{L_0} x100$$

Where, L_0 is the length before the test (mm), L_1 is the length after the test (mm).

Elongation at Break (E_1): Percentage elongation at break was determined by the standard of TS EN ISO 3376 [23]. Sample was cut into the strips and analyzed equipment (Shimadzu, Japan) set based on the sample dimensions. Pieces placed in the jaw of the analyzer in a vertical position. Analyzer run and percentage elongation caused by a specific load (E_1) as percentage is calculated as below:

$$E_1 = \frac{(L_1 - L_{0)}}{L_0} x100$$

Where, L_0 is the separation of the jaws at the specific load, L_1 is the initial separation of the jaws.

Breaking Force: Analyses of breaking force were carried by TS EN ISO 3376 [23]. Sample was cut into the strips and analyzes equipment (Shimadzu, Japan) set based on the sample dimensions. Pieces placed in the jaw of the analyzer in a vertical position. Analyzer run until pieces were broken (F).

Tensile Strength (T_n): TS EN ISO 3376 [23] was the standard used to define tensile strength. Sample was cut into the strips and analyses equipment (Shimadzu, Japan) set based on the sample dimensions. Pieces placed in the jaw of the analyzer in a vertical position. Analyzer run until pieces were broken and tensile strength in Newtons per square millimeter (T_n) is calculated as below:

$$T_n = \frac{F}{\overline{w} - \overline{t}}$$

Where, F is the highest force applied to the sample (N), is the mean width of the sample (mm), is the mean thickness of the sample (mm).

Escherichia coli: The analyses were carried by the rules of β-glucoronidase positive *E. coli* count based on ISO 16649: 2012 [24] and ISO 7218: 2007 [25] standards. Twenty five gram sample was homogenized in 1 g/L NaCl and 8 g/L peptone water using a stomacher. One mL 10^{-2} dilution was added to a petri plate following 15 mL TBX agar that was prepared and stored at 45°C. Petri plates moved inversed after stirring carefully and incubated at 44°C for 18-24 h. At the end of the incubation time, typical blue colonies were counted on TBX agar.

Sulfide-reducing Anaerobe Bacteria: Based on the ISO 15213: 2003 [26] and ISO 7218: 2007 [25] standards, 1 mL 10^{-2} dilution was added to a petri plate following 15 mL Iron Sulphite agar that was prepared and stored at 45°C. Ten mL agar was added when the inoculation became solid form. Petri plates incubated at 37°C for 24-48 h in anaerobic chamber (Bactron 300, Shellab, Sheldon Manufacturing, USA Incorporation using a mixed gas of 5% hydrogen, 5% carbon dioxide, and 90% nitrogen) and black colonies were counted.

Coagulase Positive Staphylococci: Based on the ISO 6888-1: 1999 standards [27], 1 mL 10^{-2} dilution were added to two petri plates. Inoculums were applied carefully and stored at room temperature for absorption for 15 min. Petri plates turned and stored at 37°C for 48 h. Typical colonies that seem black or grey, shiny, 1.0-1.5 mm diameter and covered by an opaque zone were counted. For correction, selected five colonies were moved to brain heart infusion broth and incubated at 37°C for 24 h. Colonies were carried to tubes containing 0.3 mL rabbit plasma surrounded 0.1 mL culture and incubated at 37°C for 4-6 h. Incubation was carried to 24 h if coagulation was not occurred. If the coagulant was more than the half of the tube, coagulase test was positive.

Coliform Group Bacteria: One mL 10^{-2} dilutions were added to two petri plates based on the TS ISO 4832: 2010 [28] and ISO 7218: 2007 [25] standards and 15 mL Violet Red Bile Lactose agar that was stored at 44-47°C bath was added and moved around. Four mL agar was added when the agar became solid form. Petri plates turned and stored at 37°C for 24 h. Colonies which were 0.5 mm diameter, purple-red color and surrounded by precipitation zone were counted. For the correction, five colonies moved tubes containing Brillant Green Lactose Bile 2% Broth and incubated at 37°C for 24 h. Coliforms formed gases.

