

Effects of *Staphylococcus carnosus* on Quality Characteristics of *Sucuk* (Turkish Dry-Fermented Sausage) During Ripening

Güzin Kaban* and Mükerrerrem Kaya

Department of Food Engineering, Faculty of Agriculture, Atatürk University, Erzurum 25240, Turkey

Abstract In this study, *Staphylococcus carnosus* isolated from traditional *sucuk* (Turkish dry-fermented sausage) was used in combination with *Lactobacillus plantarum* as a lactic culture in *sucuk* production. *Sucuk* produced with only *L. plantarum* was evaluated as a control group. Microbiological, physicochemical, and volatile profile characteristics of *sucuk* samples were investigated during ripening. In both sausages with *S. carnosus* and control group, pH value decreased to below 5.0 at the 3rd day. In all samples, Aw value decreased as the ripening time progressed. Sausages with *S. carnosus* showed the higher nonprotein nitrogen (NPN) value than control group. However, the highest mean value for thiobarbituric acid reactive substances (TBARS) was observed in control group. Enterobacteriaceae dropped to undetectable levels at the 3rd day in both groups. *S. carnosus* increased approximately 1 log unit within the first 3 days of the fermentation. In the presence of *S. carnosus*, significant changes were observed in only a few volatile compounds.

Keywords: *Staphylococcus carnosus*, volatile compound, Turkish dry-fermented sausage (*sucuk*), gas chromatography/mass spectrometry, solid phase microextraction

Introduction

Lactic acid bacteria (*Lactobacillus* and *Pediococcus*) and catalase positive cocci (coagulase negative *Staphylococcus* and *Kocuria*) are two important groups of bacteria in fermented sausage manufacture. Lactic acid bacteria accumulate lactic acid to levels that inhibit pathogenic and spoilage flora and coagulate soluble meat proteins, thereby reducing water binding capacity and facilitating drying of the product (1). Catalase positive cocci play an important part in the formation of the color by reducing nitrate to nitrite, and proteolytic and lipolytic activities contributing to the aroma of fermented sausages (2-4). They also possess catalase activity involved in stabilization of color and prevention of rancidity (5).

Coagulase negative staphylococci (CNS) and *Kocuria* are catalase positive cocci used as starter culture in manufacturing fermented sausage. Among Gram-positive catalase positive cocci, *Staphylococcus xylosus* is the predominant species in many naturally-fermented Italian (6-9) and Spanish (10-12) sausages, but *Staphylococcus saprophyticus* and *Staphylococcus carnosus* are the most frequent species isolated from traditional Greek sausages (13). Although *S. xylosus* was always isolated from *sucuk*, the most popular dry-fermented sausage in Turkey, *S. carnosus* is rarely isolated as a part of the indigenous flora of this sausage (14). There have been many reports about the effects of *Staphylococcus* species on the formation of volatile compounds in fermented sausage or model system (15-23).

S. carnosus is an important species widely used in industrial manufacturing of *sucuk*. Though, there are no

studies on the behavior and effects of *S. carnosus* in *sucuk*. Thus, the aim of the study was to determine the effects of *S. carnosus*, isolated from *sucuk*, on the microbiological, physicochemical, and volatile profile characteristics of *sucuk* during ripening.

Materials and Methods

Preparation of strains In this research, *Staphylococcus carnosus* MK93 and *Lactobacillus plantarum* GM77 (as a lactic culture), isolated from traditional *sucuk* by Kaban (23), were used. *S. carnosus* MK93 is a facultative anaerobe, and has lipolytic, proteolytic, and nitrate reducing activities. *L. plantarum* GM77 can grow at both 15 and 45°C and, on the other hand, in the presence of 6.5 and 10% NaCl, and it does not form ammonia from arginine (23).

S. carnosus MK93 was grown in TSB (Oxoid, Hampshire, UK) at 30°C for 24 hr. *L. plantarum* GM77 was grown in MRS broth (Oxoid) at 30°C for 24 hr. *S. carnosus* MK93 and *L. plantarum* GM77 were added to the *sucuk* mix to attain 10⁶ and 10⁷ CFU/g, respectively.

Production of *sucuk* The formulation consisted of beef meat and beef meat fat (80:20, w/w) and 2.5% NaCl, 1% garlic, 0.4% sucrose, 0.7% red pepper, 0.5% black pepper, 0.9% cumin, 0.25% pimento, and 0.015% NaNO₂ (24).

