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Evaluation of Quality Characteristics of Commercial Fermented Sausages (Sucuk and Heat-Treated Sucuk)

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ARTICLE INFO	A B S T R A C T						
Research Article	The study aimed to evaluate the pH, water activity (a _w), residual nitrite, lactic acid bacteria, <i>Micrococcus/Staphylococcus</i> , Enterobacteriaceae and yeast-mould in fermented sausage samples						
Received : 03-07-2023 Accepted : 28-09-2023	from different firms. A total of 30 sucuk and 30 heat-treated sucuk samples were taken different brands with different batch numbers. According to analysis results, all samples, exception of some heat-treated sucuk samples from one brand, provided pH values that wer						
<i>Keywords:</i> Nitrit pH Lactic acid bacteria Sucuk Heat-treated sucuk	the permitted limit of regulation. For sucuk, mean aw value of only one brand was below 0.90, while a_w values for heat-treated sucuk were in the range of 0.928 to 0.957. All samples had residual nitrite levels less than 15 mg/kg (in the range of 7.84-14.80 mg/kg). Yeast-mould and Enterobacteriaceae numbers were often below <2 log cfu/g. The number of <i>Staphylococcus and Micrococcus</i> showed a wide variation in both products which was <2 - 5.96 log cfu/g for sucuk and <2 - 7.85 log cfu/g for heat-treated sucuk. Lactic acid bacteria counts varied between 2-<4.0 log cfu/g in 40% of heat-treated sucuk samples. In sucuk, the number of lactic acid bacteria was <6 log cfu/g in 23.33% of the samples, and 6-<8 log cfu/g in 50% of the samples.						
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Introduction

Fermented sausages that maintain popular with their typical flavors and aromas may differ depending on raw material quality, added ingredients, and process conditions (Yu et al., 2021). Fermented sausages are classified by water activity (a_w), weight loss, moisture, moisture:protein ratio, geographical region, degree of fat comminution. They are categorized into three groups based on their moisture content: high humidity (50-60% water), semi-dry (35-50%), and dry (20-35%) sausages (Campbell-Plantt, 1995). Fermented sausages involve dry or semi-dry products such as chorizo, hard salami, peperroni, salami, summer sausage, Rohwurst, cervelat (Kaya and Kaban, 2020). However, in many countries, especially in traditional products, there isn't broad, explicit and uniform distinction among semi-dry and dry fermented sausages, and even many products with the same name can be manufactured as "dry" or "semi-dry" fermented sausages (Lücke, 2107). On the other hand, high humidity fermented sausages such as Mettwurst and Teewurst, as well as Sobrasada made in several countries, are soft-consistent products made with fermentation (Kaya and Kaban, 2020).

Nitrite, pH, aw and low oxygen levels are important hurdle effects to microbial stability in fermented sausages. In fermented sausages, salt (2-3%) exhibits partial bacteriostatic activity and lowers the water activity of the batter to 0.955-0.960, depending on the fat ratio in the formulation (Kaya and Kaban, 2020). aw of dry fermented sausages is less than 0.90, and no smoking or heat treatment is used in the manufacturing process. aw in semidry fermented sausages ranges between 0.90-0.95, and a heat treatment of 60-68°C is typically used (Caplice and Fitzgerald, 1999). The rate and degree of acid production during fermentation in fermented sausages are critical for product sensory characteristics and product safety. For controlled acid formation in industrial manufacturing, starter culture has been involved, as opposed to the spontaneous flora or back slopping application that occurs in traditional production (Kaban et al., 2022a). Semi-dry fermented sausages differ substantially from dry fermented sausages in that they have a strong distinctive flavor due to the rapid fermentation. The final pH of these products ranges from 4.7 to 5.4. However, the pH varies between 5.2 and 5.8 in dry fermented sausages, which ripened longer