Total Aerobic Mesophilic Bacteria: The analysis was carried out ISO 4833-1: 2013 [29]. One mL 10^{-2} dilution were added to a petri plate following 15 mL PCA agar addition that was prepared and stored at 45°C. Petri plates moved to mix dilution and agar homogenously. Petri plates turned when the agar became solid form and incubated at 37°C for 48 h.

Yeasts and Molds: Yeasts and molds were determined based on ISO 21527-2: 2012 [30] and TS ISO 7218: 2007 standards [31]. One mL 10^{-2} dilution were added to a petri plate following 15 mL dichloran 18% glycerol agar (DG18) addition that was prepared and stored at 45°C. Petri plates moved to mix dilution and agar homogenously. Petri plates turned when the agar became solid form and incubated at 25°C for 5 days.

Listeria monocytogenes: *Listeria monocytogenes* was determined based on TS EN ISO 11290-1: 1997 [32] and ISO 7218: 2007 [25]. Casing and half fraser broth at 1:10 ratio were weighted and incubated at 30°C for 24 h after homogenization on the first enrichment. 0.1 mL sample was added 10 mL Fraser broth and incubated at 36°C for 48 h as the pre-enrichment process. At the end of the incubation, sample was moved to petri plates containing Agar Ottaviani and Agosti agar (ALOA) and Oxford agar and incubated at 37°C for 24 h. Greenish blue color colonies surrounded by opaque zones on ALOA agar and 1.5-2.0 mm diameter greenish gray colonies on Oxford agar were detected.

***Salmonella spp.*:** TS EN ISO 6579: 2005 [33] and ISO 7218: 2007 [25] were the standards to determine *Salmonella* spp. Peptone water with sample was incubated at 37°C for 18 h. 0.1 mL culture was added to two different tubes containing 10 mL Rappaport-Vassiliadis Soya (RVS) agar and Muller-Kauffmann Tetratiyonat Novabiyosin (MKTTN) agar and incubated at 41.5°C and at 37°C for 24 h, respectively. Culture taken from RVS and MKTTN agar was planted to Xylose-Lysine-Desoxycholate agar (XLD) agar as the first selective agar and, then, planted Brillant Green agar (BGA) agar as the second selective agar. After incubation, colonies were tested.

Statistical Analysis: One way analyses of variance were applied to data procedure of SPSS22 (SPSS INC). Least significant mean values were generated and corresponding least significant difference (LSD) values were calculated at a value of 0.05. The number of replication was two for all analyses.

RESULTS

Twenty one casings were analyzed to determine some chemical and mechanical properties. Mineral component of the casings that is defined as ash content is between 0.61-3.07% (P<0.05) shown in the *Fig. 1*. The highest ash content was determined on Casing20 while Casing2 had the lowest ash value (P<0.05). As shown in the *Fig. 2*, the highest water vapor permeability was reported on Casing12 with 4.37 mg cm^2 h (P<0.05). Casing17 and Casing20 has similar water vapor permeability values that were determined as 1.02 mg cm^{2}h and 1.044 mg/cm^{2}h respectively (P<0.05). Casings could not be produced as an industrial product that may cause unstable properties; even if, one casing might exhibit different results. Thickness of Casing7 and Casing13 were measured as 0.16 mm while Casing12 and Casing14 were determined as 0.15 mm. 0.2 mm was the thickness of Casing20 (P<0.05) as presented in *Fig. 3*. One of the mechanical properties of the casings is defined as elongation at break. As in the *Fig. 4*, elongation at break was determined as 18.21% and 2.50% on Casing14 with the highest value and Casing16 and Casing19 with the lowest values respectively (P<0.05). Tensile strength which is expressed as the stability of filling during production was also determined. The strongest sample was determined as Casing20 with 44 N mm^2 while weak samples were reported as Casing12 and Casing13 with 6 N/mm^2 tensile strength (P<0.05) as shown in *Fig. 5*. Another parameter to characterize the strength was breaking force and results are presented in *Fig. 6*. The most stable sample was reported as Casing7 with 39N while the weakest samples were defined as Casing11 and Casing19 (P<0.05). Samples could not resist the load up to 20N that caused difficulties to define permanent elongation.

