

Distribution and Serotyping of *Listeria monocytogenes* in Seafood Processing Plants

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Listeria spp. were isolated from the samples submitted from various seafood plants such as raw materials, products, swab samples of plants floor and conveyor belts through the whole processing procedures. All the samples were collected from 3 kinds of seafood plants such as a imitation crab meat plant, *jeotgal* plant and frozen seafood plant. And also serotypes of the identified *L. monocytogenes* were determined. Among the 301 strains of isolated *Listeria* spp., 96 strains, 179 strains and 26 strains were identified as *L. monocytogenes*, *L. innocua* and *L. welshimeri*, respectively. While among the 145 strains of *Listeria* spp. isolated from the imitation crab meat plant, 74 (51.0%) strains, 64 (44.1%) strains and 7 (4.8%) strains were identified as *L. monocytogenes*, *L. innocua* and *L. welshimeri*, respectively. In the case of the 126 strains of *Listeria* spp. isolated from the frozen seafood plant, 22 (17.5%) strains of *L. monocytogenes*, 93 (73.8%) strains of *L. innocua*, and 11 (8.5%) strains of *L. welshimeri* were detected. Among the 30 strains isolated from a *jeotgal* plant, 22 (73.3%) strains of *L. innocua* and 8 (26.7%) strains of *L. welshimeri* were detected. The detection rates of *L. monocytogenes*, one of the very important food poisoning bacteria especially in frozen and/or refrigerated seafoods, were relatively high as 77.1% (74/96 strains) in a imitation crab meat plant and 22.9% (22/96 strains) in a frozen seafood plant, but not detected from *jeotgal* plant. Distribution of *L. monocytogenes* serotypes and characterization were examined. The serotypes of 96 *L. monocytogenes* isolated from pork skin, pork fat, the floor and conveyor belts were 1/2a (59.4%), 1/2b (6.2%), 1/2c (12.5%) and unknown serotypes (21.9%). Unknown serotypes were divided into three specific groups by the O antigen they have.

Key words: *Listeria monocytogenes*, Seafood, Serotype

Introduction

Since *Listeria monocytogenes* was recognized as a food-borne pathogen in the early 1980s, listeriosis outbreaks and sporadic cases have been linked to several food items. Documented sources of the bacteria to patients were confirmed to be from cheese, vegetables, pasteurized milk and meat products (Schlech et al., 1983; Linnan et al., 1988; Farber and Peterkin, 1991). In Korea, although this bacteria is not well known, since it has been detected in hard-shelled mussel imported from New Zealand and produced domestically, and has also been found in frozen foods such as pizza and ice cream,

L. monocytogenes has become of major concern for public health authorities and the food industry. Although human listeriosis may be caused by all 13 serotypes of *L. monocytogenes*, serotypes 1/2a, 1/2b and 4b cause most of the cases. Serotype 4b strains are most widely distributed among clinical isolates, accounting for about 40% of sporadic listeriosis in humans, and the most recent major epidemics have involved serotype 4b strains (Zheng and Kathariou, 1995; Louie et al., 1996). Because *Listeria* spp. are able to survive and grow at low temperatures, existence of *L. monocytogenes* are a major problem in the food industry. Since most seafoods are ready-to-eat foods, the possibility of food poisoning by this organism is extremely high. Furthermore, research about the incidence and serotyping of *L. monocy-*

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togenes in seafoods and seafood processing plants are frequent in many countries (Dillon et al., 1994; Rørvik et al., 1995; Johansson et al., 1999). However, in the case of Korea, although there has been research on the distribution and serotyping of *L. monocytogenes* in meat products and frozen food (Hong and An, 1998; Gu et al., 1995), there has not been enough on seafoods and seafood processing plants. Because the consumption of seafoods is high in Korea, investigation on the distribution and serotyping of *L. monocytogenes* is a very important subject.

The purpose of this study was to investigate the actual conditions of the contamination and to provide basic information for application of the HACCP system by examination of the distribution and serotyping of *L. monocytogenes* in seafood and seafood processing plants.

Materials and Methods

Sample collection

Three seafood processing plants in Korea were selected during 1999~2000 for this study. They were an imitation crab meat plant in Busan, a *jeotgal* plant in Pohang and a frozen seafood plant in Busan. Samples were collected from raw materials, raw products and processing environments by FSIS sample collection guidelines (Berry, 1996).