than semi-dry fermented sausages (Vignolo et al., 2010). According to the Turkish Food Codex Communiqué on meat, prepared meat mixtures and meat products, the maximum pH value authorized for sucuk is 5.4, while the maximum pH value allowed for heat-treated sucuk is 5.6 (Anonymous, 2019). In fermented sausages, nitrite and/or nitrate are utilized as curing agents depending on the product type and processing conditions. The fermentation process is the most important part in the production, and the level of nitrite acts as a hurdle effect for many pathogenic bacteria. Ingoing nitrite level permitted in these products is limited to 150 mg/kg. Low residual nitrite levels in the final product are also crucial for reducing the nitrosamine risk (Sallan et al., 2023).

Lactic acid bacteria and Gram (+) catalase (+) cocci are technologically essential microorganisms for fermented sausages (Kaban et al., 2022a). Lactic acid bacteria, which multiply up to around 10^8 cfu/g in the fermentation stage, retain their survival to a great extent in the further stages of ripening (Dalmış and Soyer, 2008; Soyer et al., 2005). The number of Gram (+) catalase (+) cocci in these products varies depending on the acidification during fermentation (Akköse et al., 2023). These microorganisms, which are important in the microbiota of fermented sausages, are found at lower levels in heat-treated sucuk. On the other hand, the growth of Gram (-) flora such as Enterobacteriaceae and *Pseudomonas* is suppressed by hurdles such as nitrite, pH, redox potential, a_w and their combinations (Kaya and Kaban, 2020).

Two different types of fermented sausage are manufactured in Türkiye, namely sucuk and heat-treated sucuk. While fermentation and drying procedures are used in sucuk production, fermentation, heat application, and drying procedures are applied in the production of heattreated sucuk (Armutçu et al., 2020). The moisture:protein ratio and pH value of sucuk are the most crucial product attributes. The moisture:protein ratio should be less than 2.5, and the pH should be no more than 5.4 in sucuk. Heattreated sucuk is permitted to have a greater moisture/protein ratio (3.6) and a higher pH (up to 5.6) value (Anonymous, 2019). Many studies have been carried out on the quality attributes of sausage samples obtained from market (Yücel and Karaca, 1993; Sancak et al., 1996; Atasever et al., 1998; Con et al., 2002; Kaban and Kaya, 2008; Sezer et al., 2013; Büyükünal et al., 2016; Kızılkaya et al., 2023). On the other hand, there are little information on quality attributes of heat-treated sucuk obtained from the market (Sezer et al., 2013; Kaban et al., 2022b). No research investigating the variations in quality traits between the same brand's sucuk and heat-treated sucuk based on different production durations has been also found in the literature.

The aim of the study is to determine and analyze pH and water activity values as well as residual nitrite content of sucuk and heat-treated sucuk samples received from various firms at different times. In addition, it is aimed to determine the numbers of technologically important microorganisms (Lactic acid bacteria and *Micrococcus/Staphylococcus*), Enterobacteriaceae and yeast-moulds in these products.

Material and Method

Material

Samples of sucuk and heat-treated sucuk that were collected from ten different brands in Türkiye were utilized as study materials. Vacuum packed samples were gathered from retail establishments while considering different batch numbers at three different times. The following analyses were performed on a total of 60 samples (10 sucuk and 10 heat-treated sucuk samples) which are 3 samples from each brand.

Method

Physicochemical Analysis

Water Activity (a_w) : The equipment (TH500 a_w Sprint, Novasina) was utilized to determine the a_w value of the samples. The instrument was calibrated using six different salt solutions prior to use. At 25°C, water activity was measured (Kaban et al., 2022a).

pH: 10 g samples were weighed into jars and homogenized with distilled water (100 mL) with ultra turrax (IKA Werk T25, Germany) to detect pH value. The pH meter (Mettler Toledo, Greifensee, Switzerland) was calibrated using buffer solutions of pH 4.0 and 7.0 before analysis (Kaban et al., 2022a).