Table 1 presents microbial specifications of the casings. Contaminations with *E. coli* were determined in nine samples. Casing5 and Casing6 were contaminated with *E. coli* as 4.85 log cfu/g and 4.60 log cfu/g respectively (P<0.05). Sulfide reducing anaerobe bacteria were determined in Casing12 and Casing15. Sixteen samples were contaminated with coagulase positive *Staphylococci* such as *Staphylococcus aureus* where Casing1 and Casing14 had the highest contamination values as 5.77 log cfu/g and 5.81 log cfu/g respectively (P<0.05).

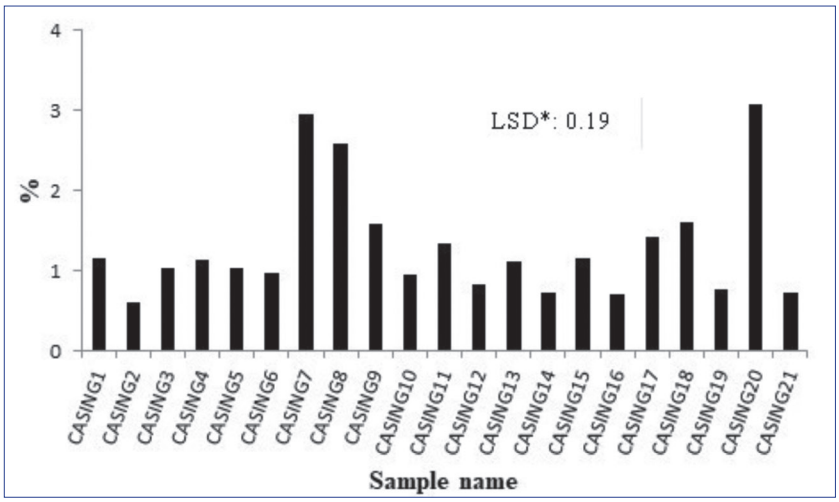

Fig 1. Ash content of natural casings (* LSD: least significant difference (calculated at α = 0.05)

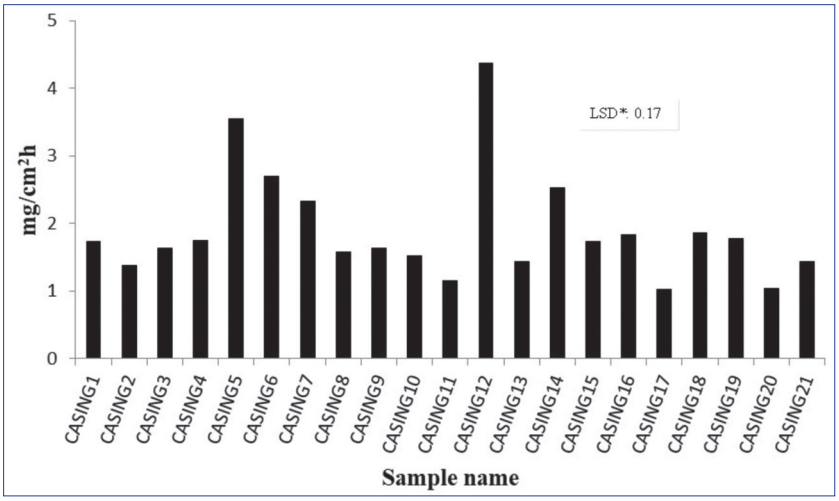

Fig 2. Water vapor permeability (* LSD: least significant difference (calculated at α = 0.05)