The *sucuk* batters were prepared in a laboratory-type cutter (Typ MTK 662; MADO, Dornhan/Schwarzwald, Germany). First, the control group batter was prepared. In this group *L. plantarum* GM77 was used as a lactic culture. Then, batter containing *S. carnosus* MK93 and *L. plantarum* GM77 was prepared. The mixture of each batter was stuffed in to collagen casings (38 mm, Naturin Darm, Germany) with a laboratory-type filling machine (Typ MTK 591; MADO), the final mass of each sausage was 200±10 g. The ripening programs were as follows: 3 days 24±1°C and relative humidity (RH) 90±2%, 4 days at

*Corresponding author: Tel: +90-442-231-2425; Fax: +90-442-236-0958
E-mail: gkaban@atauni.edu.tr
Received June 25, 2008; Revised July 23, 2008;
Accepted July 29, 2008

22±1°C and RH 85±2%, 7 days at 18±1°C and RH 80±2%. The experiment was conducted twice.

Sampling procedures Sampling was performed by randomly selecting 2 samples of each sausage group after 0, 1, 3, 7, 9, and 14 days for volatile compounds, microbiological and physicochemical analyses.

Microbiological analysis For microbiological analysis, the sausage casing was removed aseptically. Sausage was cut using a sterile knife. Twenty-five g samples from each sausage were transferred into a sterile stomacher bag and 225 mL of sterile physiological water (0.85% NaCl) were added and homogenized for 1.5 min in a stomacher (Lab Blender 400-BA 7021; Stomacher Seward Medical, London, England). Serial decimal dilutions were prepared in the same diluent and 0.1 mL samples of appropriate dilutions were spread in duplicate on selective agar plates.

Lactic acid bacteria were enumerated on de Man, Rogosa, Sharpe, agar (MRS, Oxoid, Hampshire, UK) in anaerobic conditions (Aneorocult A, Merck, Darmstadt, Germany) after 48 hr at 30°C; *Micrococcus/Staphylococcus* on Mannitol Salt Phenol-Red agar (MSA, Merck) after 48 hr at 30°C. Enterobacteriaceae were determined on Violet Red Bile Dextrose agar (VRBD, Merck), incubation was carried out at 30°C for 48 hr in anaerobic conditions.

Physicochemical analysis The pH was measured in a homogenate of the sample with distilled water (1:10 w/v) using a pH meter (ATI ORION 420; Orion, Beverly, MA, USA). Water activity (A_w) was determined using a TH-500 A_w sprin apparatus (Novasina, Pfäffikon, Switzerland).

Nonprotein nitrogen (NPN) was determined by the Kjeldahl method after protein precipitation with trichloroacetic acid, the results were expressed as g/100 g of sausage (25). Thiobarbituric acid reactive substances (TBARS) values of samples were determined according to the methods of Lemon (26) and were expressed as μmol malonaldehyde/g sample.

Volatile compounds analysis The extraction of headspace volatile compounds was used a solid phase microextraction (SPME) device (Supelco, Bellefonte, PA, USA), using fibres of 75 μm , carboxen/polydimethylsiloxane (CAR/PDMS) and performed according to Kaban (22). The compounds adsorbed by the fibre were desorbed from the injection port of the gas chromatography (GC, Agilent Technologies 6890N; Agilent, Santa Clara, CA, USA) for 6 min at 250°C with the purge valve off (splitless mode). The compounds were separated in a BD-624 (30 m, 0.25 mm i.d., 1.4- μm film, J&W Scientific, Folsom, CA, USA) capillary column. The GC was equipped with a mass selective detector (MS, Agilent Technologies 5973). Helium was used as carrier gas. The GC oven temperature programme was started when the fibre was inserted and held at 40°C for 5 min and subsequently programmed from 40 to 110°C at 3°C/min and at a rate of 4°C/min from 150°C, then at a rate of 10°C/min from 210°C where it was held for another 15 min. The total run time was 56.33 min and the GC-MS interface was maintained at 240°C. Mass spectra were obtained by electron impact at 70 arbitrary scale (eV), and data were acquired across the range 50-

500 u.m.a .

The compounds were determined by comparing the results with mass spectra from a database developed by NIST and WILEY or standards molecules (Supelco 44585-U) and by matching their retention indices with those in the literature. Quantification was based on either a total or single ion chromatogram on an eV. The results are expressed as means of 3 replicates.

Statistical analysis The data were tested by variance analysis (complete randomized design, 2 replicates) and differences between means were evaluated by Duncan's multiple-range test using statistical package for the social sciences (SPSS) 13 Statistics Software (Chicago, IL, USA, 2004).

Results and Discussion

pH The changes in pH value of *sucuk* samples during ripening are presented in Fig. 1a. The pH value has dropped a little bit more rapidly in *sucuk* inoculated with *S. carnosus* compared to the control group. However, the real source of acidification is *L. plantarum*, pH value decreased to below 5 after 3 days of fermentation in all groups (Fig. 1a).

