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This is an electronic version of an article published in
International Journal of Environmental Health Research, Vol. 16, No. 5, 2006
p. 375-383, available online at:
http://dx.doi.org/10.1080/09603120600869406

Ribotype diversity of *Listeria monocytogenes* isolates from two salmon processing plants in Norway

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Abstract

The purpose of this study was to use automated ribotyping procedure to track *Listeria monocytogenes* transmission in the cold smoked fish production chain and to characterize *L. monocytogenes* subtypes associated with the salmon processing industry. A total of 104 isolates, which had previously been obtained from a raw fish slaughter and processing plant (plant B) and an adjacent, downstream, salmon smoking operation (plant A), were characterized. These isolates had been obtained through a longitudinal study on *Listeria* presence, which covered a 31-week period, in both plants. Isolates had been obtained from samples taken from different machinery used throughout the production process. In addition, six isolates obtained from products produced in plant A two years after the initial study were included, so that a total of 110 isolates were characterized. Automated ribotyping was performed using both the restriction enzymes EcoRI and PvuII to increase the discriminatory power. The 110 *L. monocytogenes* isolates could be divided into 11 EcoRI ribotypes; PvuII ribotype data yielded multiple subtypes within 7 EcoRI ribotypes for a total of 21 subtypes based on both EcoRI and PvuII ribotyping. A total of three EcoRI ribotypes (DUP-1023C, DUP-1045B, and DUP-1053E) were isolated at multiple sampling times from both plants. In addition, one subtype (DUP-1053B) was isolated at multiple sampling times in only plant A, the salmon smoking operation. These data not only support that *L. monocytogenes* can persist throughout the salmon production system, but also showed that *L. monocytogenes* may be transmitted between slaughter and smoking operations or may be unique to smoking operations. While the majority of subtypes isolated have been rarely or never linked to human listeriosis cases, some subtypes have previously caused human listeriosis outbreaks and cases. Molecular subtyping thus is critical to identify *L. monocytogenes* transmission and niches in order to allow design and implementation of control strategies at the appropriate stage of production and in order to reduce the prevalence of *L. monocytogenes* linked to human disease.

Keywords: *Listeria, food safety, salmon, ribotyping, biofilm*

Introduction

Listeria monocytogenes is recognized as an important human pathogen causing food-borne outbreaks and sporadic infections. It may cause invasive disease such as bacteremia, meningitis and severe prenatal infections (Gellin & Broome 1989). The organism is

ubiquitous throughout nature and is frequently isolated from the food-processing industry (Jacquet et al. 1993; Fonnesbech et al. 2001; Hoffman et al. 2003; Thimothe et al. 2004). This bacterium is also regularly isolated from the fish processing industry in Norway (Rørvik et al. 1995; Rosef et al. 2002; Klæboe et al. 2005). Post-process contamination of food with *L. monocytogenes* represents a serious problem because of its ability to survive and grow at refrigeration temperature (Farber & Peterkin 1991). The zero tolerance rules by FDA (Chen et al. 2003) and the 100 cfu/g level tolerance in Europe (EC 2000) issued for ready-to-eat food presents a serious challenge to the food industry. Cold-smoked fish products are foods of particular concern due to the lack of a heat inactivation step during processing (Gombas et al. 2003; Thimothe et al. 2004).

Although the presence of *L. monocytogenes* has been demonstrated in many environments, our understanding of the ecology and transmission of this organism, particularly in complex food production systems, is still limited. The capacity of *Listeria monocytogenes* to adsorb to the inert surfaces found in the food-processing environment is well known (Mafu et al. 1990; Hood & Zottola 1997; Kalmokoff et al. 2001). *Listeria monocytogenes* in biofilms are much more resistant to disinfection than their free-living counterparts and thick complex biofilms are more difficult to remove than adhered single cells of the bacteria (Tompkin 2002), possibly contributing to the well documented ability of *L. monocytogenes* to persist in food processing plants over time (Lappi et al. 2004).