Residual Nitrite: The samples homogenized were weighed at 10 g with a 0.1 mg reliability. Ultrapure water (50 mL, 50-60°C) was added and then well mixed using glass baguette. The mixture was then transferred to flasks of 200 mL capacity. Acetonitrile (50 mL) was added to the flask following 15 minutes of mixing, after which 200 mL of ultrapure water was added. The resulting samples were filtered using nitrite-free/nitrate-free filter paper, and the filtrates were then put in vials after being run through a 0.45 μ m filter. Using HPLC/DAD, the residual nitrite concentration was calculated. The flow rate in the column was set to be 2 mL/min and the nitrite standard was used for identification. The results were obtained in mg/kg (NKML, 2000).

Microbiological Analysis

Sausage sample of 25 g was homogenized with 225 mL of sterile physiological saline in Stochmacher (Lab Stomacher Blander 400 - BA 7021). From this homogenate, serial dilutions were prepared. The lactic acid bacteria, *Micrococcus/Staphylococcus*, Enterobacteriaceae, and yeast-mould counts of the samples were determined using the spread plate technique. The results are given as log cfu/g.

Lactic Acid Bacteria: The number of lactic acid bacteria was determined using MRS Agar (de Man Rogosa Sharpe Agar, Merck). The petri plates were incubated in anaerobic conditions (Anaerocoult A, Merck) for 2 days at 30°C after adding 0.1 mL of each dilution to the medium. Catalase (-) colonies were counted at the end of the incubation to determine the number of lactic acid bacteria (Y1lmaz Oral and Kaban, 2021).

Micrococcus/Staphylococcus : MSA (Mannitol Salt Phenol Red Agar, Merck) was used for the number of *Micrococcus/Staphylococcus* and the inoculated plates were incubated at 30°C for two days. Following the incubation, the number was detected by considering the catalase (+) cocci (Yılmaz Oral and Kaban, 2021). *Enterobacteriaceae:* In order to determine the Enterobacteriaceae number of the samples, 0.1 mL of the dilutions prepared were transferred to VRBD (Violet Red Bile Dextrose, Merck) agar plates and incubation was conducted at 30°C for 2 days under anaerobic conditions (Anaerocoult A, Merck). After incubation, the number of Enterobacteriaceae was detected by counting red, rose red or purple colonies larger than 1 mm (Gökalp et al., 2015).

Yeast and Mould: To count the number of yeast and mould, RBC (Rose Bengal Chloroamphenicol, Merck) was utilized. The dilution solutions were added to the petri plates, where they underwent a 5-day incubation period at 25°C. The colonies were counted after 5 days (Gökalp et al., 2015).

Statistical Analysis

In the study, fermented sausage type (sucuk and heattreated sucuk) and brand (A, B, C, D, E, F, G, H, J, K) were taken as factors and the trials were based on randomized complete block design in 2x10 factorial design with 3 replications. Sampling was carried out at three different times with different batch numbers, and thus, a total of 60 samples, 30 of which were heat-treated sucuk and 30 sucuk samples, were examined. Analysis of variance was applied to pH, a_w and residual nitrite results in the study and the means of the results were compared with Duncan's multiple range test (Version 24, SPSS Inc., Chicago, IL, USA). For lactic acid bacteria and *Micrococcus/ Staphylococcus* numbers, graphs were prepared and evaluated using frequency distribution.