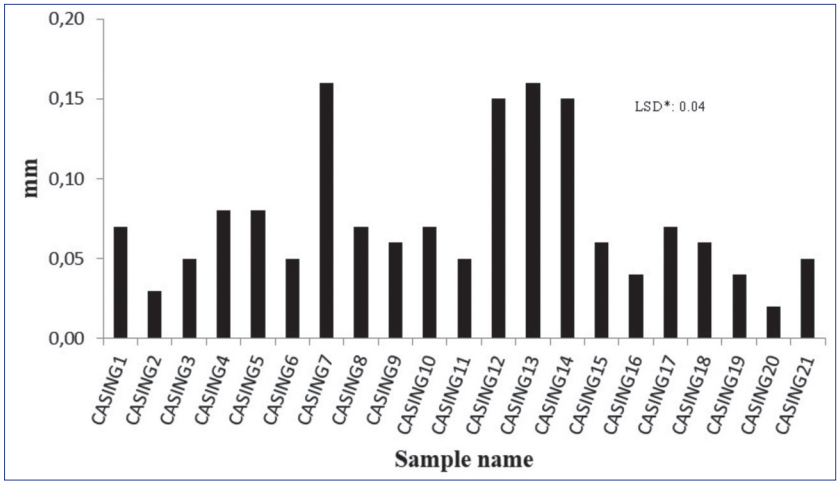

Fig 3. Thickness of natural casings (* LSD: least significant difference (calculated at α = 0.05)

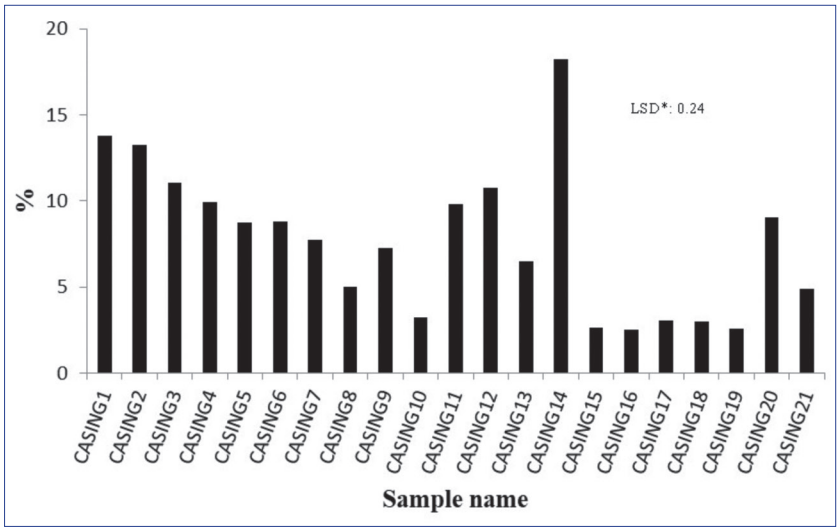

Fig 4. Elongation of break of natural casings (* LSD: least significant difference (calculated at α = 0.05)

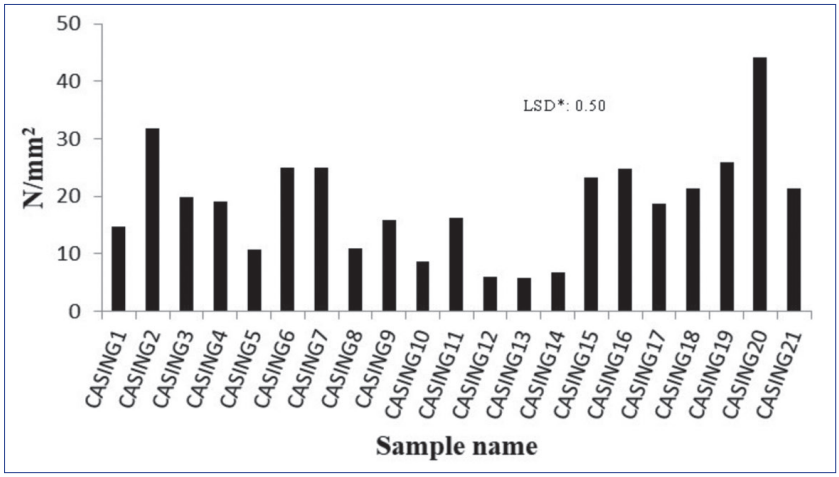

Fig 5. Tensile strength of natural casings (* LSD: least significant difference (calculated at α = 0.05)