A_w The use of starter culture had no significant effect on A_w value of *sucuk* ($p > 0.05$). This result means that acidification in both groups (*L. plantarum* and *L. plantarum*+*S. carnosus*) is sufficient and, because of this, there are no differences between these 2 groups in terms of drying. On the other hand, ripening period had a significant effect ($p < 0.01$) on A_w value. The value dropped continuously from day 3 to the end of the ripening period (Fig. 1b). The interaction of starter culture \times ripening time had no significant effect ($p > 0.05$) on A_w value.

NPN values Ripening period had a significant effect ($p < 0.01$) on the NPN value, this value has increased with time as the ripening period progressed. However, the mean values for day 0 and day 1 are statistically insignificant ($p > 0.05$). In agreement with the results of this study, other reports have shown that proteolysis during the ripening of fermented sausage is reflected by an increase in NPN value (23,27,28). Sausage inoculated with *L. plantarum* and *S. carnosus* showed generally higher NPN values during ripening period than in sausage inoculated with only *L. plantarum* (Fig. 1c). According to the variance analysis results, addition of *S. carnosus* to *sucuk* batters was important ($p < 0.05$). The higher NPN value in *sucuk* with *S. carnosus* may result from the proteolytic activity of *S. carnosus*. Similarly, Hughes *et al.* (29) reported that NPN values of the samples containing *S. carnosus* are higher than that of the control group.

TBARS values Changes in TBARS values of *sucuk* are shown in Fig. 1d. TBARS values for control group were higher during the ripening period than the group containing *L. plantarum*+*S. carnosus*. Likewise, Talon *et al.* (30) have reported that *S. carnosus* inhibits the oxidation of unsaturated free fatty acids. Ripening time was also affected TBARS value ($p < 0.01$), the highest level was detected at day 14. The interaction between starter culture

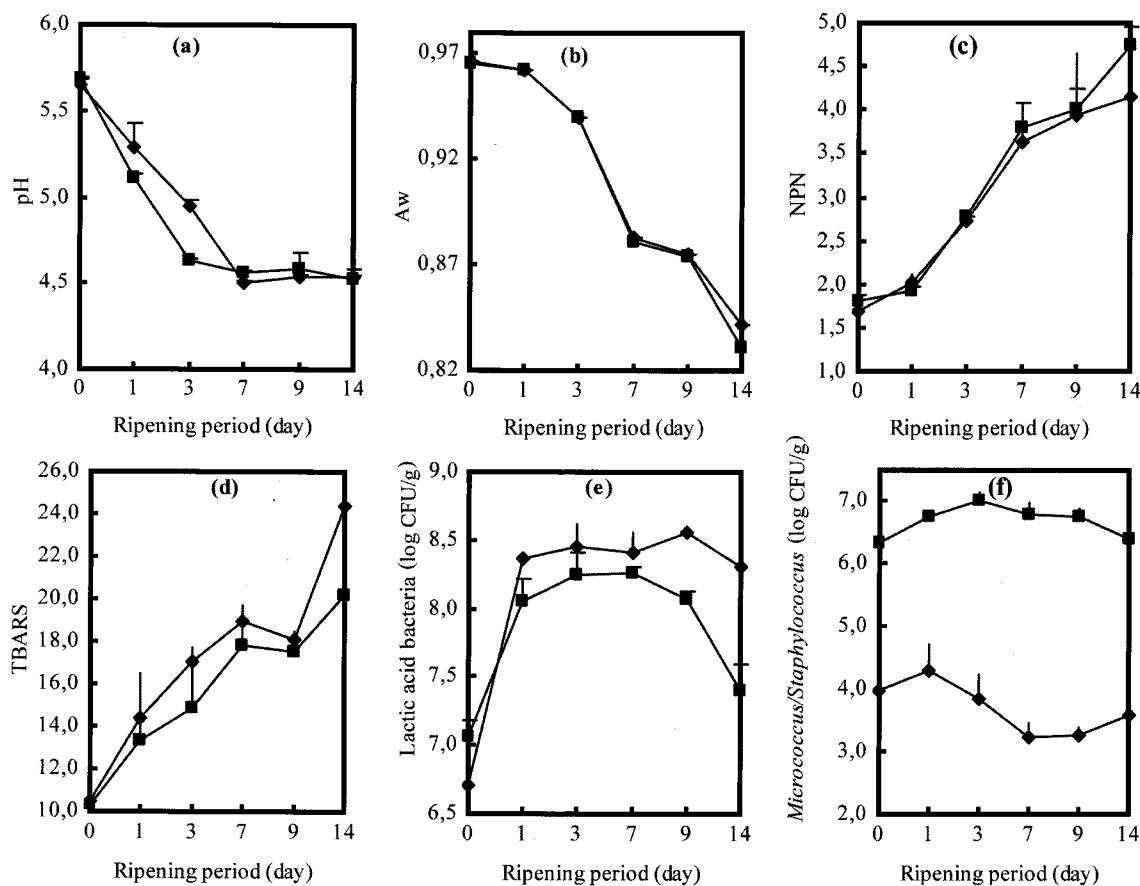


Fig. 1. The changes in pH (a), A_w (b), NPN (c), and TBARS (d) values, lactic acid bacteria (e) and *Micrococcus/Staphylococcus* (f) counts in *sucuk* with and without *S. carnosus* during ripening. -◆-, *L. plantarum*; -■-, *L. plantarum*+*S. carnosus*

and ripening period had no significant effect on the TBARS value ($p>0.05$).

