PAPER

EFFECT OF MIXED STARTER CULTURES ON BIOGENIC AMINE FORMATION DURING THE RIPENING OF TUNISIAN DRY FERMENTED CAMEL MEAT SAUSAGE

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ABSTRACT

The effect of mixed starter cultures on biogenic amine production was examined during the ripening process of dry camel meat sausage. Changes in pH, moisture content, proteolysis, microbial counts and lipid oxidation were also studied. The combination of three amine-negative bacteria, resulted in a drastic reduction of biogenic amine production. The highest total free amino acid concentration was observed in batches manufactured with mixed starter cultures. The bactericidal properties of *L. sakei* improved the hygienic quality of sausages by decreasing the number of Enterobacteriaceae. Inoculation of sausages with a mixture of strains, significantly delayed lipid oxidation and enhanced sensory characteristics.

Keywords: biogenic amine, fermented sausages, starter cultures, quality, ripening

1. INTRODUCTION

The formation of biogenic amines (BAs) in fermented sausages could affect the quality of the final products. BAs are mainly formed by the decarboxylation of amino acids. Biogenic amines could be harmful on health when they are consumed in large quantities; these molecules are responsible for food poisoning (LATORRE-MORATALLA *et al*., 2010; LORENZO *et al*., 2017). In fact, "tyramine and phenylethylamine have been associated with food histaminic intoxications and severe hypertensive crisis (MARINE-FONT *et al*., 1995)". Moreover, BOVER-CID *et al*. (1999) have reported that putrescine and cadaverine could contribute to the formation of heterocyclic carcinogenic nitrosamines.
Many factors contribute to the formation of BAs in dry fermented sausages such as ingredients, technological ripening conditions, acidification, and proteolysis during the ripening of dry fermented sausage. Many studies have reported that the production of biogenic amines in fermented meat products was related to the growth of Pseudomonas, enterobacteria, enterococci, and lactobacilli (DURLU-ÖZKAYA *et al*., 2001; SUZZI *et al*., 2003; LATORRE-MORATALLA *et al*., 2012).
Strains of lactic acid bacteria (LAB) and coagulase negative staphylococci (CNS) are usually used in the manufacture of dry fermented sausages. These strains should have the most important technological properties such as the adaptation to meat environment, development of red color, texture, and flavor of fermented meats (TALON *et al*., 2011). Moreover, the use of amine-negative strains with bacteriostatic activity can stopped the formation of BAs in the initial fermentation stage (BOVER-CID *et al*., 2000).
In the present study, the influence of mixed starter cultures of *L. sakei*, *S. carnosus* and *S. xylosus* on biogenic amine formation during the ripening of dry fermented camel meat sausage was studied. Besides the biogenic amine contents, evolution of microbial population, amino acids content, total lipid and total protein contents, pH, thiobarbituric acid (TBA), moisture content, textural and sensory characteristics were determined during the ripening of a dry fermented camel meat sausage.

2. MATERIALS AND METHODS

2.1. Sausage preparation

"The sausage formulation included 3.750 kg of camel meat (75%), 1.250 kg of hump fat (25%), 260 g of salt, 15 g of black pepper, 15 g of paprika, 60 g of glucose and 0.8 g of potassium nitrate. After chopping and mixing the ingredients, the mixture was divided into two batches (2.5 kg for each batch): batch 1, inoculated with a commercial starter culture starter A (20 g/200 kg): *L. sakei* + *S. carnosus* + *S. xylosus* (TEXEL SA-201, DANISCO, Paris, France) and batch 2, control without inoculation. Starter A was added to sausages according to manufacturer's recommendations. The mixture of each batch was stuffed into artificial casings, giving approximately 500 g as the final mass of each sausage and then placed in a fermentation chamber (BCR, CF 1 B, Antony, France). The sausages were fermented for 5 days at 24 °C and 80% relative humidity (RH). After 5 days of processing, the temperature was decreased to 14 °C for 23 days and the RH value was 80%. For sampling, three sausages of each batch at 0 day (mix before stuffing) and after 7, 14, 21 and 28 days of ripening were taken for microbiological, physicochemical and textural analysis. All reported values represent the mean of three random measurements of the sausage sample."

2.2. Microbiological analysis

"Sausage samples (10 g) of each batch were homogenized with 90 mL of sterile peptone water (Biolife, Milan, Italy) and decimal dilutions were prepared. Mesophilic LAB were enumerated on MRS (de Man, Rogosa and Sharpe) agar (Biolife) after 48 h of incubation at 30 °C. The number of staphylococci was determined on mannitol salt agar (Biolife) after incubation at 37 °C for 48 h. Yeasts and molds were enumerated on Sabouraud Dextrose Agar (Biokar, Beauvais, France) at 28 °C for 4 days. Total viable counts were determined on standard plate count agar (Biolife) at 30 °C for 48 h. Enterobacteriaceae were determined on Violet Red Bile Glucose (VRBG) (Biokar) at 37 °C for 24 h."

2.3. pH, moisture, weight loss, total lipid and total protein contents

"The pH values were measured in homogenates prepared by blending 10 g of sausage (Moulinex DPA141, Lyon, France) with 50 mL of distilled water for 2 min. Measurements were taken with a pH meter (microprocessor pH meter BT-500, Boeco, Hamburg, Germany). The moisture content was calculated by weight loss experimented by the sample (5 g) maintained in an oven (Memmert, UL 60, Schwabach, Germany) at 105 °C, until constant weight according to the ISO recommended method (ISO, 1973). Weight loss was expressed as the percentage of the initial weight (LIAROS *et al*., 2009)."
"Total lipids were extracted from 5 g of minced sausage according to the method of FOLCH *et al*. (1957). Total nitrogen was determined according to the Kjeldahl method and total protein estimated by multiplying the nitrogen content by 6.25."

