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Full Length Research Paper

Prevalence of *Listeria* species in retail quail products from Isfahan, Iran

Mohsen Panahi Dorcheh*, Rafie Sohrabi and Mohammad Salajegheh

Scientific Association of Veterinary Medicine, Faculty of Veterinary Medicine, Islamic Azad University, ShahreKord Branch, Iran.

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***Listeria* species are Gram positive, short, non sporing rods, microaerophilic. Of the six species currently recognized, *Listeria monocytogenes* is the most important as it causes a range of infections in humans and animals. This study was undertaken to determine the occurrence of *Listeria* spp. in retail quail products in Isfahan, Iran. A total of 150 samples of meat, liver, heart, kidney, and feces of quail were obtained from retail stores in Isfahan, and analyzed using standard culture methods and biochemical tests. Out of the total 150 samples, 10 (6/6%) were positive in *Listeria* spp. The occurrence of *Listeria* spp. in samples of meat, liver, heart, kidney, and feces was 10, 3, 0, 0, and 20% respectively. Only one sample was contaminated with *L. monocytogenes* (0/6%) and other samples were contaminated with *Listeria innocua* (6%).**

Key words: Quail, culture method, *Listeria* species.

INTRODUCTION

The marked increase of contamination in food industry especially meat and chicken products by pathogenic bacteria has raised a great public concern, *Listeria* species, especially *Listeria monocytogenes* has been associated with a wide variety of food sources, particularly meat and chicken (Endang et al., 2003). *Listeria* are Gram positive, facultative anaerobic, non-spore forming, rod shaped bacteria with a low C+G content. The genus consists of six species: *L. monocytogenes*, *Listeria innocua*, *Listeria seeligeri*, *Listeria welshimeri*, *Listeria ivanovii*, and *Listeria grayi*. *L. monocytogenes* is the primary human pathogen; although, there have been rates of illnesses caused by *L. selegeri*, *L. ivanovii*, and *L. innocua* (Jeyaletchumi et al., 2010). *Listeria* spp. has been isolated from poultry, red meat, and meat products in many countries around the world; although, these foods have not been associated with documented outbreaks of human listeriosis. The detection of *Listeria* spp. in meat is a particular concern in terms of consumer safety, as these organisms are capable of growing on both raw and cooked meat at refrigeration temperatures (EI-Malek et al., 2010).

In the past 25 years, *L. monocytogenes* has become increasingly, a dangerous bacteria as a food-associated pathogen. Because of its high case of fatality rate, listeriosis is one of the most frequent causes of death due to food borne illness.

L. monocytogenes is an intracellular pathogen affecting mainly children, pregnant women, the aged and immune-challenged individuals (Schlech, 2000; Liu, 2006). In addition, a wide variety of animals, including sheep, cattle, goats, pigs, rabbits, mice, birds, and fish are also infected. The pathogen is also responsible for *Listeria* infections that can lead to abortion, bacteraemia, sepsis, and meningoencephalitis (Sukhadeo and Trinad, 2009). The incidence of listeriosis is relatively rare and represents less than 0.1% of all food-borne illnesses, but causes infections with very high mortalities (20 to 30% deaths) (Mead et al., 1999).

The first epidemiologically confirmed food borne outbreak of listeriosis occurred in 1981 in Canada (Schlech et al., 1983) and was linked to the consumption of coleslaw. Other outbreak of human listeriosis have been

*Corresponding author. E-mail: jubin_2010@yahoo.com. Tel: (+98) 913-2280511. Fax: (+98) 311-6259809.

Table 1. Main laboratory tests for the differentiation of *Listeria* spp.

Species	Hemolysis	Phosphatidylinositol phosphatase	Acid production	
			L-Rhamnose	D-Xylose
L. monocytogenes	+	+	+	-
L. ivanovii	+	+	-	+
L. seeligeri	+	-	-	+
L. innocua	-	-	V	-
L. welshimeri	-	-	V	+
L. grayi	-	-	V	-

Table 2. Distribution of *Listeria* spp. in different poultry meat product.

Sample type	Number of sample		%
	Examined	Positive	
Meat	30	3	10
Heart	30	0	0
Liver	30	1	3
feces	30	6	20
Kidney	30	0	0
Total	150	10	6/6

associated with milk (Fleming et al., 1985), soft cheese (Linnan et al., 1988; James et al., 1985), jellied pork tongue, and other foods of animal or vegetable origin (Ryser and Marth, 1999).

MATERIALS AND METHODS

Culture method for isolation and identification of *Listeria* spp

In this study, 85 samples of meat, liver, heart, kidney, and feces of quail (17 samples of everyone) were obtained from retail stores in Isfahan. All products had been properly stored or refrigerated.

Detection of *Listeria* spp. in the poultry meat products was performed according to the standard culture method (Gnanou et al., 2005). At first, poultry meat purchased was divided into 25 g portions, and the portions were placed into Stomacher bags, and were stored at 3°C or frozen at -18°C. Enrichment culturing was performed according to the ISO 11290-1 reference method (Anonymous, 1996). Samples (25 g) were aseptically added to 250 ml of *Listeria* Enrichment Broth (UVM, Difco 0223) and were pre-enriched at 30°C for 20 to 24 h, and then 0.1 ml of the pre-enriched culture was transferred to Fraser Broth (Difco 0219) at 35°C for 24 to 48 h. After selective enrichment, samples were streaked into the *Listeria* Selective Agar (PALCAM) and were incubated at 35°C for 24 to 48 h. The plates were examined for the presence of *Listeria* colonies.