Studies on the epidemiology and transmission of bacterial pathogens require methods to differentiate isolates beyond the species and subspecies level (Wiedmann 2002). While a number of subtyping methods have been described for bacterial pathogens, automated ribotyping is a commonly used tool, particularly for subtyping of *L. monocytogenes* for transmission studies, since this method is highly standardized and automated, facilitating its application by industry (Bruce 1996; Wiedmann 2002). For example, a recent study on *L. monocytogenes* transmission in smoked fish plants in the United States successfully used automated ribotyping to identify persistent *L. monocytogenes* strains and their niches and transmission (Thimothe et al. 2004). In addition, a large database of EcoRI ribotypes for *L. monocytogenes* is publicly available at Pathogen Tracker (www.pathogentracker.net), allowing broad comparisons of subtype data. Because multienzyme ribotyping or other methods (e.g., PFGE) may provide greater discriminatory capabilities than single-enzyme ribotyping (Louie et al. 1996; Aarnisalo et al. 2003), we have chosen to perform ribotyping with two enzymes (EcoRI and PvuII) in this study.

Material and methods

Isolates and study plants

The majority of isolates used in this study ($n = 104$) have been randomly selected from isolates obtained as part of a previously described 31-week longitudinal study on *Listeria* presence in two seafood processing plants (Klæboe et al. 2005). The two plants these isolates had been obtained from represent a raw fish slaughter and processing plant (plant B) and an adjacent salmon smoking operation (plant A). Plant A and B are only 50 m apart; plant B processed fish, delivered fish for consumption, and provided fish for cold smoking in plant A as previously described (Klæboe et al. 2005). Seven sampling series (six series in plant A and four series in plant B) were carried out over an eight-month test period. There is a four-week interval between the series. Each series included one production week with samples taken twice daily. After series 2 in plant A and series 3 in plant B, the plants were totally cleaned by washing, disinfection and drying during a production stop for three weeks. Sampling sites in

the two plants were chosen to represent locations where the risk of cross contamination and growth of *Listeria* was high, such as sites where fish are in contact with equipment or other fish and/or areas where the humidity is high. Six sites in plant A and five in plant B were sampled. Between 25 and 50 g of sample materials, including pieces of fish meat and blended homogenous waste representing several fish, were collected from each site twice daily. Standard methods (NMKL 1999; NMKL 2004) were used for isolation; all details of *L. monocytogenes* detection and isolation have previously been described (Klæboe et al. 2005).

For plant A, isolates collected from sites before the smoking process, including the salting area, and the skinning and cold portioning machines, as well as isolates collected from sites after smoking (i.e., the skinning, slicing, and deboning machines) were characterized by subtyping. For plant B, isolates from the cleaning, filleting, deboning, sorting, and fine trimming machines were analysed. In addition to 104 *L. monocytogenes* isolates obtained during our previously reported longitudinal study (Klæboe et al. 2005), another six *L. monocytogenes* isolated from smoked salmon produced in plant A two years after the longitudinal study were also obtained and characterized. A total of 110 *L. monocytogenes* isolates were thus characterized by subtyping.

Automated ribotyping

Each isolate was characterized separately by automated ribotyping using the restriction enzymes EcoRI or PvuII. Ribotyping was performed using the DuPont Qualicon RiboPrinter as previously described (Bruce 1996). EcoRI ribotype patterns were automatically assigned a DuPont identification number (e.g., DUP-1053) by the Riboprinter®, which was confirmed by visual inspection. If this inspection indicated that a given DuPont ID included more than one distinct ribotype pattern, each pattern was designated by an alphabetically assigned letter suffix (e.g., DUP-1053B and DUP-1053E represent two distinct ribotype patterns within DuPont ID DUP-1053). Distinct ribotype patterns within a given DuPont ID generally only differed by position of a single weak band (Gray et al. 2004). EcoRI ribotype patterns were also used to classify isolates into one of the three main phylogenetic *L. monocytogenes* lineages (I, II, and III) as previously described (Wiedmann et al. 1997; Gray et al. 2004).