2.4. Lipid oxidation analysis

"Lipid oxidation was assessed by measuring thiobarbituric acid reactive substances (TBARS) during the ripening period. This analysis was performed according to the method of GENOT (1996)" as described by EL ADAB *et al*. (2016).

2.5. Free amino acid (FAA) content

"Free amino acids were extracted and analyzed by reverse phase high-performance liquid chromatography (HPLC) (Agilent L 100 system, Province, Canada) equipped with a Hypersil ODS C18 column (250 mm × 4.6 mm dimensions of the column, 5 μm porosity)", as described by EL ADAB *et al*. (2016).

2.6. Biogenic amines analysis

Biogenic amines were determined using the method described by BOULARES *et al*. (2017). "Biogenic amines are first extracted from the test sample by chloridric acid (0.1 M) and then practice derivatization. Briefly, 4 mL of HCl (0.1 M) were added to 1 g of sausage. After homogenization, samples were centrifuged at 12.000 rpm for 20 min (Sigma, 6-16S, Munich, Germany) before being filtered. After that, 2 mL of the obtained aqueous fraction were homogenized with 1 mL of sodium bicarbonate and 2 mL of dansyl chloride, and then the mixture was heated during 1 h at 40 °C. After addition of 2 mL of diethyl ether, the organic fraction was collected and evaporated under nitrogen liquid stream. The mixture was then dissolved in 1 mL of acetonitrile. A standard solution of amines was prepared similarly and used as control. Finally, 20 μL of each derivatized solution were

injected onto HPLC column where the components will be retained unequally depending on their size and composition. A Knauer eurosphère 100-RP18 reversed-phase column (250 x 4.6 mm, 5 μm, Berlin, Germany) was used for chromatographic separation. Therefore, the detection was performed at 254 nm wavelength using Acetonitrile/water as mobile phase at a constant flow rate of 0.8 mL/min for 20 min."

2.7. Texture Profile Analysis (TPA)

"Texture profile analysis (TPA) of the samples was performed with a texture analyzer (TA-XT2 Stable Micro Systems, Haslemere, UK) equipped with a cylindrical probe of 50 mm in diameter. The sausages were cut in a cylinder 1 cm thick and 3 cm in diameter and compressed twice to 50% of their original thickness. Force-time curves were recorded at a crosshead speed of 1 mm/s. Texture profile parameters (Hardness, cohesiveness, springiness, gumminess and chewiness) were evaluated during the ripening of dry fermented sausages using the method of BOURNE (1978)."

2.8. Color measurement

"Color measurements were carried out using a CR-300 colorimeter (Minolta Chroma Meter CR-300, Tokyo, Japan). Each sausage was cut and the color of the slices was measured three times for each analytical point L*, a* and b* scale coordinates were obtained: L* (lightness), a* (redness) and b* (yellowness). Before each series of measurements, the instrument was calibrated using a white ceramic tile."

2.9. Sensory evaluation

"The sensory analysis was performed by a sensory panel of ten assessors who had undergone professional training. A slice of each sample batch (5 mm thick approximately) was served to the assessors. The sensory evaluation was based on a six point hedonic scale to determine red color (10 = red and shiny; 1 = dark and dull), after taste (10 = extremely desirable; 1 = extremely undesirable), fat intensity (10 = high; 1 = low), acidity (10 = strong acidity; 1 = light acidity), hardness (10 = firm; 1 = soft) and overall acceptability (10 = high; 1 = low)."

2.10. Statistical analysis

"Data were statistically analyzed using one-way analysis of variance (ANOVA) procedure of SPSS 17.0 (SPSS, Inc., Chicago, IL). Duncan's multiple range test was used to determine any significant difference between mean values and evaluations were based on a significance level of $p<0.05$."

3. RESULTS AND DISCUSSION

3.1. Microbiological analysis

The evolutions of microbial population during the ripening of control and inoculated sausages are shown in Fig. 1. The total viable counts (TVC) and LAB counts increased ($p<0.05$) during the ripening period. The numbers of LAB and TVC in starter-mediated

sausages were significantly higher than those in control ones ($p<0.05$), which could be explained by the prior inoculation of sausages by *Lactobacillus sakei*, *Staphylococcus carnosus* and *Staphylococcus xylosus*. Results showed that the number of lactic acid bacteria in dry fermented sausages increased ($p<0.05$) during the two first weeks of ripening. A decrease of their number ($p<0.05$) was observed during the two last weeks of ripening which could be due to the exhaustion of the sugar. These results were similar to those reported by QINXIU *et al*. (2016) and MEJRI *et al*. (2017).
Enterobacteriaceae counts decreased ($p<0.05$) during ripening in both control and starter-mediated sausages. This result was similar to that reported by LU *et al*. (2015). The number of Enterobacteriaceae is lower ($p>0.05$) in inoculated sausages than those measured on control ones. Enterobacteriaceae numbers dropped below 1 logarithmic unit in starter-mediated batches. This drop is due to bactericidal properties of starters (LORENZO *et al*., 2007; CIUCIU *et al*., 2014). Staphylococci profiles showed no significant differences ($p>0.05$). At day 0, staphylococci counts in starter-mediated sausages were more than five logarithmic units higher than those of the control samples. A decrease of the number of staphylococci in sausages was observed after seven days of ripening. Our results match with those found by ZHAO *et al*. (2011) in dry fermented mutton sausages. Yeast and molds counts increased ($p>0.05$) during the first seven days of ripening and then decreased ($p>0.05$) to reach at day 28 values of 4.15 and 3.77 log CFU/g, respectively, for control and inoculated batches.