Microbiological analysis

The isolation procedure was started within 12 hr. Viable cell counts were examined by the A.P.H.A. method (Busta et al., 1984). For the isolation of *Listeria* spp., 25 mL of sterile saline including the swabs or sponges, 25 mL of the water sample or 25 g of raw product were inoculated in 225 mL of *Listeria* Enrichment Broth as described by Lovett et al. (1987) and stomached (Lab Blender Stomacher 400, Seaward Co.) at 230 rpm for 120 sec. All samples were incubated at 30°C for 48 hr. A portion of the enrichment broth was streaked on PALCAM agar plate. After 24~48 hr incubation at 37°C, the plates were examined for typical *Listeria* colonies, which were streaked on sheep blood agar plates for CAMP test. Typical colonies were confirmed as *Listeria* spp. by Gram stain, catalase test, motility

test, CAMP test and tests for fermentation of mannitol, rhamnose and xylose. Furthermore, *L. monocytogenes* was reconfirmed by API *Listeria* kit (bio-Merieux Co., France).

Serotyping

The serotyping scheme designed by Seeliger and Höhne (1979) was as followed. Serotyping was performed using commercial *Listeria* antisera (Denka Seiken, Tokyo, Japan) as described by the manufacturer (Denka Seiken, 1995).

Results and Discussion

The results of viable cell counts were similar in each plant and the most contaminated point was the floor (Table 1). Of the 44 samples from the imitation crab meat processing plant, *Listeria* spp. were detected in 15 samples and the detection rate was 34.1%. Among the 145 strains of *Listeria* spp. isolated from the 15 samples, 74(51.0%) *L. monocytogenes*, 64(44.1%) *L. innocua* and 7(4.8%) *L. welshimeri* were detected. *L. monocytogenes* was detected in samples from the floor, after freezing material and conveyor belts. Of the 63 samples from the frozen seafood processing plant, *Listeria* spp. were detected in 32 samples and the detection rate was 50.8%. Among the 126 strains of *Listeria* spp. isolated from the 32 samples, 22(17.5%) *L. monocytogenes*, 93(73.8%) *L. innocua*, and 11(8.5%) *L. welshimeri* were detected (Table 2). *L. monocytogenes* was detected from pork fat, pork skin and the floor. Of the 16 samples from the *jeotgal* processing plant, *Listeria* spp. were detected in 5 samples and detection rate was 31.3% (Table 3). Among the 30 strains of *Listeria* spp. isolated from the 5 samples, 22(73.3%) strains of *L. innocua* and 8(26.7%) strains of *L. welshimeri* were detected but *L. monocytogenes* was not detected. Among the three plants, the frozen seafood plant was the most contaminated by *Listeria* spp. In the frozen seafood plant, although the isolation of *L. monocytogenes* was lower than the imitation crab meat plant, the detection rate of *Listeria* spp. (50.8%) was high and detected from most of the isolated samples. In the *jeotgal* plant, the detection rate of *L. innocua* was the highest.

In the imitation crab meat plant, detection of *L.*

Table 1. Distribution of *Listeria monocytogenes* and other *Listeria* spp. in samples from the imitation crab meat processing plant

