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INVESTIGATION OF LISTERIA MONOCYTOGENES IN BRANDED AND NON-BRANDED SAUSAGE CONSUMED IN KONYA PROVINCE

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ABSTRACT

Twentyfour branded and non-branded 24 sausage specimens were examined for presence of Listeria spp. Listeria isolation were performed in two enrichment periods, according to FDA (Food and Drug Administration) approved method as described by Lovett and Hitchins. After 48-hours enrichment period, one Listeria monocytogenes (4.16%), two Listeria innocua (8.33%) proliferated in branded sausages, and there were two L.monocytogenes (8.33%), four L.innocua (16.66%) in non-branded sausages (total 9, 18.75%). There were difference in Listeria numbers between in branded and in non-branded sausages (p>0.05). We may conclude that sausages are not contaminated with L. monocytogenes with a potential to create infection.

Keywords: *Listeria monocytogenesis, sausage*

KONYA İLİNDE TÜKETİLEN MARKALI VE MARKASIZ SUCUKLARDA LISTERIA MONOCYTOGENES'İN ARAŞTIRILMASI

ÖZET

Konya ilinde tüketime sunulan markalı ve markasız toplam 48 sucuk örneği Listeria türleri yönünden incelendi. Listerialar'ın izolasyonları FDA tarafından öngörülen Lovett ve Hitchins'in bildirdiği metoda göre iki ayrı zenginleştirme süresi ile yapılmıştır. 48 saat zenginleştirme süresinde markalılarda 1 adet (%4.16) Listeria monocytogenes, 2 adet (%8.33) Listeria innocua, markasızlarda 2 adet (%8.33) L.monocytogenes, 4 adet (%16.66) L.innocua olmak üzere toplam 9 adet (%18.75) Listeria türü izole edilmiştir. Sonuçlar istatistiksel olarak chi square metodu ile analiz edilmiştir. Markalı ve markasız sucuk örneklerinde üreyen Listeria türleri arasında anlamlı bir fark bulunmuştur(p>0.05). Konya'da tüketime sunulan sucukların Listeriozis açısından rezerv oluşturmayacağı düşüncesine varılmıştır.

Anahtar Kelimeler: *Listeria monocytogenesis, sucuk*

INTRODUCTION

Listeriosis is a specific bacterial disease mostly encountered in new-borns and immune deficient patients and is characterized with granulomatous lesions and abscesses (Hugas et al., 1996). Disease progresses as primary bacteremia and more commonly memnengitis in humans (Foodborne Listeriosis report, 1988; Howard et al., 1987; Buncic et al., 1991). Listerias are frequently encountered in the nature, reproduce easily and are resistant to several environments. These make them contaminate food quite easily (Carpenter and Harrison, 1989). Listeriosis, previously reported in single cases secondary to contaminated food, resulted in death rates reaching 30% in endemics in Canada in 1981, in USA in 1985, in Swiss between 1983-1987 (Lovett and Hitchins,1988; McLauchlin and Gilbert, 1990; Kampelmacher, 1988; Hugas et al., 1995). Listeriosis in humans was primarly discovered to be secondary to consumption of contaminated milk and milk products (Fleming et al., 1985). Later studies confirmed that the other food materials, such as meat and meat products, could be contaminated copiously with this microorganism (Çiftçioğlu and Derbentli, 1988; Cherubin et al., 1981). Other authors reported that meat products like ground meat, sausage harbor high numbers of listeria species (Seeliger, 1988; Sümbüloğlu; Tekinşen et al., 1980). Some characteristics of listeria species, such as wide-spread appearance in nature, resistance to several environments, reproduction in refrigerator temperatures increases the dimensions of health hazards (Erdal, 1993). Development of strategies to control *L. monocytogenes* in last product are important to prevent food based listeriosis cases (Erdal, 1993). We aimed to investigate frequency of *L. monocytogenes* in branded and non-branded sausages, produced in Konya.

MATERIAL AND METHODS

Twentyfour branded sausage specimens were bought from stores and delicatessens, and non-branded 24 sausage specimens were bought from local bazaars and butchers, in Konya.

Tryptone Soy Broth (TSB-Oxoid CM 128) as basic enrichment media in isolation and identification of Listeria species and Yeast Extract (YE-Oxoid L 21), as a secondary enrichment media were used in the study. Nalidicsic acid (Sigma N 4282), Akriflavine (Sigma A 8251) and cycloheximide (Sigma C 6225) were added to the growth media as supplements. Listeria Selective Agar Base (Oxoid CM 856) and Listeria Selective Suplement (Oxoid SR 140) as selective growth media and correction media, Triptone Soy Agar (TSA Oxoid CM 131) were used.