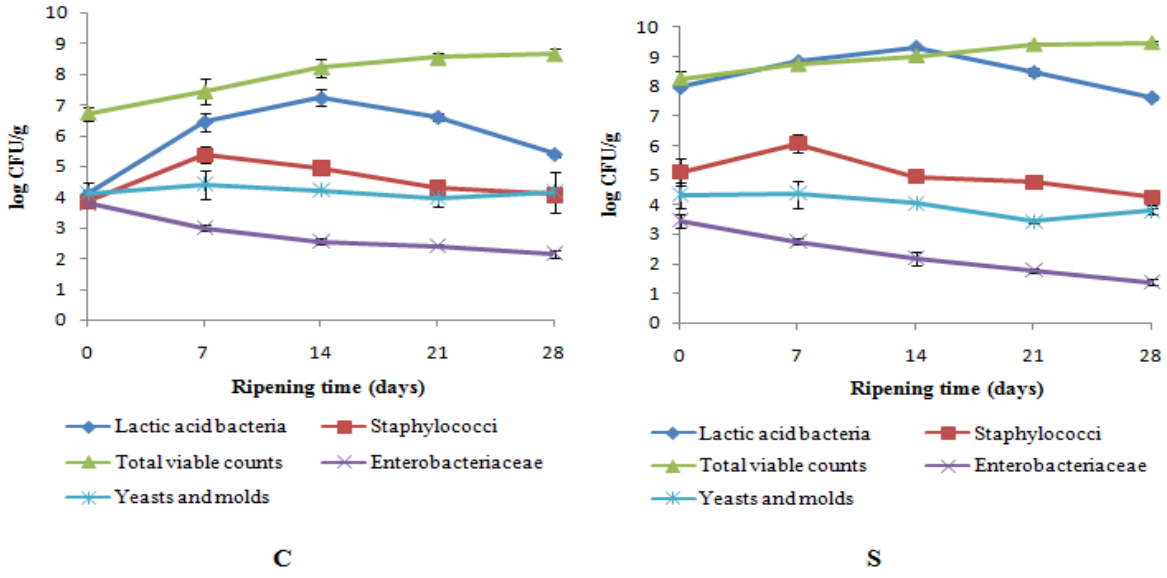

Figure 1. Evolution of microbial population during the ripening of control and inoculated sausages: C (control sausage), S (sausage inoculated with mixed starter cultures).

3.2. pH, moisture, weight loss, total lipid and total protein contents

The pH of control and starter-mediated sausages decreased ($p<0.05$) from 6.22 to 5.79 and 5.01 after 14 days of ripening, respectively (Fig. 2A). The pH decrease could be attributed to lactic acid production by LAB (NIE *et al*., 2014). Beyond the 14th day, pH values increased for both control and inoculated sausages. This may be caused by proteolytic

processes and mold growth on the sausage surface (ESSID and HASSOUNA, 2013). The weight of control sausages and those inoculated with mixed starter cultures decreased ($p<0.05$) during the ripening period (Fig. 2B). These results match with those found by JIN *et al*. (2010) and LIAROS *et al*. (2009). The moisture content decreased ($p<0.05$) in all the samples (Fig. 2C). However, no significant difference ($p>0.05$) was found between the different batches during the ripening process. This water loss is due to the elevated temperature of fermentation (24°C) and to the decrease of pH of sausages to their isoelectric pH (HAMOEN *et al*., 2013).

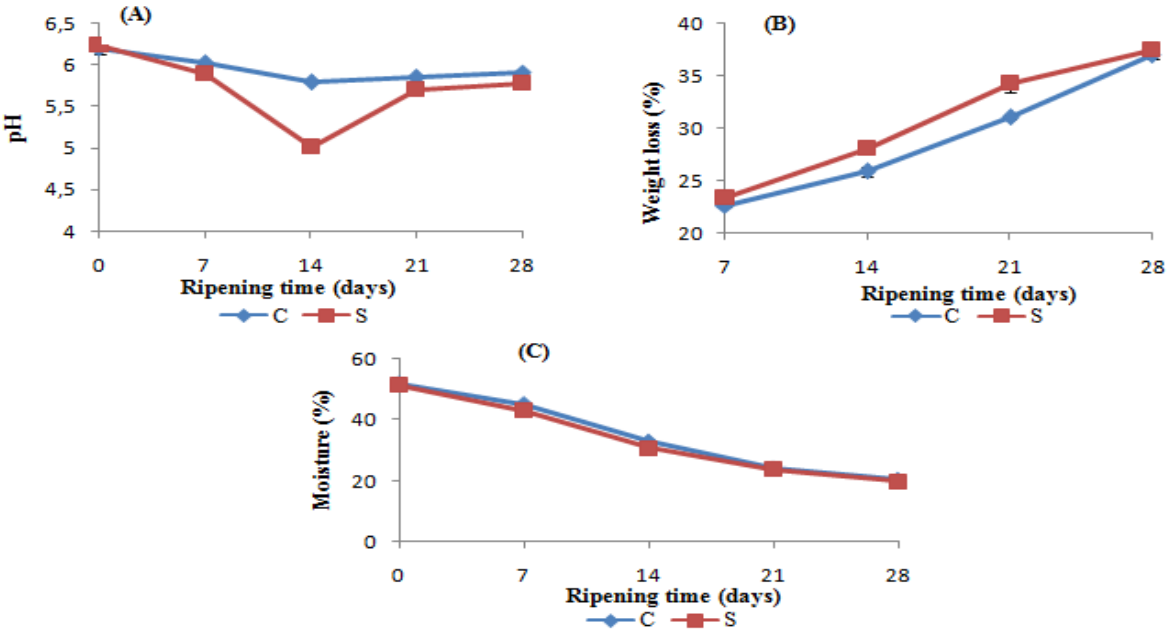

Figure 2. Evolution of pH (A), weight loss (B) and moisture (C) during the ripening of control and inoculated sausages: C (control sausage), S (sausage inoculated with mixed starter cultures).