Results and Discussion

Physicochemical Results

The effects of fermented sausage type and brand factors on pH and a_w values are given in Table 1. According to the results, both fermented sausage type and brand factor showed a significant effect on pH and a_w values (P<0.05). The lowest mean values in terms of aw and pH was found in sucuk. The mean pH value of both sucuk and heattreated sucuk is under the limit values given in the Turkish Food Codex Communiqué on meat, prepared meat mixtures and meat products (maximum pH: 5.4 for sucuk; maximum pH: 5.6 for heat-treated sucuk) (Anonymous, 2019). There were also differences between the brands in terms of pH value. The brand \times fermented sausage type interaction was given in Figure 1a.As can be seen from Figure 1a, pH values showed a wide variation in both types of fermented sausage samples. However, only pH values of sucuk and heat-treated sucuk samples from the B and E brands differed statistically. The mean pH in heat-treated sucuk sample of B brand (pH=6.1) was higher than the communiqué's limit value. In other brands, the pH value is below the specified limit values (Figure 1a) (Anonymous, 2019). Previous investigations on sucuk yielded similar findings (Yücel and Karaca, 1993; Kaban and Kaya, 2008). Additionly, Sancak et al. (1996) reported that the pH value for sucuk ranged between 4.99-6.21 (mean pH: 5.50), whereas Atasever et al. (1998) stated that the pH value for sucuk ranged between 4.45-6.43 (mean pH: 5.24). Mean pH of sucuk was found by Kızılkaya et al. (2023) to be below 5.4. However, in another research, the mean pH of sucuk was 6.18 (min: 4.94, max: 6.97) while heat-treated sucuk was 6.74 (min: 6.4, max: 6.92) (Sezer et al., 2013). Vural (1998) detected a pH range of 5.16 to 5.55 for semidry fermented sausage. Kaban et al. (2022b) also reported that pH value of commercial heat-treated sucuk samples varied between 4.28-5.47. High pH in sucuk and heattreated sucuk indicates that either fermentation is not performed sufficiently or not performed at all. The pH value is an important hurdle for fermented sausages such as sucuk and heat-treated sucuk (Leistner and Gorris, 1995; Kaya and Kaban, 2020). In the present study, only the heattreated sucuk of B brand did not comply with the regulation (Figure 1a) (Anonymous, 2019).

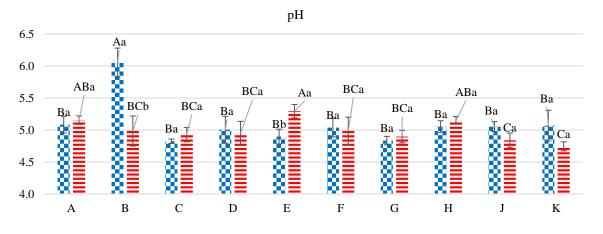
The brand \times fermented sausage type interaction, which was found to have a very significant effect on the a_w value of the samples, is given in Figure 1b. aw of sucuk and heattreated sucuk was in the range of 0.880-0.950 and 0.928-0.957, respectively. As can be seen from Figure 1b, in brands of D, G and K, sucuk gave lower aw value than heattreated sucuk. On the other hand, there was no statistically significant difference in aw among the samples of sucuk and heat-treated sucuk (P>0.05) (Figure 1b). As can be understood from the results, the aw values of 0.94 - 0.95 indicate a limited degree of drying in these products. The aw of dry fermented sausages is below 0.90, whereas aw of semi-dry fermented sausages ranged from 0.90 to 0.95 (Caplice and Fitzgerald, 1999). In the present study, the mean aw of sucuk was found below 0.90 only in D brand (mean aw:0.88). In K brand, the mean aw of sucuk was 0.905 (Figure 1b). Kızılkaya et al. (2023) found a_w values in the range of 0.937-0.961, while Kaban and Kaya (2008) in the range of 0.801-0.913 in the sucuk samples obtained from the market. As can be seen from Figure 1b, the mean aw of heat-treated sucuk was quite high in D brand (a_w:0.957) and G brand (a_w:0.951). Kaban et al. (2022b) reported lower aw values (min aw: 0.913, max aw: 0.940 mg/kg) for heat-treated sucuk. pH and aw are considered as two significant hurdles in fermented sausages. The pH level of semi-dry fermented sausages with a higher a_w than dry sausages is generally below 5.0 (Sallan and Kaya, 2021). In the present study, while the a_w is high in the majority of the brands, the pH value is below 5.0. According to the a_w and pH results of this study, both sucuk and heat treated sucuk should be kept in cold storage.