Results

EcoRI and PvuII ribotyping results

The 110 *L. monocytogenes* isolates could be divided into 11 EcoRI ribotypes. PvuII ribotyping allowed further differentiation of seven EcoRI ribotypes (Table I) and combined analysis of PvuII and EcoRI ribotypes differentiated a total of 21 subtypes (we will refer to these subtypes as ''combined EcoRI-PvuII subtypes''). Ribotype DUP-1023C was dominating with 50 of the 110 isolates found during the entire test period. DUP-1053E and DUP-1053B were isolated with 17 and 12 respectively (Table I). Eight of the EcoRI ribotypes (102 isolates) represented lineage II, while three ribotypes (8 isolates) represented lineage I; no isolates were classified into lineage III.

Contamination patterns

Seven EcoRI ribotypes (e.g., DUP-1023C, DUP-1053E; Table I) were isolated from both plant A and B; this finding was further confirmed by PvuII ribotyping as at least one combined EcoRI-PvuII subtype within each EcoRI ribotypes was also found in both plants

Table I. Distribution of *Listeria monocytogenes* ribotypes among 110 isolates from two salmon processing plants.

EcoRI ribotype[a]	No. of isolates	PvuII ribotype	No. of isolates	No. of isolates from Plant A	No. of isolates from Plant B
DUP-1023C	50	1	48	31	17
		2	1	1	–
		3	1	1	–
DUP-1027B (DUP-18605)	3	1	3	2	1
DUP-1038B	2	1	2	2	–
DUP-1039C	5	1	3	2	1
		2	1	–	1
		3	1	1	–
DUP-1042B	3	1	2	–	2
		2	1	–	1
DUP-1045B	8	1	7	5	2
		2	1	1	–
DUP-1053B	12	1	11	11	–
		2	1	1	–
DUP-1053C (DUP-18602)	4	1	3	2	1
		2	1	1	–
DUP-1053E	17	1	14	8	6
		2	2	1	1
		3	1	–	1
DUP-1062B	3	1	3	2	1
DUP-1062D (DUP-18596)	3	1	3	3	–

[a]EcoRI ribotype patterns were automatically assigned a DuPont identification number (e.g., DUP-1053); if visual inspection indicated that a given DuPont ID included more than one distinct ribotype pattern, each pattern was designated by an alphabetically assigned letter suffix (e.g., DUP-1053B and DUP-1053E); ribotype identification number in parenthesis represent identification numbers that were recently added to the RiboPrinter and correspond to the identification number listed first.

(e.g., the DUP-1023C – PvuII-1 subtypes represented 31 and 17 isolates from plants A and B). In addition, one EcoRI ribotype (DUP-1042B) was only found in plant B, while two EcoRI ribotypes (DUP-1053B and DUP-1062D) were only found in plant A (Table I).

The most prevalent ribotype (DUP-1023C) represented 32 of the 74 isolates from plant A and 18 of the 36 isolates from plant B. Interestingly, while the frequency of this ribotype was extremely high before the main sanitation intervention, which was conducted after sampling series 2 and 3 in plants A and B, the frequency of this ribotype was considerably reduced among isolates collected after this intervention (only 3 of 27 isolates obtained after intervention were DUP-1023C) (Tables II and III). In particular, DUP-1023C was not found among any isolates from the fish slaughtering plant (plant B) that were obtained after the main sanitation intervention, while DUP-1023C was recovered from samples collected from plant A after main sanitation intervention, including isolates collected during series 4 and 7 and isolates from smoked salmon collected two years after the initial study. Similarly, ribotype DUP-1053B, representing the third most common ribotype identified in this study, was found among isolates from plant A collected before and after the main sanitation intervention; this ribotype actually showed a higher frequency among plant A isolates after the main sanitation intervention (representing 8 of 20 isolates) as compared to before cleaning (Table II). Ribotype DUP-1045B, although overall less frequent, was isolated from both plants A and B before and after the main cleaning. Overall, the ribotype distribution changed with the main sanitation process; before the main sanitation intervention ribotypes DUP

Table II. Distribution of ribotypes among isolates ($n = 74$) from plant A.