Samples	No. of samples ¹⁾	No. of isolates from samples for				Viable cell counts (CFU/mL or g)	
		<i>Listeria</i> spp.	<i>L. monocytogenes</i>	<i>L. innocua</i>	<i>L. welshimeri</i>	Range	Average
Mixture	1 / 4	1	0	1	0	$2.1 \times 10^5 \sim 1.6 \times 10^5$	1.9×10^5
After sealing	1 / 2	2	0	2	0	$1.8 \times 10^5 \sim 2.2 \times 10^5$	2.0×10^5
Cooling	0 / 2	0	0	0	0	2.1×10^3	2.1×10^3
Before freezing	0 / 2	0	0	0	0	$2.0 \times 10^2 \sim 8.0 \times 10^2$	5.0×10^2
After freezing	1 / 2	24	18	6	0	$8.6 \times 10^4 \sim 1.1 \times 10^5$	9.8×10^4
Before disinfection	0 / 2	0	0	0	0	$2.1 \times 10^5 \sim 3.3 \times 10^5$	2.7×10^5
After disinfection	0 / 2	0	0	0	0	>30	>30
Floor	8 / 8	76	37	33	6	$4.5 \times 10^5 \sim 1.8 \times 10^8$	5.5×10^7
Wall	0 / 4	0	0	0	0	$9.3 \times 10^3 \sim 9.4 \times 10^5$	3.2×10^4
Water	0 / 2	0	0	0	0	>30~ 5.4×10	> 2.7×10
Worker	0 / 2	0	0	0	0	$2.5 \times 10^2 \sim 2.5 \times 10^2$	2.5×10^2
Knife	1 / 2	4	0	4	0	7.6×10^3	7.6×10^3
Silent cutter	2 / 4	3	0	3	0	$2.7 \times 10^3 \sim 4.3 \times 10^6$	3.5×10^4
Forklift truck	1 / 2	10	0	9	1	5.7×10^5	5.7×10^5
Conveyor belt	1 / 2	25	19	6	0	$7.1 \times 10^4 \sim 3.2 \times 10^6$	1.6×10^5
Pan	0 / 2	0	0	0	0	2.4×10^3	2.4×10^3
Total	15/44	145	74/145 ²⁾ (51.0%)	64/145 ³⁾ (44.1%)	7/145 ⁴⁾ (4.8%)		

¹⁾No. of samples that were contaminated with *Listeria* spp. / No. of submitted samples.

²⁾No. of *L. monocytogenes* / No. of *Listeria* spp. (%).

³⁾No. of *L. innocua* / No. of *Listeria* spp. (%).

⁴⁾No. of *L. welshimeri* / No. of *Listeria* spp. (%).

Table 2. Distribution of *Listeria monocytogenes* and other *Listeria* spp. in samples from the frozen seafood processing plant

Samples	No. of samples ¹⁾	No. of isolates from samples for				Viable cell counts (CFU/mL or g)	
		<i>Listeria</i> spp.	<i>L. monocytogenes</i>	<i>L. innocua</i>	<i>L. welshimeri</i>	Range	Average
Shrimp	0 / 1	0	0	0	0	1.6×10^5	1.6×10^5
Tuna	1 / 1	6	0	6	0	2.2×10^5	2.2×10^5
Pork fat	4 / 4	24	12	10	2	$8.0 \times 10^2 \sim 7.2 \times 10^5$	1.3×10^5
Pork skin	4 / 4	16	7	8	1	$1.7 \times 10^4 \sim 1.3 \times 10^6$	1.1×10^5
Sausage product	1 / 1	1	0	1	0	8.0×10^5	8.0×10^5
Condiments	1 / 4	4	0	3	1	$5.4 \times 10^4 \sim 7.4 \times 10^5$	6.7×10^4
Bread crumb	1 / 4	5	0	5	0	$2.7 \times 10^4 \sim 6.4 \times 10^5$	3.3×10^5
After mixing	2 / 4	2	0	2	0	$1.8 \times 10^2 \sim 2.5 \times 10^3$	2.5×10^2
After molding	2 / 4	6	0	6	0	$2.7 \times 10^4 \sim 8.4 \times 10^5$	5.5×10^5
After B&B	2 / 4	8	0	6	2	$2.5 \times 10^5 \sim 1.4 \times 10^6$	8.2×10^5
After freezing	2 / 4	2	0	2	0	$7.8 \times 10^4 \sim 2.2 \times 10^5$	1.5×10^5
Wall	3 / 6	14	0	14	0	$9.3 \times 10^3 \sim 1.2 \times 10^4$	1.1×10^4
Floor	8 / 8	14	3	8	3	$4.5 \times 10^5 \sim 2.3 \times 10^6$	1.4×10^6
Worker	1 / 2	3	0	3	0	$2.5 \times 10^2 \sim 7.5 \times 10^4$	3.7×10^4
Water	1 / 4	4	0	2	2	$8.7 \times 10^4 \sim 2.7 \times 10^4$	8.7×10^6
Silent cutter	3 / 4	8	0	8	0	$2.7 \times 10^3 \sim 3.5 \times 10^4$	1.9×10^4
Board	1 / 2	9	0	9	0	$2.5 \times 10^4 \sim 6.1 \times 10^4$	4.3×10^4
Conveyor belt	0 / 2	0	0	0	0	$2.6 \times 10^3 \sim 7.1 \times 10^3$	4.5×10^3
Total	32/63	126	22/126 ²⁾ (17.5%)	93/126 ³⁾ (73.8%)	11/126 ⁴⁾ (8.5%)		

¹⁾No. of samples that were contaminated with *Listeria* spp. / No. of submitted samples.