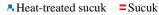
In the study, there was no significant effect of fermented sausage type and brands factors on residual nitrite (P>0.05). Similarly, there was no significant effect of brand × fermented sausage type interaction on residual nitrite (P>0.05) (Table 1). Residual nitrite for both sucuk and heat-treated sucuk was below 15 mg/kg (Table 1). In a previous study on sucuk, higher residual nitrite amounts (min: 29.66 mg/kg, max: 89.0 mg/kg, mean: 46.87 mg/kg) were determined (Yücel and Karaca, 1993). Büyükünal et al. (2016) determined the mean nitrite level for sucuk as 24.83 mg/kg. In addition, Kızılkaya et al. (2023) determined the residual nitrite content in sucuk samples to be 5.81–17.65 mg/kg, and Kaban et al. (2022b) found the residual nitrite content in all heat-treated sucuk samples to be below 10 mg/kg. Low residual nitrite level is an important factor in terms of nitrosamine in fermented sausages. It is even more important to have low residual nitrite levels in products cooked before consumption such as sucuk and heat-treated sucuk (Sallan et al., 2023). Residual nitrite in fermented sausages, however, is vital for color stabilization, and a residual nitrite in the level of 10-15 mg/kg is widely suggested as a reservoir for the regeneration of cured meat color (Alahakoon et al., 2015).

Treatments		n pH a _w		Residual Nitrite (mg/kg)		
Fermented Sausage Type (ST)		-				
Heat-treated sucuk		5.090 ^a	0.941 ^a	10.671ª		
Sucuk		4.992 ^b	0.929 ^b	11.053ª		
SEM		0.021	0.001	0.538		
Significance		**	**	ns		
Brand (Br)						
А	6	5.118 ^b	0.943 ^{ab}	9.602ª		
В	6	5.517ª	0.929 ^{cd}	9.117ª		
С	6	4.875 ^c	0.937 ^{bc}	10.868^{a}		
D	6	4.975 ^{abc}	0.919 ^e	10.778ª		
E	6	5.095 ^{ab}	0.938 ^{abc}	11.292ª		
F	6	5.010 ^{abc}	0.942^{ab}	11.855ª		
G	6	4.865 ^c	0.948 ^a	11.343ª		
Н	6	5.102 ^{ab}	0.943 ^{ab}	11.127ª		
J	6	4.947 ^{bc}	0.930 ^{cd}	14.800 ^a		
K	6	4.905°	0.920 ^{de}	7.837ª		
SEM		0.048	0.003	1.202		
Significance		**	**	ns		
ST×Br		**	**	ns		

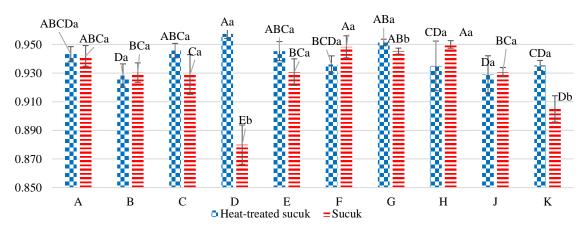
Table 1. Overall effect of fermented sausage type and different brands on pH, a_w and residual nitrite values in samples

ns: not significant; **: P<0.01





(a) a_w



⁽b)

Figure 1. Mean pH (a) and a_w (b) values of heat treated sucuk and sucuk samples obtained from different brands (A-D: Different capital letters indicate significant differences between brands for fermented sausage type. a-b: Different lower case letters indicate significant differences between fermented sausage types for brand)