	EcoRI ribotypes[a]						
Site of sampling	Series 1[b]	Series 2[b]	Series 4[b]	Series 5[b]	Series 6[b]	Series 7[b]	After two years[c]
Salting area	1045B		1023C 1053E 1053C				
Skinning machine (before smoking)	1023C (2) 1053E (2)			1027B	1039C	1039C	
Portion machine	1023C (2) 1045B (2) 1053E (2) 1053B 1062B 1062D (2)	1023C (4) 1053E		1039C 1053B (2)	1038B 1053B	1023C (3)	
Filleting	1023C (2)	1023C 1053E			1038B 1053B		
Skinning machine (after smoking)	1023C (3) 1053B 1053E 1062B	1023C (6) 1053E		1027B 1053B	1053B (3) 1053C	1045B	
Slice	1023C (4) 1053B	1023C (3)					
Smoked salmon							1023C 1045B (2) 1053B 1053C 1062B

[a]Number in parenthesis indicate the number of isolates with a given ribotype; [b]Series 1 – 7 represents isolates from six sampling series (each series included one production week with samples taken twice daily) that were carried out in both plants over an eight-month test period as described by Klæboe et al. 2005; [c]Isolates obtained from raw materials 2 years after series 1 through 7 were completed.

Table III. Distribution of ribotypes among isolates ($n = 36$) from plant B.

	EcoRI ribotypes[a]			
Site of Sampling	Series 2[b]	Series 3[b]	Series 5[b]	Series 6[b]
Head cutting	1053E	1023C 1039C	1042B	1045B
Filleting		1039C	1042B 1053C	
Deboning	1023C	1023C (4) 1053E (2)		
Sorting	1053E (2)	1023C (6) 1045B 1053E	1042B	
Fine trimming	1023C (3) 1053E	1023C (3) 1053E (2)	1027B	

[a]Number in parenthesis indicate the number of isolates with a given ribotype; [b]Series 2 – 6 represents isolates from four sampling series (each series included one production week with samples taken twice daily) that were carried out in both plants over an eight-month test period as described by Klæboe et al. 2005.

1023C, DUP-1053B and 1053E dominated, while a greater diversity of uncommon and apparently sporadic EcoRI ribotypes, in addition to some persistent ribotypes, was found after the main sanitation intervention.

Discussion

Listeria monocytogenes causes a rare, severe human food-borne disease, and in Norway, is responsible for 15–20 hospitalized cases annually (MSIS 2001; MSIS 2002). In order to better understand the transmission of this food-borne pathogen in cold smoked salmon, a commodity that has previously been shown to often have a high prevalence of *L. monocytogenes* contamination (FDA/FSIS 2003), we conducted subtype analyses of 110 *L. monocytogenes* that have been isolated from a raw fish slaughter and processing plant (plant B) and an adjacent, downstream, salmon smoking operation (plant A). Our data show that: (i) two enzyme (EcoRI and PvuII) ribotyping provides improved discrimination for *L. monocytogenes* over single EcoRI ribotyping, (ii) specific *L. monocytogenes* subtypes persist in both raw fish slaughter and smoking operation, representing both common and operation specific subtypes, and (iii) *L. monocytogenes* subtypes isolated represented predominantly lineage II, consistent with the proposed classification of lineage II as a more environmentally adapted lineage, as compared to lineage I, which appears to be more common among human listeriosis cases and outbreaks (Nightingale et al. 2005; Chen et al. 2006; Sauders et al. 2006).