²⁾No. of *L. monocytogenes* / No. of *Listeria* spp. (%).

³⁾No. of *L. innocua* / No. of *Listeria* spp. (%).

⁴⁾No. of *L. welshimeri* / No. of *Listeria* spp. (%).

Table 3. Distribution of *Listeria monocytogenes* and other *Listeria* spp. in samples from the *jeotgal* processing plant

Samples	No. of samples ¹⁾	No. of isolates from samples for				Viable cell counts (CFU/mL or g)	
		<i>Listeria</i> spp.	<i>L. monocytogenes</i>	<i>L. innocua</i>	<i>L. welshimeri</i>	Range	Average
Wall	1 / 4	0	0	0	0	$9.2 \times 10^5 \sim 9.8 \times 10^5$	9.4×10^5
Floor	1 / 2	15	0	12	3	$2.5 \times 10^5 \sim 5.1 \times 10^6$	2.1×10^6
Chopping board	0 / 2	2	0	2	0	$1.9 \times 10^4 \sim 3.5 \times 10^4$	2.7×10^4
Drain	0 / 2	8	0	3	5	$1.5 \times 10^6 \sim 2.0 \times 10^6$	1.7×10^6
Board	1 / 2	0	0	0	0	6.5×10^4	6.5×10^4
Water	0 / 2	0	0	0	0	$5.2 \times 10 \sim 5.6 \times 10$	5.4×10
<i>Changran</i>	2 / 2	5	0	5	0	$2.6 \times 10^6 \sim 3.0 \times 10^6$	2.8×10^6
Total	5 / 16	30	0/30 ²⁾ (0.0%)	22/30 ³⁾ (73.3%)	8/30 ⁴⁾ (26.7%)		

¹⁾No. of samples that were contaminated with *Listeria* spp. / No. of submitted samples.

²⁾No. of *L. monocytogenes* / No. of *Listeria* spp. (%).

³⁾No. of *L. innocua* / No. of *Listeria* spp. (%).

⁴⁾No. of *L. welshimeri* / No. of *Listeria* spp. (%).

monocytogenes from frozen product was presumed that the product may have been contaminated during transfer on conveyor belts which were already contaminated by *L. monocytogenes*. *L. innocua* was detected from all isolated samples. Especially, *L. innocua* was even detected from a forklift truck which is not associated with the processing procedure directly. These results suggest that *Listeria* spp. were ubiquitous in imitation crab meat processing environments.

In the frozen seafood plant, *L. monocytogenes* was detected from pork fat and pork skin. These results were similar to other research which had already reported (Farber and Peterkin, 1991) that *L. monocytogenes* was detected frequently from meat such as pork, beef and poultry. In the present study, *L. monocytogenes* was detected just from the floor, except in the case of pork fat and pork skin in the frozen seafood plant, however, *L. innocua* was detected from all isolated samples including water and workers. These results were a considerable problem for food safety in processing environments.

In the *jeotgal* plant, most of *Listeria* spp. were detected from the floor, drains and *Changran*. In the case of the *jeotgal* processing plant, the isolation rate of *L. welshimeri* was higher than those of other plants.

Among the 106 strains of *Listeria* spp. isolated from raw materials and raw products in each plant, the isolation rates of *L. monocytogenes*, *L. innocua*, and *L. welshimeri* were 34.9%, 59.4% and 5.7%,

respectively (Table 4). Thirty-seven *L. monocytogenes* were detected from pork fat, pork skin and frozen product. In the case of the 195 strains of *Listeria* spp. taken from processing environments (Table 5), the detection rate of *L. monocytogenes*, *L. innocua* and *L. welshimeri* was 30.3%, 59.5% and 10.3%, respectively. Fifty-nine *L. monocytogenes* were detected from the floor and conveyor belts. In raw materials and raw products, *L. monocytogenes* was detected from pork fat, pork skin and frozen product, however, *L. innocua* was detected from most of the isolated samples. Especially, in the case of raw materials such as tuna, *Changran*, pork fat, and pork skin, the isolation rate of *Listeria* spp. was over 90.0%. In the case of processing environments, *Listeria* spp. were detected from wide range of environments. Also, in the frozen seafood plant, the contamination of *Listeria* spp. was more wide spread than in the imitation crab meat plant. In all investigated plants, *L. monocytogenes* was detected mainly from the floor and also the isolation rate of *Listeria* spp. from the floor was over 94.4%. *L. innocua* was detected from all isolated samples and the mean value of isolation rates was 59.8%. *L. welshimeri* was detected from the floor, drains, water and a forklift truck. In the present study, *Listeria* spp. were widely distributed in all investigated seafood processing environments. Although the isolation rate of *L. monocytogenes* was the highest value in the imitation crab meat plant, the distribution of *L. innocua* was more wide spread in the frozen