Microbiological evaluation As it can be seen from Fig. 1e, *L. plantarum*, which was used as a lactic culture in both groups, has adapted well to the ripening conditions. But, *L. plantarum* alone demonstrated a good growth in the control group. The use of *S. carnosus* had significant effect ($p<0.01$) on *Micrococcus/Staphylococcus* counts. *S. carnosus*, which was added to *sucuk* batter at 10^6 CFU/g level, increased approximately 1 log unit within the first 3 days of the fermentation. There are no significant differences in the number *S. carnosus* in the following days of the ripening (Fig.1f). This result shows that this strain has adapted well in *sucuk*. In contrast, indigenous *Micrococcus/Staphylococcus* had not grown in the control group; the counts were under initial level during the ripening period, except day 1. This result is similar to the findings of Lücke and Hechelmann (2), Kaban and Kaya (31) who considered fast acidification as the main cause of micrococccaeae inhibition in dry fermented sausage. Enterobacteriaceae were progressively eliminated from the ripened *sucuk*, the initial enterobacteriaceae counts were 10^3 CFU/g level, and it dropped to undetectable levels at the 3rd day in both groups. This was caused by the rapid drop of pH.

Volatile compounds Forty compounds were selected from the volatile compounds identified from *sucuk* during

ripening, and mean peak areas and variance analysis results for these 40 compounds are shown in Table 1. There were differences between 2 groups in terms of the following compounds: β -pinene ($p<0.05$), 4-terpinenol ($p<0.01$), benzene-1,3-bis (1,1-dimethylethyl) ($p<0.001$), and hexadecane ($p<0.001$). However, the mean peak areas of these 4 compounds are quite low. The real effect was demonstrated by the ripening period (Table 1). Acetic acid was shown as the major peak at day 7 in both in control group (*L. plantarum*) and in sausage inoculated with *L. plantarum*+*S. carnosus*. 1-Propene,3,3-thiobis was demonstrated high levels in the beginning days and dropped in the following days. β -Pinene was shown higher levels at day 7. D-Limonene, β -myrcene, α -phellandrene, and α -terpinen were demonstrated increases at the last 2 days. *O*-Cymene, propanal-2-methyl-3-phenyl, γ -terpinene, linalol, and disulphide-di-2-propenyl which were major among all compounds, showed significant changes during the ripening (Table 1). The significance of these compounds for *sucuk* is also reported by Kaban (22). One of the important compounds, linalol, was also significantly affected ($p<0.05$) by the interaction between starter culture and ripening period. Other researchers have reported that different *Staphylococcus* species produce different aroma compounds in different levels (15,18,32). The different results of our study may be explained by large and rapid acidification, short ripening period, and other processing parameters.

Table 1. The effects of starter culture and ripening period on the volatile compounds from *sucuk* samples