Automated ribotyping differentiated isolates into 11 EcoRI ribotypes and 21 ribotypes when both EcoRI and PvuII ribotyping data were used. These findings are consistent with previous studies which also showed increased subtype discrimination for *L. monocytogenes* when PvuII ribotyping is performed in addition to the commonly used standard EcoRI ribotype analysis (Gendel & Ulaszek 2000; De Cesare et al. 2001). Discrimination of 21 subtypes among the isolates tested here not only supports that the subtyping approach used is appropriate to track *L. monocytogenes* contamination, but also indicates considerable *L. monocytogenes* diversity in the two-plant food production system studied, consistent with previous studies which also found considerable within-plant *L. monocytogenes* diversity (e.g., Lappi et al. 2004).

In order to better understand *L. monocytogenes* transmission from raw to finished products in the cold smoked fish production chain, *L. monocytogenes* from a raw fish slaughter and processing plant (plant B) and an adjacent, downstream, salmon smoking operation (plant A), were characterized by automated ribotyping. *L. monocytogenes* had previously been isolated from 81% and 50% of the environmental samples collected in plants A and B, respectively. In addition, *L. monocytogenes* was isolated from 6% of cold-smoked fish produced in plant A (Klæboe et al. 2005). Molecular subtyping analysis showed that a total of seven *L. monocytogenes* ribotypes, including the most common ribotype, DUP-1023C, were found in both plants. Contamination between the plants A and B can easily occur by movement of people, equipment, and raw product (from plant B to A), particularly due to the short distance between these two plants. Our findings thus support the importance of controlling traffic of equipment and people between and within plants (Tompkin et al. 1999).

Molecular subtyping analysis also revealed that a number of *L. monocytogenes* subtypes persisted in the production system studied, including two ribotypes (DUP-1023C and DUP-1053B) that were isolated from salmon products two years after the main investigation, indicating persistence for up to 2 years. These findings are consistent with a number of studies that revealed persistence of *L. monocytogenes* in processing plants (e.g., Lappi et al. 2004). While some subtypes were isolated over time in one or the other plant, other subtypes were found in both plants, potentially indicating persistence in both plants or persistence in one

plant with continuous introduction to the other plant through personnel or equipment movement or through raw materials.

Since the isolates characterized were obtained from plant A and B before and after a major sanitation and *L. monocytogenes* control effort, including strict hygienic enforcement of the zones of the production area, changing of conveyer bands on all machinery, improvement of personal hygiene, removal of waste from the floor, and reduction of external traffic to the factory to a minimum (Klæboe et al. 2005), the subtyping data also allowed us to evaluate the effect of these intervention on subtype-specific *L. monocytogenes* contamination patterns. While the previously reported prevalence data showed that the interventions did not have a significant effect on the prevalence of *L. monocytogenes* in plant B (Klæboe et al. 2005), ribotype DUP-1023C was not found among the isolates characterized that were obtained after the interventions, indicating that presence of this subtype had at least been reduced. In plant A, on the other hand, ribotypes DUP-1023C and DUP-1053B were both found among isolates obtained both before and after implementation of interventions, indicating that these subtypes had established themselves in a niche that was not eliminated, e.g., through formation of biofilms (Bremer et al. 2001; Djordjevic et al. 2002). The broad presence of the persistent subtypes supports the hypothesis that subtypes are spread throughout the plant over time by the movement of equipment, fish smoking racks and possibly employees (Tompkin et al. 1999).