Microbiological Results

Lactic acid bacteria, Micrococcus/Staphylococcus, yeast-mould and Enterobacteriaceae counts of the brands with respect to the fermented sausage type are given in Table 2. Figures 2a and 2b, respectively, show frequency distribution graphs of lactic acid bacteria and Micrococcus/Staphylococcus counts of the samples. As can be shown from Table 2, lactic acid bacteria count in sucuk and heat-treated sucuk showed a wide variation. In addition, significant variations were also found between different batch numbers of the same brand (Table 2). For example, the lowest number of lactic acid bacteria was 3.80 log cfu/g and the highest count was obtained as 7.88 log cfu/g for sucuk from A brand. A similar situation exists in sucuk samples of B brand (min:2.48 log cfu/g, max: 7.90 log cfu/g). This circumstance was interpreted as an evidence that these firms do not produce in a standardized manner. As can be seen in Figure 1a, which was created by considering all sucuk and heat-treated sucuk samples, the number of lactic acid bacteria were found in 50% of the 30 sucuk samples at the level of 6.00 - 8.00 log cfu/g and 26.67% of sucuk samples at the level of $> 8.00 \log cfu/g$. Lactic acid bacteria constitute the dominant flora in fermented sausages without heat treatment such as sucuk and their numbers vary between 10⁶-10⁸ cfu/g (Gökalp et al., 2004; Kaban, 2013). In a previous research, it was reported that total aerobic mesophilic bacteria count in sucuk varied between 3.0x104 - 2.2x108 cfu/g (mean 2.9x10⁷ cfu/g) (Çon et al., 2002). Kaban and Kaya (2008) detected more than 1×10^8 cfu/g lactic acid bacteria number in all of the sucuk samples examined. In a study conducted by Sancak et al. (1996), it was reported minimum, maximum and mean lactic acid bacteria counts as 1.7 x 10⁶ cfu/g, 2.4×10^9 cfu/g and 3.3×10^8 cfu/g, respectively. In the current study, 10% of the samples had less than 4.0 log cfu/g of lactic acid bacteria and 13.33% of the samples had lactic acid bacteria counts between 4.0 - <6.0 log cfu/g for sucuk (Figure 2a). This result indicates that these products were subjected to a heat treatment. Indeed, it is stated in the regulation that this product is a fermented meat product without heat treatment (Anonymous, 2019). In heat-treated sucuk, 40% of the samples varied between 2.0 - <4.0 log cfu/g and 40% varied between 4.0 - <6.0 log cfu/g. In this product, only 3.3% of the samples had $> 8.0 \log cfu/g$ of lactic acid bacteria count. These results are thought to be due to the difference in the heat treatment parameters applied in the firms, and also the formulation. Lactic acid bacteria are microorganisms that contribute significantly to both product safety and sensory properties by producing acid during fermentation (Gökalp et al., 2004).

Table 2. Lactic acid bacteria, *Micrococcus/Staphylococcus*, yeast-mould and Enterobacteriaceae counts of the brands with respect to the fermented sausage type