Identified compounds	KI ²⁾	ID ³⁾	Starter culture (SC)														p-value ¹⁾		
			<i>L. plantarum</i>							<i>L. plantarum</i> + <i>S. carnosus</i>									
			Ripening period (RP, day)							Ripening period (RP, day)									
Hexane	600	A	0.21 ⁴⁾ 0.16 ⁵⁾	1.51 2.14	-	-	-	-	-	-	0.08 0.01	1.81 2.56	-	-	-	-	-	-	-
1,3-Epithio-propene	612	B	2.32 0.76	2.18 1.34	9.57 0.91	12.39 0.26	5.22 0.93	11.56 2.43	2.34 0.47	1.42 2.01	7.63 2.01	12.67 3.85	6.14 0.76	12.34 1.97	-	-	-	<0.001	
Acetic acid	717	A	-	-	0.14 0.20	3.47 0.60	2.12 0.71	1.24 0.69	-	-	0.33 0.11	4.04 0.13	1.55 0.08	2.24 0.16	-	-	-	<0.001	
Allyl methyl sulfide	731	B	1.67 0.33	1.18 0.22	1.25 0.14	1.13 0.01	1.68 0.07	0.99 0.63	1.22 0.11	1.64 0.48	0.96 0.01	0.75 0.51	2.14 0.06	1.49 0.81	-	-	-	<0.05	
Toluene	791	A	1.35 0.41	1.67 0.86	1.11 0.21	1.27 0.23	3.27 0.82	1.35 0.12	0.88 0.08	8.87 9.37	0.72 0.07	1.42 0.08	3.16 0.37	1.58 0.03	-	-	-	-	
Hexanal	849	B	-	-	-	0.01 0.01	0.31 0.09	0.08 0.07	-	-	-	-	0.36 0.10	0.23 0.06	-	-	-	<0.001	
3,3-Thiobis, 1-propene	888	B	4.45 1.29	3.02 0.30	1.64 0.18	0.74 0.03	1.76 0.25	1.35 0.20	3.36 0.18	4.17 0.62	1.07 0.05	0.52 0.02	1.91 0.24	1.67 0.09	-	-	-	<0.001	
1 R- α -pinene	951	B	0.35 0.08	0.48 0.12	0.59 0.01	0.90 0.02	0.74 0.00	0.97 0.08	0.42 0.01	0.36 0.06	0.59 0.04	0.98 0.03	0.76 0.06	0.90 0.04	-	-	-	<0.001	
2-Metil-propenyl, disulfide	955	B	0.48 0.68	2.76 1.00	0.80 0.10	0.24 0.05	0.67 0.30	0.36 0.21	-	2.57 0.25	0.29 0.02	0.22 0.02	0.23 0.05	0.43 0.04	-	-	-	<0.001	
β -Pinene	988	B	1.50 0.34	1.94 0.40	2.26 0.20	3.65 0.14	2.20 0.28	2.66 0.16	1.51 0.03	1.67 0.23	2.76 0.15	4.41 0.26	2.48 0.16	2.77 0.06	<0.05	<0.001	-	<0.001	
β -Myrcene	1,005	B	0.30 0.42	0.40 0.56	1.25 0.70	0.78 0.46	3.05 0.10	3.08 0.21	0.71 0.35	1.15 0.21	0.81 0.11	0.81 0.12	2.37 0.13	2.88 0.78	-	-	-	<0.001	
α -Phellandrene	1,019	B	0.67 0.26	0.60 0.13	0.85 0.11	0.90 0.02	1.38 0.02	1.19 0.09	0.42 0.01	0.70 0.18	0.75 0.21	0.87 0.06	1.71 0.07	1.23 0.03	-	-	-	<0.001	
3-Carene	1,022	B	0.56 0.12	1.18 1.45	0.65 0.05	0.67 0.06	0.91 0.47	1.12 0.20	0.60 0.03	0.32 0.08	0.48 0.04	0.67 0.01	1.64 0.17	1.20 0.13	-	-	-	-	
α -Terpinene	1,030	B	0.24 0.08	0.20 0.14	0.28 0.05	0.07 0.01	0.49 0.05	0.38 0.03	0.14 0.03	0.25 0.06	0.07 0.02	0.05 0.01	0.57 0.15	0.40 0.01	-	-	-	<0.001	
D-Limonene	1,054	B	2.05 0.49	1.63 0.65	2.25 0.21	1.44 0.11	3.65 0.50	3.24 0.34	1.54 0.04	1.71 0.42	1.31 0.16	1.49 0.12	4.39 0.06	3.49 0.35	-	-	-	<0.001	
O-Cymene	1,058	B	20.93 7.41	18.63 9.80	17.29 2.63	11.17 0.96	27.89 0.59	20.73 1.08	13.14 0.01	18.75 5.85	11.85 1.32	13.10 1.29	36.80 0.21	24.71 0.71	-	-	-	<0.001	