Thirteen samples were contaminated with Coliform group bacteria. The highest coliform contamination was reported in Casing14 with 5.56 log cfu/g ($P<0.05$). Similarly, contamination of Casing6 was recorded as 5.54 log cfu/g ($P<0.05$). Casing9 was just the sample determined as clean from total aerobic mesophilic bacteria. Casing14 had the highest total aerobic mesophilic bacteria reported as 6.49 log cfu/g while Casing6 and Casing1 were recorded as 6.32 log cfu/g and 6.34 log cfu/g respectively ($P<0.05$). Casing4 was detected as clean by molds and yeasts. The highest contamination with mold and yeast was determined on Casing10 and Casing5 as 4.09 log cfu/g and 4.57 log cfu/g respectively ($P<0.05$). *Salmonella* spp. was detected on Casing13 and Casing19. Casings were reported as clean from *L. monocytogenes*.

DISCUSSION

In this study, the properties of natural casings ready to apply in the sucuk production in the market were investigated. Natural casings are derived from gastrointestinal system of farmed animals such as cattle and lamb where the quality and specifications could be effected by age, feed components, breeding, grown condition etc.[12,18]. Intestine is mostly processed to casings in unsuitable and unhygienic conditions in small scale enterprises with lack of standardization, thus, 21 casings were supplied from various regions in Turkey and determined mechanical and microbiological properties as a reference study for meat industry.

The ash content of the casings were varied between 0.61-3.07% ($P<0.05$). During the production of the casing, some solutions and preservative agents such as sodium chloride, phosphate salts, lactates, and organic acids are applied to extent shelf-life and protect from microbial spoilage [15,34]. Similarly, Wijnker *et al.*[35] indicated that treatment with mixture of phosphate and sodium chloride (NaCl) prevents some diseases such as foot-and-mouth disease and classical swine fever. However, usage of these agents in uncontrolled high levels may be cause of undesired residuals that is related with health risks for consumers

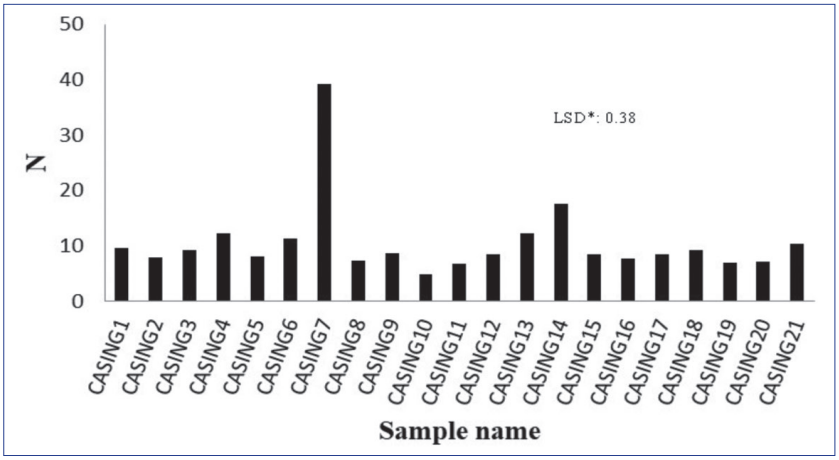

Fig 6. Breaking force of natural casings (* LSD: least significant difference (calculated at α = 0.05)

product in safety issues or to match traditional properties. Sucuk is defined as a fermented meat product where the humidity must be less than 40% in Turkish Food Codex [36]. In this point of view, water vapor permeability of the casings becomes essential to achieve high quality fermented product and to reach the required moisture content. Water Vapor Permeability of the casings was determined between 1.02-4.37 mg/cm²h (P<0.05). The porous structure that leads transition of water vapor increases the importance and demand of natural casings. Similarly, Djordevic *et al.*[11] and Simelana [37] indicate lower Water Vapor Permeability of artificial casings made of