Brand	to the termented sausage type Heat-treated Sucuk					Suc	uk	
	LAB	MS	EB	YM	LAB	MS	EB	YM
А	7.38	4.23	$< 10^{2}$	$< 10^{2}$	3.80	5.30	$< 10^{2}$	$< 10^{2}$
	7.60	3.30	$< 10^{2}$	$< 10^{2}$	7.88	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$
	7.77	3.85	$< 10^{2}$	$< 10^{2}$	6.93	4.70	$< 10^{2}$	$< 10^{2}$
В	4.11	$< 10^{2}$	<10 ²	$< 10^{2}$	2.48	3.94	$< 10^{2}$	$< 10^{2}$
	5.30	4.60	$< 10^{2}$	$< 10^{2}$	7.43	5.60	$< 10^{2}$	$< 10^{2}$
	7.85	4.69	$< 10^{2}$	$< 10^{2}$	7.90	5.20	$< 10^{2}$	$< 10^{2}$
С	3.65	7.85	$< 10^{2}$	3.66	8.23	5.96	$< 10^{2}$	$< 10^{2}$
	3.69	2.95	$< 10^{2}$	$< 10^{2}$	6.78	5.70	$< 10^{2}$	$< 10^{2}$
	4.69	3.00	$< 10^{2}$	$< 10^{2}$	7.30	4.00	$< 10^{2}$	$< 10^{2}$
D	2.60	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$	8.30	3.30	$< 10^{2}$	$< 10^{2}$
	4.65	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$	8.30	4.78	$< 10^{2}$	$< 10^{2}$
	3.00	6.90	$< 10^{2}$	$< 10^{2}$	8.78	5.71	$< 10^{2}$	$< 10^{2}$
Е	2.60	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$	8.52	4.85	2.47	$< 10^{2}$
	4.40	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$	6.39	2.30	$< 10^{2}$	$< 10^{2}$
	2.48	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$	7.35	3.00	$< 10^{2}$	$< 10^{2}$
F	3.48	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$	7.00	$< 10^{2}$	$< 10^{2}$	2.60
	4.60	3.30	$< 10^{2}$	$< 10^{2}$	6.60	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$
	3.60	3.30	$< 10^{2}$	$< 10^{2}$	8.54	3.30	$< 10^{2}$	$< 10^{2}$
G	2.47	3.60	$< 10^{2}$	$< 10^{2}$	4.77	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$
	2.85	3.77	$< 10^{2}$	$< 10^{2}$	3.60	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$
	5.14	3.74	$< 10^{2}$	$< 10^{2}$	4.30	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$
Н	7.90	6.08	2.69	$< 10^{2}$	6.60	3.00	$< 10^{2}$	3.17
	8.10	5.69	2.00	$< 10^{2}$	4.85	$< 10^{2}$	$< 10^{2}$	3.60
	4.60	4.30	$< 10^{2}$	$< 10^{2}$	4.30	$< 10^{2}$	$< 10^{2}$	2.60
J	4.88	5.00	$< 10^{2}$	$< 10^{2}$	8.48	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$
	4.77	4.98	$< 10^{2}$	$< 10^{2}$	7.69	$< 10^{2}$	$< 10^{2}$	3.48
	2.00	3.93	$< 10^{2}$	$< 10^{2}$	7.81	$< 10^{2}$	$< 10^{2}$	$< 10^{2}$
К	4.97	3.90	<10 ²	$< 10^{2}$	7.90	5.30	<10 ²	<10 ²
	4.08	3.92	$< 10^{2}$	$< 10^{2}$	8.48	4.95	$< 10^{2}$	$< 10^{2}$
	2.30	3.60	$< 10^{2}$	$< 10^{2}$	7.71	4.54	$< 10^{2}$	$< 10^{2}$

LAB: Lactic Acid Bacteria (log cfu/g); MS: *Micrococcus/Staphylococcus* (log cfu/g); EB: Enterobacteriaceae (log cfu/g); YM: Yeast-Mould (log cfu/g)

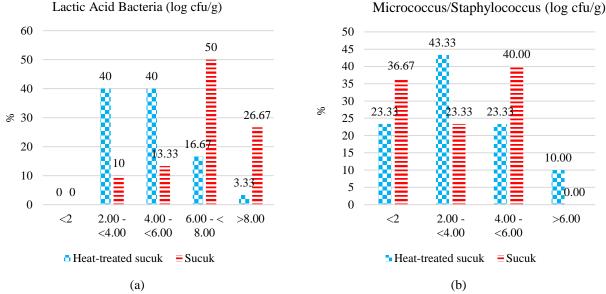


Figure 2. Frequency distribution of heat-treated sucuk and sucuk samples in terms of lactic acid bacteria and Micrococcus/Staphylococcus numbers