Changes in total lipid and total protein contents during ripening of dry fermented sausages are summarized in Table 1.

Table 1. Evolution of the chemical composition during ripening of the control batch and starter-inoculated sausages.

	Samples	Ripening time (days)				
		0	7	14	21	28
Chemical composition						
Protein (%)	C	15.75±0.21^{a}	19.25±0.44^{b}	20.13±0.88^{b}	21.02±0.88^{b}	27.78±0.22^{c}
	S	15.97±0.44^{a}	19.47±0.22^{b}	21.22±0.66^{c}	22.97±0.22^{d}	28.87±0.44^{e}
Fat (%)	C	18.1±0.1^{a}	21.50±0.1^{b}	26.65±0.05^{c}	34.7±0.5^{d}	36.1±0.7^{e}
	S	18.2±0.2^{a}	22.1±1.9^{b}	30.1±1.3^{c}	37.8±0.6^{d}	39.1±0.7^{d}

Samples: C, control camel meat sausage; S, sausage inoculated with mixed starter cultures. Data are means±standard deviation. Different letters in the same row indicate significant differences ($p<0.05$).

Results showed that protein content and lipid content increased ($p<0.05$) during ripening of control and inoculated sausages. However, protein content and lipid content showed no significant differences ($p>0.05$) between control batches and those inoculated with a mixture of strains. DALLA SANTA *et al*. (2014) reported that there was significant difference between control and inoculated Italian sausages in protein content at the end of ripening. However, they did not show any significant difference in lipid content.

3.3. Lipid oxidation analysis

The TBARS values increased ($p<0.05$) during ripening from 0.25±0.04 to 0.95±0.16 and 0.8±0.1 mg MDA/kg of sample, respectively, in control and inoculated sausages (Fig. 3). Results showed that there was no significant difference ($p>0.05$) between the different samples. Many factors could affect lipid oxidation such as "chemical composition of raw material, processing conditions, light, and access to oxygen (AHN *et al*., 2002)". The TBARS values are lower ($p>0.05$) in starter-mediated sausages than those found in control ones. Similar results were found by KARGOZARI *et al*. (2014) and EL ADAB *et al*. (2016) who reported that *S. xylosus* and *S. carnosus* could limit lipid oxidation in dry fermented sausages due to their antioxidant activity.

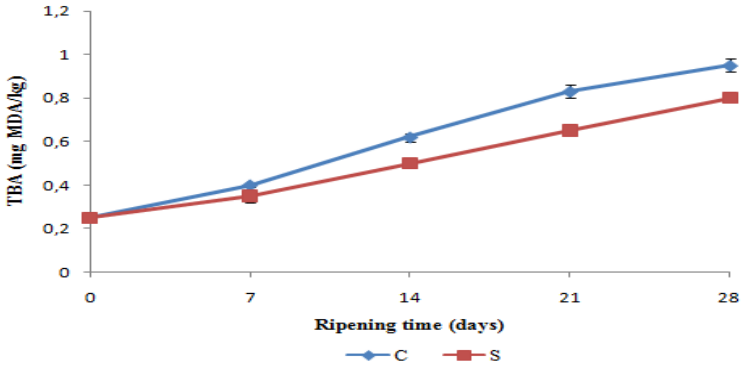

Figure 3. Changes in thiobarbituric acid (TBA) values during the ripening of control and inoculated sausages: C (control sausage), S (sausage inoculated with mixed starter cultures).

3.4. FAA content

The contents of total FAAs during ripening are shown in Table 2. The total FAAs increased through ripening ($p<0.05$); they reached at the day 28 values of 398.82 and 534.06 mg/100 g, respectively, for control and inoculated sausages. The highest total free amino acid content was found in inoculated samples. Similar results were found by ESSID and HASSOUNA (2013) and MEJRI *et al*. (2017). Many factors could affect the proteolysis in fermented sausages such as product formulation, technological ripening conditions and mixed starter cultures (HUGES *et al*., 2002).
The main amino acids present in the initial mixture were Arginine, threonine, glycine, alanine and glutamine with a concentration higher than 148.02 mg/100 g of dry matter. "The FAA content affects sensory properties impacting on fresh taste (glutamic acid and aspartic acid), sweet taste (glycine and alanine), bitter taste (arginine and leucine), sweet and bitter (lysine) and sour or salty (other FAA)" (DOMÍNGUEZ *et al*., 2016).

Table 2. Free amino acid content (mg amino acids/100 g of sausage) during ripening of the control batch and starter-inoculated sausages: C (control camel meat sausage), S (sausage inoculated with mixed starter culture).