Table 1. Continued

	Starter culture (SC)												p-value	
	<i>L. plantarum</i>						<i>L. plantarum</i> + <i>S. carnosus</i>							
	Ripening period (RP, day)						Ripening period (RP, day)							
Hexanoic acid	1,059	A	-	-	0.03	0.06	0.06	0.01	-	-	0.03	0.07	-	<0.001
γ -Terpinene	1,079	B	6.20	5.10	7.84	6.94	9.73	9.39	4.51	6.49	6.27	8.05	9.87	10.18
			1.97	0.19	1.17	0.38	0.49	0.58	0.01	1.39	0.72	0.94	0.42	0.24
Di-2-propenyl -disulfide	1,138	C	7.74	6.22	4.59	2.64	2.10	3.07	7.90	6.51	3.90	1.46	0.70	2.62
			0.04	3.34	0.05	0.26	1.08	0.22	0.08	0.06	0.04	0.11	1.00	0.07
$C_6H_{10}S_2$	1,154	C	1.23	1.57	1.48	0.98	0.59	0.59	1.62	1.43	2.19	0.96	0.25	0.55
			0.93	1.52	0.81	0.18	0.05	0.16	0.16	0.35	0.11	0.30	0.05	0.22
Linalol	1,161	B	6.15	4.29	7.13	4.82	5.40	5.36	6.28	6.40	5.60	5.53	4.55	5.41
			0.88	1.47	0.75	0.51	0.41	0.11	0.35	0.30	0.48	0.71	0.17	0.13
Methyl-2-propenyl trisulfide	1,185	C	0.06	0.24	0.16	-	-	-	0.09	0.28	0.12	-	-	-
			0.08	0.25	0.01	-	-	-	0.05	0.01	0.04	-	-	-
Camphor	1,233	B	0.18	0.21	0.19	0.21	0.12	0.14	0.17	0.21	0.18	0.13	0.15	0.13
			0.04	0.07	0.00	0.01	0.00	0.00	0.00	0.03	0.01	0.01	0.01	0.01
4-Terpinenol	1,240	B	0.18	0.15	0.26	0.19	0.17	0.20	0.19	0.22	0.17	0.09	0.16	0.14
			0.01	0.02	0.02	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.00	0.06
3-Cyclohexene-1-methanol, α , α ,4-trimethyl	1,256	C	-	0.10	0.27	0.27	0.23	0.25	-	0.11	0.20	0.07	0.24	0.12
				0.08	0.11	0.09	0.00	0.08	-	0.16	0.04	0.01	0.02	0.10
Benzene,1,3-bis(1,1-dimethylethyl)	1,278	B	-	-	0.18	0.17	0.14	0.06	-	-	0.07	0.07	0.16	0.02
					0.01	0.04	0.02	0.01	-	-	0.02	0.00	0.01	0.02
Propanal,2-methyl-3-phenyl	1,334	B	21.73	14.36	13.24	11.32	5.24	5.19	31.97	17.87	20.73	12.27	2.64	5.62
			9.41	16.28	0.92	1.08	0.88	0.33	0.41	0.71	2.16	2.04	0.26	0.69
Benzene,1-methoxy-4-(1-propenyl)	1,342	B	0.11	0.09	0.11	0.39	0.09	0.12	0.18	0.08	0.23	0.34	0.05	0.08
			0.07	0.12	0.01	0.03	0.01	0.04	0.04	0.03	0.06	0.04	0.02	0.01
Phellandral	1,358	C	0.04	0.03	0.02	0.02	-	-	0.09	0.06	0.05	-	-	-
			0.05	0.04	0.00	0.02	0.02	0.03	0.03	0.02	0.03	-	-	-
Benzenemethanol,4-(1-methylethyl)	1,382	B	0.04	0.12	1.05	5.19	1.33	2.48	0.07	0.29	4.07	5.18	0.50	1.94
			0.06	0.17	0.70	0.07	0.50	0.36	0.05	0.06	0.04	0.95	0.06	0.37
Copaene	1,435	C	-	-	0.01	0.06	0.04	0.07	-	0.01	0.02	0.01	0.04	0.05
					0.01	0.00	0.01	0.01	-	0.01	0.03	0.01	0.02	0.02
Beta elemene	1,453	B	0.05	0.03	0.07	0.15	0.08	0.14	0.05	0.05	0.13	0.12	0.05	0.08
			0.04	0.01	0.01	0.05	0.01	0.50	0.01	0.01	0.02	0.01	0.01	0.01