From 52 poultry meat products, 12 samples which were brown-greenish and surrounded by a black halo, were transferred to Trypticase Soy Agar supplemented with 0.6% yeast extract (TSA-YE, Difco) and were incubated at 30°C for 24 to 48 h.

Biochemical tests such as the presence of catalase, hemolytic, fermentation of xylose and rhamnose, oxidase, and umbrella-shaped growth in motility in sulfide-indole-motility (SIM) medium (sulfur reduction test, indole production, motility). Gram staining was also performed on the doubtful colonies. The main tests are presented in Table 1 (Janzten et al., 2006).

RESULTS

Out of a total of 150 samples, 10 (6/6%) were positive for *Listeria* spp. *Listeria* spp. were isolated from meat, liver, heart, kidney, and feces of quail samples. Distribution of *Listeria* spp. in products considered in this study is presented in Table 2. The level of contamination of food samples by *Listeria* spp. varied. The highest rate was observed in feces samples (20%), followed by meat (10%) and liver (3%).

DISCUSSION

Meat and chicken products have been frequently contaminated with *L. monocytogenes* and may serve as means of other pathogenic organisms. The frequent occurrence of *L. monocytogenes* in meat and chicken may pose a potential risk for consumers. Human infections primarily result from eating contaminated food and may lead to serious and potentially life-threatening listeriosis (EI-Malek et al., 2010). Increasing evidence suggests that substantial portions of cases of human listeriosis are attributable to the food borne transmission of *L. monocytogenes* (Low and Donachie, 1997).

Molla et al. (2004) reported that raw meat products as expected showed a high level of contamination with *Listeria*

spp. (50.6%). It is generally assumed that such products cannot be free from *Listeria*, because of the slaughter methods (evisceration) and food processing that allows greater chance for contamination. Furthermore, *Listeria* spp. are ubiquitous in the environment (Vitas et al., 2004). People handling food at different levels can also be sources of contamination.

In this study, 52 poultry meat samples were examined for the presence of *Listeria* spp. Of the total of 52 meat samples, 11 (21.15%) isolates were contaminated with *L. innocua* and only one sample was contaminated with *L. monocytogenes*. The occurrence of *Listeria* spp. in samples of meat, liver, heart, kidney, and feces were 15, 5, 0, 0, and 30%, respectively. This was comparable with results of surveys undertaken in other countries. This suggests the presence of a significant public health hazard linked to the consumption of foods contaminated with *Listeria* spp.

In this study, *L. innocua* was the predominant isolated species, and it is more frequently isolated than *Listeria* spp. When the results of this study were compared with those of other researchers, a considerably higher level of contamination of poultry raw meat from supermarkets in Spain was reported by Capita et al. (2001). *Listeria* was found in as much as 95% of examined carcasses, of which 32% of them were recognized as *L. monocytogenes* and 66% as *L. innocua.* Vitas et al. (2004) reported 36.1% positive samples of raw poultry their research carried out in Northern Spain. Kosek-Paszkowska et al. (2005) reported 63% contamination from poultry meat products. When several studies in various countries are compared, *L. monocytogenes* isolation rates seem to vary significantly. This wide variation may be explained in terms of geographic location, isolation methods, kinds of media employed and hygienic production, HACCP application, etc.

Researchers found out that *L. innocua* grows faster than the pathogenic species in enrichment broth media and may therefore overgrow than *L. monocytogenes*. Adzitey and Huda (2010) reported that *L. innocua* occupies the same ecological niche and its high incidence signifies potential contamination by *L. monocytogenes*. Higher records were reported by several investigators, such as Hassan et al. (2001) who found *Listeria* spp. in 17 (73.9%) of the 23 samples of imported frozen beef in Malaysia, Inoue et al. (2000) isolated *L. monocytogenes* in 12.2% of minced meat samples in Japan, and Buncic (1991) detected *L. monocytogenes* in 69% of minced meat samples in Yugoslavia. It is interesting to note that *L. innocua* was isolated predominantly among *Listeria* spp.

Other studies indicated that *L. innocua* was the most predominantly isolated species in a variety of meat samples; Yucel et al. (2005) reported *L. innocua* in 83.3% of the raw minced meat, 57.6% of the raw chicken meat, 63.1% of the raw beef, 9.6% of the cooked red meat, and10.7% of the cooked chicken samples. Furthermore, detection of *L. monocytogenes* in foods can be difficult as these bacteria are normally found in very low numbers in the presence of a heterogeneous micro flora. The most frequent *Listeria* isolated from food are *L. monocytogenes* and *L. innocua*. Several studies have demonstrated that *L. innocua* is found in food more frequently than *L. monocytogenes* (Walsh et al., 1998). The reasons for the higher frequency of recovery of *L. innocua* remain unclear yet. However, this may result from either a naturally higher prevalence or from preferential selection of *L. innocua* during laboratory detection procedures (Gnanou et al., 2005).

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