Isolation procedure was done in two different enrichment periods according to method reported by Lovett and Hitchins (Kılıçtugay, 1987). Twentyfive grams of sausage specimen were delivered into 225 mls of basic enrichment media and incubated at 30ºC for 24 hours. Following this procedure, 0.1 ml specimen was drawn from each growth media and incubated for 24 hours in secondary growth media. After the first enrichment, specimens were incubated in main enrichment growth media for one week at 30ºC. Following the enrichment procedures, specimens were taken from growth media and diluted in 1/10 ratio in %0.5 KOH solutions. Specimens of 0.1 ml volume from these dilutions were delivered to selective LSA growth media and incubated for 24 hours at 30ºC. S type colonies surrounded by black zone from this media were taken and passaged to TSA for confirmation. After incubation at 30ºC for 24 hours. Gram, catalase and motility tests were done to yellow-white colonies and those subspecies with positive results were identified as Listeria. Isolaties were named with identification tests. Chi square method was used to analize differences in times of enrichment for production of Listeria and to determine species differences of listeria in branded and non-branded sausages, proliferating in 48 hours and 1 week (Breuer and Prandl, 1988).

RESULTS

Species and numbers of Listeria in branded and non-branded sausages , proliferating in 48 hours and 1 week periods are given in Table-1. In 48-hours enrichment period, 3 (12.5%) Listeria species were found in branded sausage specimens. One (4.16%) was *L. monocytogenes*, two (8.33%) were *L. innocua*.

Table 1: Species and numbers of Listeria in branded and non-branded sausages , proliferating in 48 hours and 1 week periods.

Species	48 –hour enrichment period			1 week enrichment period		
	Branded n=24	Non-branded n=24	Total=48	Branded n=24	Non-branded n=24	Total=48
Listeria	3 (%12,50)	6 (%25,00)	9 (%18,75)	1 (%4,16)	4 (%16,66)	5 (%10,41)
L. monocytogenes	1 (% 4,16)	2 (% 8,33)	3 (% 6,25)	-	1 (% 4,16)	1 (% 2,08)
L. innocua	2 (% 8,33)	4 (%16,66)	6 (%12,50)	1 (%4,16)	3 (%12,50)	4 (% 8,33)

In the same period, there was no statistical difference concerning proliferating *Listeria* numbers between branded and non-branded sausages ($p>0.05$). In one week enrichment period, one listeria species (*L.innacua*) was found in branded sausages (4.16%). *L. monocytogenes* could not be isolated. Four *Listeria* species were isolated in non-branded sausages: one was *L. monocytogenes* (4.16%), three were *L. innocua* (12.5%). In this period, there was no statistical difference in numbers of Listeria proliferating in specimens from branded and non-branded sausages ($p>0.05$). There were also no difference for numbers of Listeria species in branded and non-branded sausages, proliferating in 48 hours and 1 week enrichment periods ($p>0.05$). There was no difference in enrichment periods for proliferating *Listeria* ($p>0.05$).

DISCUSSION

In this study, we reproduced 3 *L. monocytogenes* (6.25%), 6 *L. innocua* (12.5%) (total 9) *Listeria* (18.75%) species in total of 48 branded and non-branded sausage specimens. Kaya and Gokalp (Kaya and Gökalp,) found 11 Listeria spp (39%), Cıftcıoglu and Ugur found 11 (11%) *Listeria spp*, Erdal (Vanderlinde and Grav, 1991) found 2 (2.4%) Listeria spp. Our numbers were lower than their findings, but higher than those of other authors (Erol et al., 1999; Vanderlinde and Grav, 1991). These differences in numbers may be secondary to contamination of different supplements and different production methods. In this study, there was no difference Listeria numbers between branded and non-branded sausages in 48-hours time period. However the proliferation ratio (25%) in non-branded sausages was higher in non-branded sausages then branded sausages (12.5%). For this reason, we think that non-branded sausages are more contaminated with *Listeria* and pose a greater potential of Listeriosis. In the other countries, *Listeria* contaminations were reported to vary between 40-97% (Cherubin et al., 1981; Seeliger, 1988; Farchmin, 1983). Apparently, our results were lower then studies in other countries. Total mesophilic aerobic bacteria and lactobacilla overwhelming flora after maturation of fermented sausages may prevent proliferation of *Listeria*. Some authors reported that *Lactobacillus sake* (Lb.706) and similar spp, producing bacteriosin and playing role in maturation of sausage inhibits proliferation of *Listeria* spp (Erdal, 1993; Weis, 1989; Johnson et al., 1989). Other authors reported that *L. monocytogenes* does not proliferate in food with high total microorganism numbers (Çiftçioğlu and Ugur 1992; Çetin and Derbentli, 1988). Similar studies demonstrated that food in our country are rich in total microorganism numbers

(Çetin and Derbentli, 1988, Weis, 1989). We think that lower number of *Listeria* in sausage specimens could be secondary to high bacterial contamination and presence of microorganisms producing bacteriocin.

We performed Listeria isolation in two enrichment periods, according to FDA approved method as described by Lovett and Hitchins (Kılıçtugay, 1987). We could not demonstrate any difference in enrichment periods of Listeria ($p>0.05$). In total, Listeria proliferating rate was 20% in 48 hours, 11% in one week. Taking into account the great difference in proliferation ratios, 48-hour incubation time yield better results. Finally, we may conclude that sausages produced in our country are not contaminated with *L. monocytogene*s with a potential to create infection.

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