FAA	Ripening time (days)				
	0	14		28	
	C	C	S	C	S
Aspartic acid	14.12±0.49^{e}	22.84±1.26^{d}	42.12±2.23^{b}	30.14±0.58^{c}	49.22±1.23^{a}
Glutamic acid	28.34±0.34^{e}	43.62±1.80^{d}	63.22±1.56^{b}	58.78±2.41^{c}	81.12±1.12^{a}
Serine + Glutamine + Histidine	8.62±1.30^{d}	9.02±2.31^{cd}	10.22±1.14^{c}	23.15±0.05^{b}	28.12±2.12^{a}
Arginine + Threonine + Glycine	84.32±2.50^{e}	95.56±2.14^{d}	130.02±0.77^{b}	120.45±1.36^{c}	145.02±0.89^{a}
Alanine	35.36±0.25^{e}	43.14±1.11^{d}	50.41±0.93^{c}	60.98±0.12^{b}	90.45±2.14^{a}
Lysine	10.65±1.18^{e}	22.34±2.23^{d}	25.17±1.25^{c}	26.23±0.04^{b}	28.78±1.45^{a}
Tryptophane	18.42±0.32^{e}	23.55±0.22^{d}	29.47±2.24^{b}	37.45±1.86^{c}	51.78±1.23^{a}
Isoleucine	3.12±0.45^{e}	4.65±1.01^{d}	9.12±0.88^{b}	7.52±1.03^{c}	13.12±0.58^{a}
Leucine	13.68±2.08^{d}	22.14±0.55^{c}	33.45±0.78^{b}	34.12±2.02^{b}	46.45±1.66^{a}
Total	216.63±0.79	286.86±0.71	392.2±0.58	398.82±0.85	534.06±0.49

Data are means±standard deviation.
Different letters in the same row indicate significant differences ($p<0.05$).

3.5. Biogenic amines contents

Total biogenic amine content in starter-mediated sausages was lower than in control ones during ripening ($p<0.05$) (Fig. 4). The total biogenic amine content was 285.67 mg/kg in sausages at the beginning of fermentation, which increased significantly during the first seven days of ripening to reach values of 338.28 and 310.14 mg/kg respectively, in the control sausages and sausages inoculated with *L. sakei*, *S. xylosus* and *S. carnosus*. Beyond the 7th day, total BA concentrations decreased significantly ($p<0.05$) to reach at the end of ripening respectively, values of 256.06 and 116.91 mg/kg. After 28 days of ripening, the total BA content in starter-mediated sausages was 54.3% lower than that in the control samples ($p<0.05$). The combination of three amine-negative bacteria, resulted in a drastic reduction of biogenic amine production. LEE *et al*. (2016) reported that *L. sakei* and *S. xylosus* could degrade BAs formed during fermentation through biogenic amine oxidases enzymes. These findings match with those found by HU *et al*. (2007) and NIE *et al*. (2014).
Changes in putrescine, cadaverine, spermine, spermidine and histamine concentrations during ripening of camel meat sausages are shown in Fig. 5. There was a decrease in all biogenic amines in the analyzed samples during the ripening. Spermine and spermidine were the predominant amine compounds in the sausages, followed by putrescine, histamine, and cadaverine. Our result is not in agreement with the study of GONZALEZ-FERNANDEZ *et al*. (2003) and LU *et al*. (2010), who reported that cadaverine and putrescine were the predominant amine compounds respectively, in Spanish pork sausage and traditional Chinese smoked horsemeat sausage. Many factors could contribute to the variability between the different types of products such as the microbiological quality of raw materials, ingredients, diameter of sausage, acidification, proteolysis and technological ripening conditions (BOVER-CID *et al*., 2001: BOZKURT and ERKMEN, 2002; LATORRE-MORATALLA *et al*., 2012).

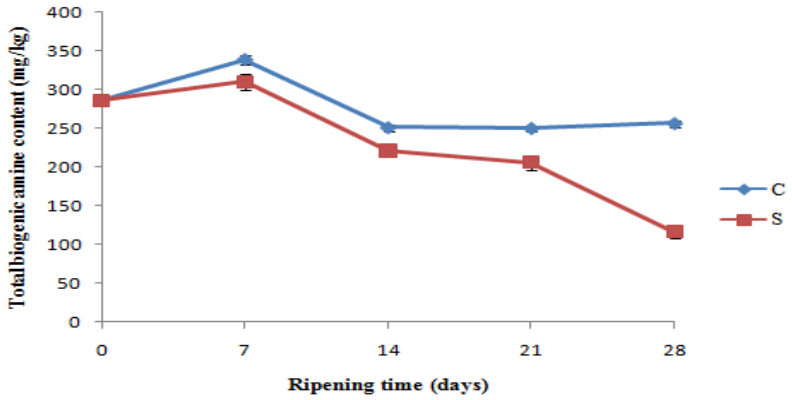

Figure 4. Changes in the total concentration of biogenic amines (mg/kg) in control and inoculated sausages: C (control sausage), S (sausage inoculated with mixed starter cultures) during ripening.

Spermidine and spermine are natural amines that always appear in fresh meat (HERÁNDEZ-JOVER *et al*., 1997). BOVER-CID *et al*. (2001) showed that these two BAs could be a source of nitrogen for some microorganisms, which could explain the decrease of spermidine and spermine concentrations during the ripening of dry fermented sausage.
The histamine concentrations were much lower in the starter-mediated sausages than that in the control batches (Fig. 5). At the end of ripening, the histamine content in inoculated sausages was 41.1% lower than that of the control ones ($p<0.05$). Many studies showed that histamine toxicity depended on the concentration upon absorption and it could be enhanced by the presence of cadaverine and putrescine (BOVER-CID *et al*., 2001; RENES *et al*., 2014). The drastic reduction of biogenic amine production is related to the drop of the pH of inoculated samples, which contribute to the decrease of the number of Enterobacteriaceae ($p<0.05$).
"Cadaverine can be used as an indicator of food hygiene (CHANG *et al*., 2012)." Cadaverine contents increased ($p<0.05$) during the first seven days of ripening from 0 mg/kg to 2.84 and 1.8 mg/kg for the control and starter-mediated sausages, respectively (Fig. 5). At the end of ripening, cadaverine accumulation was significantly ($p<0.05$) inhibited by 63.8% in starter-mediated sausages compared to the spontaneously fermented sausages. Results showed that there was a significant difference ($p<0.05$) between control batches and those inoculated with mixture of starter cultures. KOMPRDA *et al*. (2009) and RABIE *et al*. (2009) reported that the formation of cadaverine in fermented sausages is related to the presence of enterobacteria, which could be used as a chemical indicator of raw material and manufacturing practice hygiene. Values obtained for cadaverine indicate application of good hygiene in all phases of production.
The putrescine concentrations increased ($p<0.05$) during the initial 7 days. At the end of ripening, the putrescine concentration in inoculated sausages was 58.8% lower than that of the control ($p<0.05$) (Fig. 5).