Micrococcus/Staphylococcus numbers of sucuk and heat-treated sucuk samples showed a large variation between brands, and even in different samples of the same brand. Micrococcus/Staphylococcus counts in all heattreated sucuk samples of E brand were found below the detectable limit. In two samples of E brand and in one sample each of B and F brands, the number was also found below the detectable limit. As can be seen in Figure 2b, the number of Micrococcus/Staphylococcus was below the detectable limit in 23.3% of the heat-treated sucuk samples. This result is considered to be a good indicator that heat treatment is involved in the sucuk production. In addition, 40% of the samples gave Micrococcus/Staphylococcus Since counts of 4.0 - <6 log cfu/g in sucuk. *Micrococcus/Staphylococcus* are acid sensitive microorganisms, rapid acidification during fermentation can significantly inhibit the development of these products (Akköse et al., 2023). 23.3% of the sucuk samples gave *Micrococcus/Staphylococcus* counts of 2.0 - <4 and, 36.6% gave Micrococcus/Staphylococcus counts below the detectable limit (Figure 2b). These findings suggest that heat treatment is included in the sucuk process rather than a rapid acidification. Micrococci and staphylococci are technologically important microorganisms in these products and contribute to product quality with nitrate reductase and catalase activities. These microorganisms have also effects on flavour with their proteolytic and lipolytic activities (Kaya and Kaban, 2020).

In heat-treated sucuk, Enterobactericeae were detected at a level of 2 log cfu/g in two samples of only one brand (H), and the number was below the detectable number in the other samples. In sucuk, Enterobactericeae was detected only in 1 sample of E brand (Table 2).

These results show that some firms also apply heat treatment to the sucuk. Çon et al. (2002) reported that the number of Enterobacteriaceae in sucuk varied between $<10 - 1.1 \times 10^4$ cfu/g (mean: 1.3×10^3 cfu/g). Kaban and Kaya (2008), on the other hand, found the Enterobacteriaceae count under the detectable limit in the samples of 5 brands (10 samples) and 10^2 cfu/g in two brands (4 samples).

Yeast-mould count of heat-treated sucuk was found below the detectable limit in all samples of the other brands except C. In sucuk, the number of yeast-mould was determined at the level of 10^2 - 10^3 cfu/g in all three samples of H brand. In addition, it was determined at the level of 10^2 cfu/g in 1 sample of F brand and 10^3 cfu/g in the sample of J brand (Table 2). Con et al. (2002) determined the number of yeast-mould in sucuk between $<10 - 1.4 \times 10^5$ cfu/g (mean: 1.2×10^4 cfu/g) and Sancak et al. (1996) determined between 10^2 - 10^6 cfu/g (mean: 10^5 cfu/g). In the research conducted by Atasever et al. (1998), it was also determined as 6.4×10^4 cfu/g. In our study, the fact that the number of yeast-mould in sucuk is generally below the detectable number indicates the application of heat treatment in sucuk production.

Conclusion

pH is an important criterion for sucuk and heat-treated sucuk. The mean pH value was found below 5.4 in all sausage samples obtained from different firms. The pH value of the heat-treated sucuk is also below the limit value (5.6) in all except one brand (B). However, in terms of a_w value, a wide variation was detected in both the sucuk and heat-treated sucuk. In sucuk, the mean aw value is below 0.90 in only one brand (D). In another brand (K), the mean a_w value was found to be 0.905. In other brands, high a_w values were determined, including 0.95. These results indicate that these products were not subjected to adequate drying process. In heat-treated sucuk, the a_w value varied between 0.928-0.957. However, the residual nitrite level in all samples was less than 15 mg/kg. Enterobacteriaceae and yeast-mould counts in heat-treated sucuk and sucuk were generally below the detectable limit. Lactic acid bacteria counts were <6 log cfu/g in 23.33% of the sucuk samples and <6 log cfu/g in 80% of the heat-treated sucuk samples. In 23.3% of the sucuk samples, the Micrococcus/Staphylococcus count was 2 - <4 log cfu/g, while in 36.6%, the count was below the detectable limit. These findings suggest that heat treatment was applied in the sucuk production process. On the other hand, considerable differences were obtained between different batch numbers of the same product in some brands. This demonstrates that these firms do not produce in a standardized manner.

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