Table 4. Distribution of *Listeria monocytogenes* and other *Listeria* spp. in samples from raw materials and processing products

Samples	No. of samples ¹⁾	No. of isolates from samples for				Viable cell counts (CFU/mL or g)	
		<i>Listeria</i> spp.	<i>L. monocytogenes</i>	<i>L. innocua</i>	<i>L. welshimeri</i>	Range	Average
Mixture	1 / 4	1	0	1	0	$2.1 \times 10^5 \sim 1.6 \times 10^5$	1.9×10^5
After sealing	1 / 2	2	0	2	0	$1.8 \times 10^5 \sim 2.2 \times 10^5$	2.0×10^5
Cooling	0 / 2	0	0	0	0	2.1×10^3	2.1×10^3
Before freezing	A ²⁾ 0 / 2	0	0	0	0	$2.0 \times 10^2 \sim 8.0 \times 10^2$	5.0×10^2
After freezing	1 / 2	24	18	6	0	$8.6 \times 10^4 \sim 1.1 \times 10^5$	9.8×10^4
Before disinfection	0 / 2	0	0	0	0	$2.1 \times 10^5 \sim 3.3 \times 10^5$	2.7×10^5
After disinfection	0 / 2	0	0	0	0	>30	>30
Shrimp	0 / 1	0	0	0	0	1.6×10^5	1.6×10^5
Tuna	1 / 1	6	0	6	0	2.2×10^5	2.2×10^5
Pork fat	4 / 4	24	12	10	2	$8.0 \times 10^2 \sim 7.2 \times 10^5$	1.3×10^5
Pork skin	4 / 4	16	7	8	1	$1.7 \times 10^4 \sim 1.3 \times 10^6$	1.1×10^5
Sausage product	1 / 1	1	0	1	0	8.0×10^5	8.0×10^5
Condiments	B ³⁾ 1 / 4	4	0	3	1	$5.4 \times 10^4 \sim 7.4 \times 10^5$	6.7×10^4
Bread crumb	1 / 4	5	0	5	0	$2.7 \times 10^4 \sim 6.4 \times 10^5$	3.3×10^5
After mixing	2 / 4	2	0	2	0	$1.8 \times 10^2 \sim 2.5 \times 10^3$	2.5×10^2
After mclding	2 / 4	6	0	6	0	$2.7 \times 10^4 \sim 8.4 \times 10^5$	5.5×10^5
After B&B	2 / 4	8	0	6	2	$2.5 \times 10^5 \sim 1.4 \times 10^6$	8.2×10^5
After freezing	2 / 4	2	0	2	0	$7.8 \times 10^4 \sim 2.2 \div 10^5$	1.5×10^5
Changran	C ⁴⁾ 2 / 2	5	0	5	0	$2.6 \times 10^6 \sim 3.0 \times 10^6$	2.8×10^6
Total	25/53	106	37/109 ⁵⁾ (34.9%)	63/109 ⁶⁾ (59.4%)	6/109 ⁷⁾ (5.7%)		

¹⁾ No. of samples that were contaminated with *Listeria* spp. / No. of submitted samples.

²⁾ Imitation crab meat processing plant.

³⁾ Frozen seafood processing plant.

⁴⁾ Jeotgal processing plant.

⁵⁾ No. of *L. monocytogenes* / No. of *Listeria* spp. (%).

⁶⁾ No. of *L. innocua* / No. of *Listeria* spp. (%).

⁷⁾ No. of *L. welshimeri* / No. of *Listeria* spp. (%).

seafood plant.