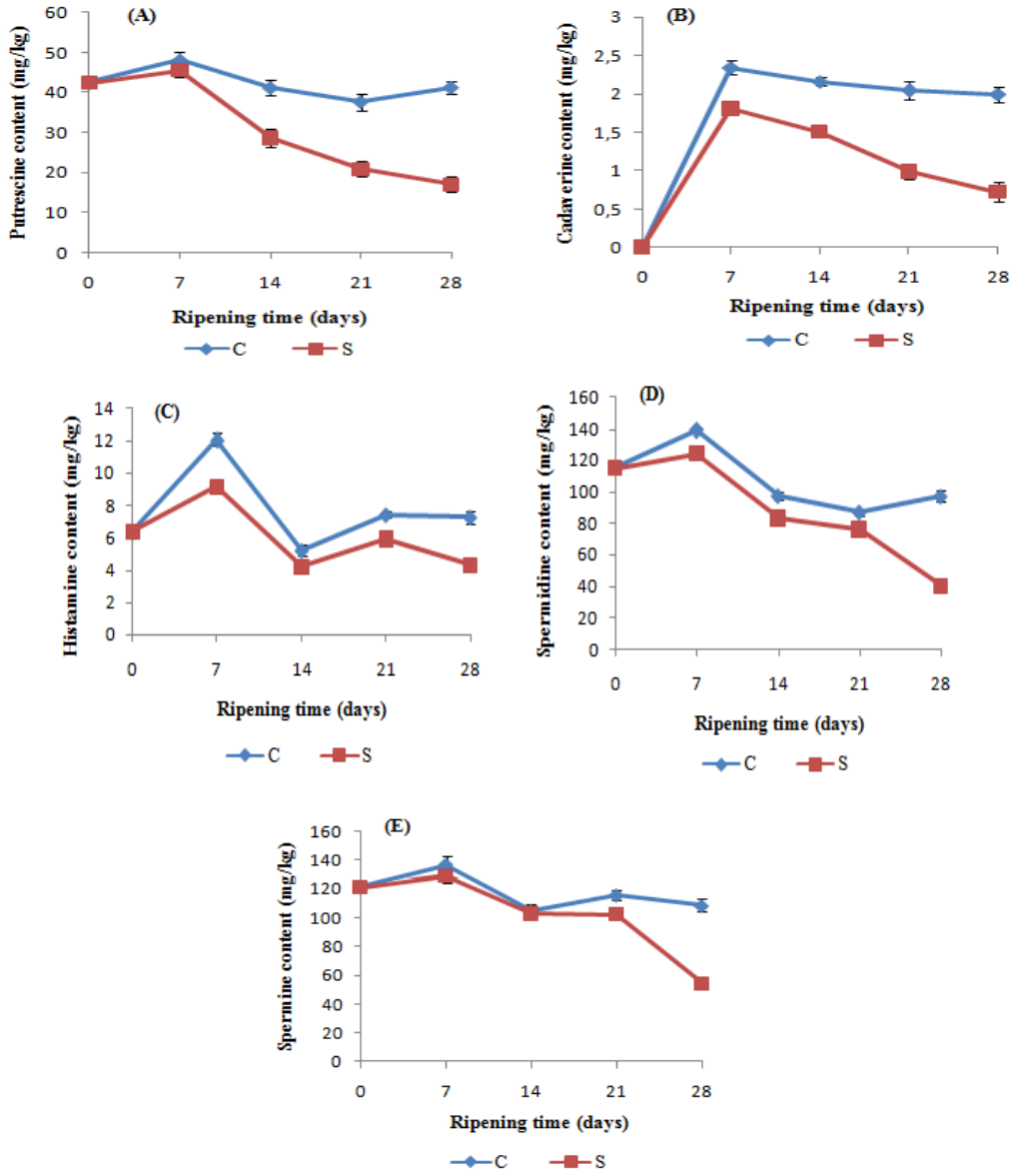

Figure 5. Changes in the amounts of putrescine (A), cadaverine (B), histamine (C), spermidine (D), and spermine (E) (mg/kg) in control and inoculated sausages: C (control sausage), S (sausage inoculated with mixed starter cultures) during ripening.

Our results showed that the combination of three bacterial strains could more effectively inhibit the formation of biogenic amines. Many studies reported the important role of *L. sakei* in the inhibition of the formation of biogenic amines (GENÇCELEP *et al*., 2012; LATORRE-MORATALLA *et al*., 2010; BAKA *et al*., 2011). "*L. sakei* is highly adapted to the fermented meat products and the optimal temperature of growth is between 15 and 25°C, which is the temperature range for sausages manufacture (BOVER-CID *et al*., 2001)." Moreover, GONZALEZ-FERNÁNDEZ *et al*. (2003) reported that *L. sakei* could reduce the formation of biogenic amine due to its strong acidifying activity. "BOVER-CID *et al*. (2001)

also reported that this specie is able to inhibit the production of biogenic amines in Spanish fermented sausage. However, when *L. sakei* was combined with *S. carnosus* or *S. xylosus* an even more effective reduction of amine accumulation was achieved compared with the effect of each strain used alone (LATORRE-MORATALLA *et al*., 2012)."
"MASSON *et al*. (1996) showed that *S. carnosus* and *S. xylosus* can be used as safe starter cultures. Whereas, STRAUB *et al*. (1995) found that *S. carnosus* contribute to the production of biogenic amines, but they did not find this for *S. xylosus*."