Table 1. Continued

	Starter culture (SC)															
	<i>L. plantarum</i>							<i>L. plantarum</i> + <i>S. carnosus</i>								
	Ripening period (RP)							Ripening period (RP, day)							<i>p</i> -value ¹⁾	
Eugenol	1,460	B	0.17	0.28	0.37	1.41	0.22	0.46	0.37	0.17	1.03	1.66	0.09	0.30	-	<0.001
			0.14	0.37	0.08	0.22	0.06	0.07	0.09	0.05	0.23	0.33	0.01	0.06	-	
Trans caryophyllene	1,473	B	0.36	0.29	0.34	0.24	0.31	0.34	0.39	0.38	0.47	0.21	0.32	0.29	-	<0.05
			0.05	0.14	0.04	0.04	0.08	0.08	0.00	0.01	0.01	0.01	0.04	0.00	-	
Benzene, 1,2-dimethoxy-4-(2-propenyl)	1,466	B	0.77	1.05	1.28	5.03	0.76	1.55	1.41	0.58	3.54	5.91	0.28	1.15	-	<0.001
			0.49	1.27	0.27	0.86	0.25	0.35	0.16	0.18	0.54	1.29	0.01	0.18	-	
5-Methyl-1,2,3,4-tetrahydro-cyclohexane	1,474	C	0.27	0.11	0.12	-	-	-	0.17	0.25	0.05	-	-	-	-	<0.001
			0.04	0.05	0.02	-	-	-	0.08	0.01	0.01	-	-	-	-	<0.001
Caryophyllene	1,490	B	0.93	1.09	2.16	3.80	1.69	2.47	1.08	1.00	2.92	3.82	1.17	2.26	-	<0.001
			0.23	0.60	0.02	0.51	0.41	0.21	0.01	0.11	0.04	0.24	0.08	0.13	-	
Alpha caryophyllene	1,504	B	0.08	0.09	0.14	0.28	0.17	0.25	0.09	0.07	0.22	0.23	0.10	0.17	-	<0.001
			0.04	0.04	0.01	0.01	0.01	0.04	0.01	0.01	0.01	0.05	0.01	0.01	-	<0.001
Benzene, 1,2-dimethoxy-4-(1-propenyl)	1,558	B	0.09	0.10	0.17	0.42	0.28	0.42	0.18	0.14	0.42	0.15	0.04	0.08	-	-
			0.07	0.14	0.01	0.04	0.04	0.42	0.08	0.07	0.11	0.06	0.02	0.04	-	-
Hexadecane	1,600	B	-	-	-	-	0.66	1.70	-	-	-	-	0.64	0.59	<0.001	<0.001
			-	-	-	-	0.18	0.28	-	-	-	-	0.06	0.01	<0.001	<0.001

¹⁾Significant levels from analysis of variance. Only significant values listed ($p < 0.05$).

²⁾Kovats index calculated for DB-624 capillary column (J& W Scientific: 30 m, 0.25 mm i.d., 1.4- μ m film thickness) installed on a gas chromatograph equipped with a mass-selective detector.

³⁾Reliability of identification: A, mass spectrum and retention time identical with an authentic sample; B, mass spectrum and Kovats index from literature in accordance; C, tentative identification by mass spectrum.

⁴⁾Mean peak area.

⁵⁾Standard deviation.

In conclusion, *S. carnosus*, isolated from traditional dry fermented sausage, demonstrated a good growth during the fermentation, and remained in high levels during drying. NPN value increased in the presence of this strain due to its proteolytic activity. In the presence of *S. carnosus*, lipid oxidation is limited as well. However, *S. carnosus* did not significantly change the profiles of the volatile compounds as expected.