L. monocytogenes has been found in a wide range of foods such as dairy products, meat products, vegetables and seafood products (Farber and Peterkin, 1991). However, seafood products have received less study than other foods. Weagant et al. (1988), upon examining 57 samples of frozen seafood products, found 15 samples, including shrimp, crab meat, lobster tail, fin fish and surimi-based seafood, to be positive for *L. monocytogenes*. Very little work has been done to examine the growth of *L. monocytogenes* in seafoods (Lovett et al., 1990) and the contamination with *L. monocytogenes* in seafood processing plants (Rørvik et al., 1995; Johansson et al., 1999). In the domestic case, there have been a few studies on the distribution of *L. monocytogenes* in meat products (Hong and An, 1998), but in the

case of seafoods, there has been very little. Hong and An (1998) upon examining 234 samples of pork fabrication processing environments, found 17.5% *L. monocytogenes* and 34.2% *Listeria* spp.

In the present study, a total 301 strains of *Listeria* spp. were collected from raw materials, raw products and processing environments in three seafood processing plants. Among the samples, the detection rates of *L. monocytogenes*, *L. innocua* and *L. welshimeri* was 31.9%, 59.5% and 8.6%, respectively. *L. monocytogenes* can be found in several seafood processing environments and raw materials. However, *L. innocua* was widely distributed in seafood processing plants included workers, water and even a forklift truck. Furthermore, more focus is needed on the existence of *L. innocua*, because *L. innocua* has been reported to grow faster than *L. monocyto-*

Table 5. Distribution of *Listeria monocytogenes* and other *Listeria* spp. in samples from processing environments

Samples	No. of samples ¹⁾	No. of isolates from samples for				Viable cell counts (CFU/mL or g)	
		<i>Listeria</i> spp.	<i>L. monocytogenes</i>	<i>L. innocua</i>	<i>L. welshimeri</i>	Range	Average
Floor	8 / 8	76	37	33	6	4.5×10 ⁵ ~1.8×10 ⁸	5.5×10 ⁷
Wall	0 / 4	0	0	0	0	9.3×10 ³ ~9.4×10 ⁵	3.2×10 ⁴
Water	0 / 2	0	0	0	0	>30~5.4×10	>2.7×10
Worker	0 / 2	0	0	0	0	2.5×10 ²	2.5×10 ²
Knife	A ²⁾ 1 / 2	4	0	4	0	7.6×10 ³	7.6×10 ³
Silent cutter	2 / 4	3	0	3	0	2.7×10 ³ ~4.3×10 ⁶	3.5×10 ⁴
Forklift truck	1 / 2	10	0	9	1	5.7×10 ⁵	5.7×10 ⁵
Conveyor belt	1 / 2	25	19	6	0	7.1×10 ⁴ ~3.2×10 ⁶	1.6×10 ⁵
Pan	0 / 2	0	0	0	0	2.4×10 ³	2.4×10 ³
Wall	3 / 6	14	0	14	0	9.3×10 ³ ~1.2×10 ⁴	1.1×10 ⁴
Floor	8 / 8	14	3	8	3	4.5×10 ⁵ ~2.3×10 ⁶	1.4×10 ⁶
Worker	1 / 2	3	0	3	0	2.5×10 ² ~7.5×10 ⁴	3.7×10 ⁴
Water	B ³⁾ 1 / 4	4	0	2	2	8.7×10 ⁴ ~2.7×10 ⁴	8.7×10 ⁶
Silent cutter	3 / 4	8	0	8	0	2.7×10 ³ ~3.5×10 ⁴	1.9×10 ⁴
Board	1 / 2	9	0	9	0	2.5×10 ⁴ ~6.1×10 ⁴	4.3×10 ⁴
Conveyor belt	0 / 2	0	0	0	0	2.6×10 ³ ~7.1×10 ³	4.5×10 ³
Wall	1 / 4	0	0	0	0	9.2×10 ⁵ ~9.8×10 ⁵	9.4×10 ⁵
Floor	1 / 2	15	0	12	3	2.5×10 ⁵ ~5.1×10 ⁶	2.1×10 ⁶
Chopping board	C ⁴⁾ 0 / 2	2	0	2	0	1.9×10 ⁴ ~3.5×10 ⁴	2.7×10 ⁴
Drain	0 / 2	8	0	3	5	1.5×10 ⁶ ~2.0×10 ⁶	1.7×10 ⁶
Board	1 / 2	0	0	0	0	6.5×10 ⁴	6.5×10 ⁴
Water	0 / 2	0	0	0	0	>5.4×10	>5.4×10
Total	33/70	195	59/195 ⁵⁾ (30.3%)	116/195 ⁶⁾ (59.5%)	20/195 ⁷⁾ (10.3%)		

¹⁾ No. of samples that were contaminated with *Listeria* spp. / No. of submitted samples.