3.6. Texture profile analysis (TPA)

Fig. 6 shows the hardness, gumminess, chewiness, springiness, cohesiveness and resilience of control and inoculated sausages. Results showed that there were no significant differences between batches in any of the textural parameters studied.

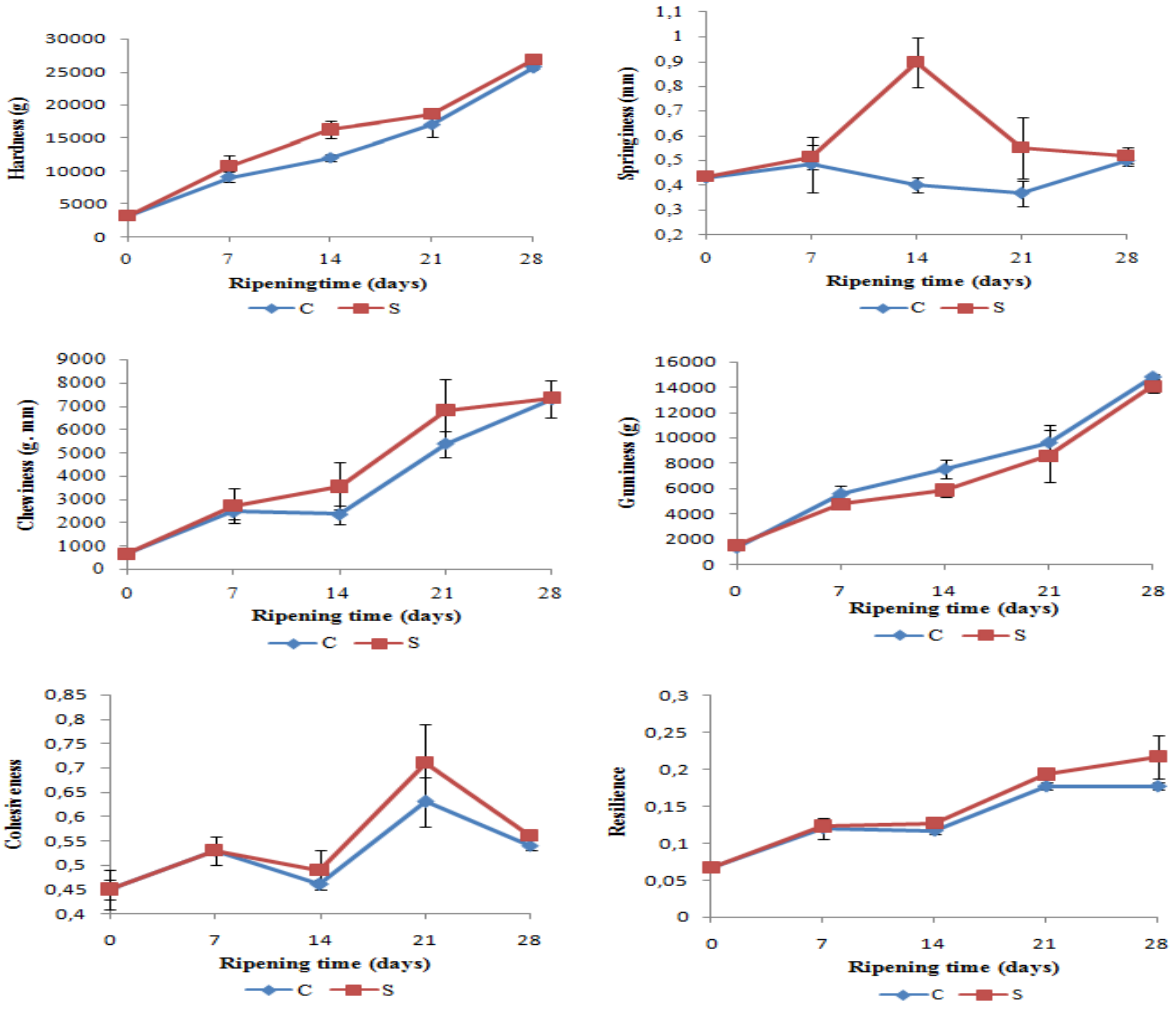

Figure 6. Changes in textural parameters during the ripening of control and inoculated sausages: C (control sausage), S (sausage inoculated with mixed starter cultures).

Hardness and springiness increased ($p<0.05$) during the ripening of control and inoculated sausages. Similar results were found by GONZALEZ-FERNANDEZ *et al*.

(2006) and BOZKURT and BAYRAM (2006) who reported that the increase of hardness could be explained by the elevated temperature during fermentation (24°C). The increase of springiness indicated that elasticity of camel meat sausage increased during the ripening period. Gumminess and chewiness values increased ($p<0.05$) during the whole process. These results are in agreement with those found by QINXIU *et al*. (2016).

3.7. Color properties

The color parameters, lightness (L*), redness (a*) and yellowness (b*) are shown in Fig. 7. L* values decreased ($p<0.05$) during ripening due to weight loss and higher myoglobin content (KADIM *et al*., 2008; OLIVARES *et al*., 2010). Moreover, results showed that L* values were significantly affected ($p<0.05$) by ripening time and not by the addition of mixed starter cultures ($p>0.05$). In relation to a* values, an increase ($p<0.05$) was observed during the first two weeks of ripening of dry fermented sausages followed by a significantly decrease which probably due to partial or total denaturation of nitrosomyoglobin because of the production of lactic acid (PEREZ-ALVAREZ *et al*., 1999; RUBIO *et al*., 2008). The evolution of a* and L* values found in this study was similar to that described by other authors (KAYAARDI *et al*., 2003; MEJRI *et al*., 2017). The b* values decreased ($p<0.05$) during ripening of both control and inoculated sausages. This finding was similar to that found by BOZKURT and BAYRAM (2006) during the ripening of sucuk.