References

- Lücke KF. Utilization of microbes to process and preserve meat. *Meat Sci.* 56: 105-115 (2000)
- Lücke FK, Hechelmann H. Starter cultures for dry sausages and raw ham composition and effect. *Fleischwirtschaft* 67: 307-314 (1985)
- Martin A, Colin B, Aranda E, Benito MJ, Cordoba MG. Characterization of Micrococcaceae isolated from Iberian dry-cured sausages. *Meat Sci.* 75: 696-708 (2007)
- Park WM, Choi WH, Yoo IJ, Kim YS, Kim WJ, Chung DH. Effects of lactic acid bacteria isolated from fermented foods on the physicochemical properties of fermented sausages. *Food Sci. Biotechnol.* 6: 104-108 (1997)
- Sanz Y, Vial R, Toldra F, Nieto P, Flores J. Effect of nitrate and nitrite curing salts on microbial changes and sensory quality of rapid ripened sausages. *Int. J. Food Microbiol.* 37: 225-229 (1997)
- Comi G, Citterio B, Manzano M, Cantoni C, Bertoldi M. Evaluation and characterization of Micrococcaceae strains in Italian dry fermented sausages. *Fleischwirtschaft* 72: 1679-1683 (1992)
- Coppola R, Lorizzo M, Saotta R, Sorrentino E, Grazia L. Characterization of micrococci and staphylococci isolated from Soppressata Molisana, A southern Italy fermented sausage. *Food Microbiol.* 14: 47-53 (1997)
- Cocolin L, Manzano M, Aggio D, Cantoni C, Comi G. A novel polymerase chain reaction (PCR)-denaturing gradient gel electrophoresis (DGGE) for the identification of Micrococcaceae strains involved in meat fermentations. Its application to naturally fermented Italian sausages. *Meat Sci.* 57: 59-64 (2001)
- Blaiotta G, Pennacchia C, Villani F, Ricciardi A, Tofalo R, Parente E. Diversity and dynamics of communities of coagulase-negative staphylococci in traditional fermented sausages. *J. Appl. Microbiol.* 97: 271-284 (2004)
- Miralles MC, Flores J, Perez-Martinez G. Biochemical tests for the selection of *Staphylococcus* strains as potential meat starter cultures. *Food Microbiol.* 13: 227-236 (1996)
- Garcia-Varona M, Santos EM, Jaime I, Rovira J. Characterisation of Micrococcaceae isolated from different varieties of Chorizo. *Int. J. Food Microbiol.* 54: 189-195 (2000)
- Martin B, Garriga M, Hugas M, Bover-Cid S, Veciana-Nogues MT, Aymerich T. Molecular, technological, and safety characterization of Gram-positive catalase-positive cocci from slightly fermented sausages. *Int. J. Food Microbiol.* 107: 148-158 (2006)
- Papamanoli E, Kotzekidou P, Tzanetakis N, Litopoulou-Tzanetaki E. Characterization of Micrococcaceae isolated from dry fermented sausage. *Food Microbiol.* 19: 441-449 (2002)
- Kaban G, Kaya M. Identification of lactic acid bacteria and Gram-positive catalase-positive cocci isolated from naturally fermented sausage (*sucuk*). *J. Food Sci.* 73: M385-M388 (2008)
- Berdagué JL, Montel P, Montel MC, Talon R. Effects of starter cultures on the formation of flavour compounds in dry sausage. *Meat Sci.* 35: 275-287 (1993)
- Stahnke LH. Aroma components from dried sausages fermented with *Staphylococcus xylosum*. *Meat Sci.* 38: 39-53 (1994)
- Stahnke LH. Dried sausages fermented with *Staphylococcus xylosum* at different temperatures and with different ingredient levels-Part II. Volatile components. *Meat Sci.* 41: 193-209 (1995)
- Stahnke LH. Volatiles produced by *Staphylococcus xylosum* and *Staphylococcus carnosus* during growth in sausage minces. Part I. Collection and identification. *Lebensm.-Wiss. Technol.* 32: 357-364 (1999)
- Stahnke LH. Volatiles produced by *Staphylococcus xylosum* and *Staphylococcus carnosus* during growth in sausage minces. Part II. The influence of growth parameters. *Lebensm.-Wiss. Technol.* 32: 365-371 (1999)
- Johansson G, Berdagué JL, Larsson M, Tran N, Borch E. Lipolysis, proteolysis, and formation of volatile components during ripening of a fermented sausage with *Pediococcus pentosaceus* and *Staphylococcus xylosum* as starter cultures. *Meat Sci.* 38: 203-218 (1994)
- Sondergaard AK, Stahnke LH. Growth and aroma production by *Staphylococcus xylosum*, *S. carnosus*, and *S. equorum*- A comparative study in model system. *Int. J. Food Microbiol.* 75: 99-109 (2002)
- Olesen PT, Meyer AS, Stahnke LH. Generation of flavour compounds in fermented sausage-the influence of curing ingredients, *Staphylococcus* starter culture and ripening time. *Meat Sci.* 66: 675-687 (2004)
- Kaban G. Isolation and identification of lactic acid bacteria and catalase-positive cocci in traditionally-produced *sucuk*, their possible uses in production, and their effects on volatile compounds. PhD thesis, Atatürk University, Erzurum, Turkey (2007)
- Kaya M, Gökalp HY. The behavior of *Listeria monocytogenes* in *sucuks* produced with different lactic starter cultures. *Turk. J. Vet. Anim. Sci.* 28: 1113-1120 (2004)
- Anonymous. Official Collection of Test Procedures Pursuant to § 35 LMBG. Investigation of food. Determination of amount of NPN-substance in meat products. L07.00-41, Germany (1989)
- Lemon DW. An Improved TBA Test for Rancidity. New Series Circular. No 51. Halifax-Laboratory, Halifax, Nova Scotia, Canada (1975)
- DeMasi TW, Wardlaw FB, Dick RL, Acton JC. Nonprotein nitrogen (NPN) and free amino acid contents of dry fermented and nonfermented sausages. *Meat Sci.* 27: 1-12 (1990)
- Gençcelep H, Kaban G, Kaya M. Effects of starter cultures and nitrite levels on formation of biogenic amines in *sucuk*. *Meat Sci.* 77: 424-430 (2007)
- Hughes MC, Kerry JP, Arendt EK, Kenneally PM, McSweeney PLH, O'Neill EE. Characterization of proteolysis during the ripening of semi-dry fermented sausages. *Meat Sci.* 62: 205-216 (2002)
- Talon R, Walter D, Montel MC. Growth and effect of staphylococci and lactic acid bacteria on unsaturated free fatty acids. *Meat Sci.* 54: 41-47 (2000)
- Kaban G, Kaya M. Effect of starter culture on growth of *Staphylococcus aureus* in *sucuk*. *Food Control* 17: 797-801 (2006)
- Montel MC, Talon JR, Berdagué JL, Rousset-Akrim S. Biochemical activities of Micrococcaceae and their effects on the aromatic profiles and odours of a dry sausage model. *Food Microbiol.* 13: 489-499 (1996)