Table 1. *Microbiological properties of natural casings (log cfu/g)*

Sample Name	*Escherichia coli*	Sulfide-reducing Anaerob	Coagulase Positive Staphylococci	Coliform	Total Aerobic Mesophilic	Mold - Yeast	*Salmonella* Spp.	*Listeria monocytogenes*
CASING1	3.30	<2.0	5.77	4.20	6.34	2.60	-	-
CASING2	<2.0	<2.0	3.63	<2.0	5.08	3.18	-	-
CASING3	<2.0	<2.0	4.70	2.20	5.56	2.70	-	-
CASING4	<2.0	<2.0	3.08	<2.0	3.38	<2.0	-	-
CASING5	4.85	<2.0	5.60	4.87	5.49	4.57	-	-
CASING6	4.60	<2.0	<2.0	5.54	6.32	3.60	-	-
CASING7	<2.0	<2.0	2.70	<2.0	2.70	3.04	-	-
CASING8	4.00	<2.0	5.72	5.08	5.51	4.05	-	-
CASING9	<2.0	<2.0	2.90	<2.0	<2.0	2.48	-	-
CASING10	4.48	<2.0	5.67	5.68	5.34	4.09	-	-
CASING11	<2.0	<2.0	<2.0	<2.0	5.34	2.60	-	-
CASING12	<2.0	2.04	<2.0	3.38	5.34	2.60	-	-
CASING13	3.38	<2.0	5.63	3.40	6.08	3.43	+	-
CASING14	3.51	<2.0	5.81	5.56	6.49	3.74	-	-
CASING15	2.62	2.79	<2.0	2.64	5.62	3.46	-	-
CASING16	<2.0	<2.0	<2.0	<2.0	3.86	2.95	-	-
CASING17	2.40	<2.0	3.32	2.96	5.52	2.99	-	-
CASING18	<2.0	<2.0	2.48	2.71	5.49	3.63	-	-
CASING19	<2.0	<2.0	2.30	<2.0	4.36	2.36	+	-
CASING20	<2.0	<2.0	2.04	<2.0	4.43	2.73	-	-
CASING21	<2.0	<2.0	2.70	2.06	5.28	2.93	-	-
LSD*	0.57	-	0.34	0.25	0.30	0.19	-	-

** **LSD:** least significant difference (calculated at α = 0.05)*

and technological inadequacy of the product.

Fermentation is one of the crucial steps where the specific taste and texture formed by the activity of starter culture and chemical changes under controlled conditions such 90% relative humidity [7,8]. Moreover, some restrictions are available for the last synthetic and polymeric materials that lead a decrease the potential to usage in fermented meat products.

Thickness of casings were measured between 0.02 and 0.16mm (P<0.05). Although artificial casings can be produced under controlled conditions, Simelena [37] reported difference on thick-ness of heat-cured collagen

films where the various temperatures were applied.

Responsibility of the casings is shaping the dough, supplying suitable conditions for fermentation and maturation and protecting the product from external influences from the production step to the fork. Casing should resist the pressure while meat dough is stuffing which increases the importance of mechanical properties in meat industry from the economic and technical aspects [17,18]. Elongation of the casings were determined between 2.5-18.21% while tensile strength was reported between 5.88-44.08 N/mm^2 (P<0.05). Simelena [37] was investigated elongation and tensile strength of whey protein isolate films and collagen films as artificial casing and reported as approximately 28-58% and 2.70-13.00 respectively. Increased on elongation may be correlated with decreased tensile strength as suggested by Simelena [37]. Moreover, Amin and Ustunol [16] investigated some properties of whey protein based films as a replacement for collagen and natural casings and stated the correlation of increased tensile strength with decreased percentage at break and water vapor permeability. In that study, cross-linking of proteins by enzymatic, physical or chemical ways was suggested to increase strength mechanical properties while decreasing water vapor permeability which is a disadvantage of artificial casings for traditional meat products [16]. Mechanical properties of casings may be influenced by age, genus, gender and feed content and conditions and agents applied during processing [18].