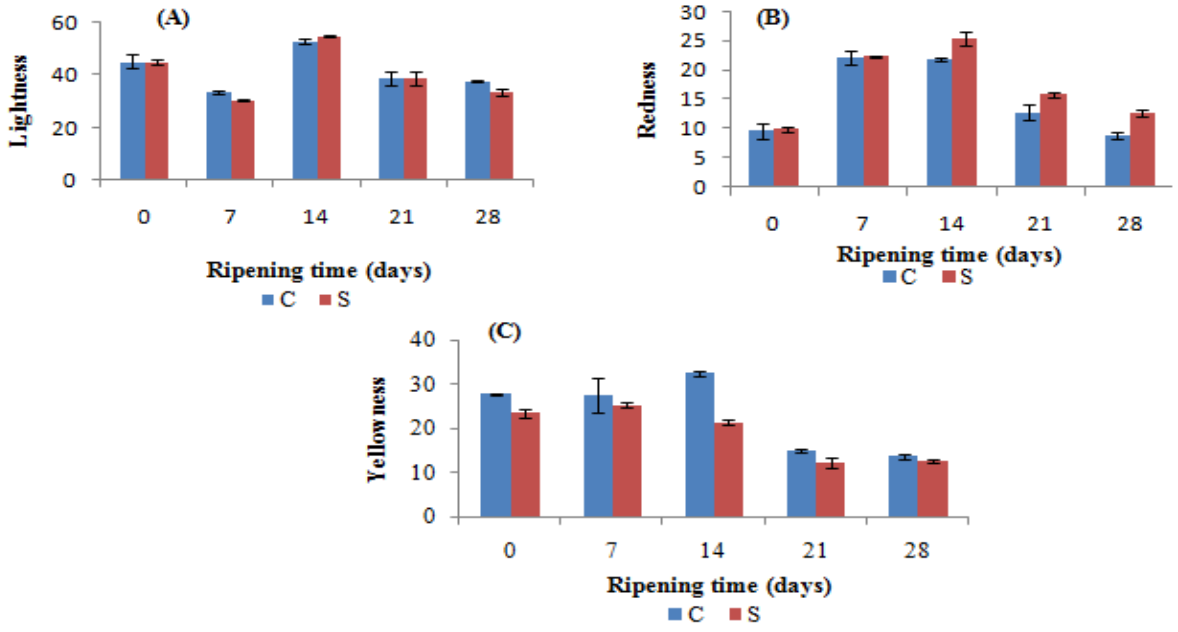

Figure 7. Changes in lightness (A), redness (B) and yellowness (C) during the ripening of control and inoculated sausages: C (control sausage), S (sausage inoculated with mixed starter cultures).

3.8. Sensory evaluation

The results of a sensorial evaluation of control and inoculated sausages are shown in Fig. 8. In fact, the starter-mediated sausages showed a more pronounced red color when

compared to control ones ($p<0.05$). RAVYTS *et al*. (2010) reported that the red color of fermented sausages was often related to the nitrate reductase activity of CNS. Moreover, inoculated sausages had higher scores of acidity ($p<0.05$) which could be explained by the lactic acid produced from bacterial carbohydrate metabolism. Additionally, sausages inoculated with mixed starter cultures showed a significantly greater overall aromatic intensity than that noted on control samples, which could be due to the lipolytic, acidifying and proteolytic activities of strains of *S. xylosus*, *S. carnosus* and *L. sakei* inoculated in meat products. Our results showed that there was no significant difference ($p>0.05$) in fat intensity. Inoculated sausages showed a significantly higher firmer texture ($p<0.05$) than those found on control samples. Similar results were found by FONSECA *et al*. (2013).

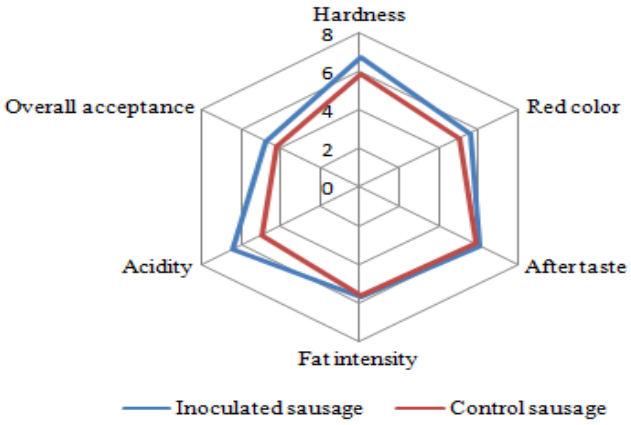

Figure 8. Sensory evaluation of control sausages and sausages inoculated with mixed starter cultures.

4. CONCLUSION

This study focused on the effect of mixed starter cultures on microbiological, biochemical and sensory characteristics of a dry fermented camel meat sausage. The bactericidal properties of *L. sakei* improved the hygienic quality of sausages by decreasing the number of Enterobacteriaceae. Inoculation of dry sausages with a mixture of strains, significantly delayed lipid oxidation and improved sensory characteristics. Moreover, the total biogenic amine concentration in starter-mediated sausages was much lower than that in the control samples. These results suggest that *L. sakei*, *S. xylosus* and *S. carnosus* could be used as safe mixed starter cultures in dry sausage production to inhibit biogenic amine formation and enhance sensory quality.

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