²⁾ Imitation crab meat processing plant.

³⁾ Frozen seafood processing plant.

⁴⁾ Jeotgal processing plant.

⁵⁾ No. of *L. monocytogenes* / No. of *Listeria* spp. (%).

⁶⁾ No. of *L. innocua* / No. of *Listeria* spp. (%).

⁷⁾ No. of *L. welshimeri* / No. of *Listeria* spp. (%).

genes in selective enrichment broths, which makes the detection of *L. monocytogenes* even more difficult in the presence of *L. innocua* (Curiale and Lewus, 1994; McDonald and Sutherland, 1994). Also, due to its frequent occurrence in food, *L. innocua* can be considered an indicator bacterium for the presence of *L. monocytogenes* (Jeyasekaran et al., 1996). From this point of view, distribution of *L. innocua* in seafood processing environments will be a very important point to prevent the presence of *L. monocytogenes*. Also, these results show that the most critical point of contamination by *Listeria* spp. was the floor in all investigated plants. *L. monocytogenes* was mostly isolated from the

floor and the isolation rate of *Listeria* spp. from the floor was over 94.4%. These results suggest that each plant needs to clean its floors and take care in the handling of raw materials to prevent proliferation of *Listeria* spp., especially *L. monocytogenes* in processing environments.

L. monocytogenes isolates were serotyped by commercial *Listeria* antisera (Denka Seiken, Tokyo, Japan). Among the 96 isolated *L. monocytogenes* from pork skin, pork fat, after freezing product, the floor and conveyor belts, three types of typical serotypes were detected (Table 6). The detection rates of serotype 1/2a, 1/2b, 1/2c and unknown was 59.4%, 6.2%, 12.5% and 21.9%, respectively. Nineteen *L.*

Table 6. Distribution of *L. monocytogenes* serotype in samples

Source of samples	Number of isolates	<i>L. monocytogenes</i> serotype (%)			
		1/2a	1/2b	1/2c	Unknown
Pork skin	7	4	0	2	1
Pork fat	12	0	0	10	2
After freezing	18	14	2	0	2
Conveyor belt	19	14	0	0	5
Floor	40	25	4	0	11
Total	96	57(59.4)	6(6.2)	12(12.5)	21(21.9)

monocytogenes isolated from pork skin and pork fat mainly belonged to serotype 1/2c but serotype 4b was not detected. These results were different from other research (Farber and Peterkin, 1991; Hong and An, 1998). As other research has already reported (Farber and Peterkin, 1991; Hong and An, 1998), the major serotype isolated from pork was serotype 4b. However, the presence of serotype 1/2a and 1/2b was a remarkable result although serotype 4b was not detected. Among the 77 *L. monocytogenes* isolated from frozen product, conveyor belts and the floor, the major serotype was 1/2a. Serotype 1/2b was isolated from frozen product and the floor but serotype 1/2c was not detected. Unknown serotypes were separated into three specific types by the O antigen they have (Table 7). Serotyping is not indispensable for the identification of *L. monocytogenes*, but recent data suggest that the majority of *L. monocytogenes* occurring in food and associated with human listeriosis belong to serotypes 1/2a, 1/2b and 4b (Rocourt, 1994). Therefore, serotyping is important work for the investigation of potential danger caused by this organism. In the present study, the isolates of *L. monocytogenes* mainly belong to serotype 1/2a. These results were similar to other previous research (Jemmi, 1990; Loncarevic et al.,

Table 7. Unknown serotypes of *L. monocytogenes* in samples

Serotype	O antigen present	H antigen present	
1/2a	I, II, (III)	A, B	Adapted from
1/2b	I, II, (III)	A, B, C	Seeliger and
1/2c	I, II, (III)	B, D	Höhne (1979)
Unknown type I	I, II, V	A, B, C	11
Unknown type II	I, II, V, VI	A, B, C	5
Unknown type III	V	A, B, C	2

1996; Boerlin et al., 1997; Nørrung and Skovgaard, 1993; Rørvik et al., 1997).

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