Intestine is contaminated with microorganisms by its nature. However, microbial quality of natural casing is affected by processing, transportation and storage conditions as a last product. On the other hand, casing is in contact with meat dough during the process such as fermentation and storage period that leads to another microbial activity [10,18]. Intestine track of farm animals such as cattle knows as the natural reservoir of *E. coli* which may be related with food-borne diseases occurred as bloody diarrhea, abdominal cramps and fever. Akkaya *et al*.[38] investigated the hygiene and sanitary conditions of processing beef carcasses and reported 3.2% *E. coli* O157 in 250 samples while 43% of the casings were contaminated in our study. Another pathogen group related with insufficient sanitary system is stated as sulfide reducing anaerobic bacteria where the intestine track is stated as a source. *Clostridium* spp. can produce spores and cause food-borne diseases even if in anaerobic conditions [10]. Two casings were contaminated with this pathogen shown in the *Table 1*. Coagulase positive staphylococci was detected in 16 samples (P<0.05). In a study, contamination with coliform bacteria was determined as 5.54 log cfu/g and 3.93 log cfu/g on pork and lamb casings respectively [18]. In another study, combination of washing with de-ionized water and 5kGy gamma radiation was successful to eliminate coliform bacteria on lamb and pork casings [6]. Thirteen samples were contaminated with coliform where the highest contamination was reported as 5.56 log cfu/g (P<0.05) as shown in *Table 1*. Moreover, total aerobic mesophilic bacteria was reported in 20 cattle casings where the highest count was 6.49 log cfu/g (P<0.05) *(Table 1)*. Similarly, initial total aerobic bacteria was determined as 6.3 log cfu/g and 5.9 log cfu/g in pork and lamb casings while contamination was reported as 6.78 log cfu/g and 6.61 log cfu/g in pork and lamb casings in another study [18]. Mold and yeast might be deterioration on quality and hazardous to human health that should be under control and safety issues. Mold and yeast were reported in 20 samples (P<0.05) shown as in *Table 1*. Akkaya *et al*.[38] investigated 250 beef carcasses and determined 10% samples contaminated with *Salmonella* spp. while 9.5% of cattle casings was contaminated shown in *Table 1*. Human salmonellosis caused by *Salmonella* spp. is one of the reasons of diarrhea that kills three million children every year in developed countries. Unfortunately, *Salmonella* spp. was reported in fecal samples in many countries that spreads easily in the processing environment related with animals [38]. Wjinker [6] determined *Salmonella* spp. in beef, sheep and hog casings. Well-known foodborne disease of *L. monocytogenes* is called listeriosis where the pathogen can resist to high level of salt as high as 12-13%, low level of water activity as low as 0.90 and pH as low as 4.4 and tolerate to refrigeration conditions in long term [39]. *Listeria monocytogenes* was not detected in our samples although Wjinker [6] reported the pathogen in beef casings. Moreover, Akkaya *et al*.[38] determined *L. monocytogens* 6.8% of beef carcasses. Three contamination phases defined as before, during and after slaughter was stated by Akkaya *et al*.[38].

Intestine is very sensitive part of the animal that should be processed as soon as possible after slaughtering because of the potential risk on the microbial contamination. Therefore, some food safety assurance systems such as Hazard Analysis of Critical Control Point (HACCP), Good Hygiene Practice (GHP), Food Safety Management System (ISO 22000) should be applied during slaughtering, handling and processing of natural casings. Difficulties to have standardization on industrial production of natural casings lead to use some agents to improve mechanical properties and microbial stability. If the processes do not carry under proper conditions, economical loss may be occur.

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