Characterization of isolates into *L. monocytogenes* lineages revealed that the majority of isolates represent lineage II, consistent with the proposal that this is a more environmentally adapted lineage, which is less likely to cause human listeriosis, as compared to lineage I, which appears to be more common among human listeriosis cases and outbreaks (Nightingale et al. 2005; Chen et al. 2006; Sauders et al. 2006). Furthermore, the persistent ribotypes DUP-1023C, DUP-1053 B, and DUP-1053E have not previously linked to human listeriosis cases in the US or to human listeriosis outbreaks, as determined by a search in the PathogenTracker database (www.pathogentracker.net). The predominant presence of *L. monocytogenes* subtypes not linked to human listeriosis cases may help to explain why human listeriosis cases have rarely been linked to consumption of smoked seafoods. Importantly though, two of the ribotypes that were sporadically isolated from plant B (DUP-1042B) and plant A (DUP-1038B) represent subtypes equivalent to epidemic clone I (Kathariou 2002) which has been linked to human listeriosis outbreaks in Anjou (France, 1976), Nova Scotia (Canada, 1981), Vaud (Switzerland, 1983–1987), Los Angeles (1985) (15) and two outbreaks in Massachusetts (1979 and 1983) (Gray et al. 2004). In addition, these two ribotypes have been shown to be common among human listeriosis cases in the US (Gray et al. 2004), supporting the importance of stringent control strategies for *L. monocytogenes* in the plants studied here as well as in the fisheries sector (Ericsson et al. 1997).

Conclusion

While a number of studies have used molecular subtyping data to elucidate *L. monocytogenes* transmission and ecology in smoked fish plans (e.g., Lappi et al. 2004), inclusion of a raw fish slaughter and processing plant and an adjacent, downstream, salmon smoking operation supplied with raw materials from this fish slaughter plant provided a unique opportunity for a more system-wide, fish slaughter to finished product analysis. While *L. monocytogenes* subtypes were found to persist in both the slaughter and processing plants, at least on subtype (DUP-1053B) was unique to the processing operation and found on various food contact surfaces. This supports an emerging consensus that post processing cross contamination from environment (Farber 1991; Rørvik et al. 1995; Rørvik et al. 1997; Autio et al. 1999) rather

than contamination on incoming raw materials that survives the cold smoking process is the main source of finished product contamination. Application of sensitive molecular subtyping methods can not only help elucidate complex transmission patterns, but can also help classify *L. monocytogenes* into lineages and subgroups that may differ in their likelihood to cause human disease (Chen et al. 2006).

Acknowledgements

The study was financially supported by The Research Council of Norway.

References

Aarnisalo K, Autio T, Sjoberg AM, Lunden J, Korkeala H, Suihko ML. 2003. Typing of *Listeria monocytogenes* isolates originating from the food processing industry with automated ribotyping and pulsed-field gel electrophoresis. J Food Protect 66(2):249–255.

Autio T, Hielm S, Miettinen M, Sjoberg AM, Aarnisalo K, Bjorkroth J, Mattila-Sandholm T, Korkeala H. 1999. Sources of *Listeria monocytogenes* contamination in a cold-smoked rainbow trout processing plant detected by pulsed-field gel electrophoresis typing. Appl Environ Microbiol 65(1):150–155.

Bremer PJ, Monk I, Osborne CM. 2001. Survival of *Listeria monocytogenes* attached to stainless steel surfaces in the presence or absence of *Flavobacterium* spp. J Food Prot 64(9):1369–1376.

Bruce JL. 1996. Automated system rapidly identifies and characterized microorganisms in food. Food Technol 50(1):77–81.

Chen YH, Ross WH, Scott VN, Gombas DE. 2003. *Listeria monocytogenes*: Low levels equal low risk. J Food Prot 66(4):570–577.

Chen YH, Ross WH, Gray MJ, Wiedmann M, Whiting RC, Scott VN. 2006. Attributing risk to *Listeria monocytogenes* subgroups: Dose response in relation to genetic lineages. J Food Protect 69(2):335–344.

De Cesare A, Bruce JL, Dambaugh TR, Guerzoni ME, Wiedmann M. 2001. Automated ribotyping using different enzymes to improve discrimination of *Listeria monocytogenes* isolates, with a particular focus on serotype 4b strains. J Clin Microbiol 39(8):3002–3005.

Djordjevic D, Wiedmann M, McLandsborough LA. 2002. Microtiter plate assay for assessment of *Listeria monocytogenes* biofilm formation. Appl Environ Microbiol 68(6):2950–2958.

EC. 2000. Commission decision on control on *Listeria monocytogenes* for certain categories of ready-to-eat food of animal origin. In: SANCO 594/2000 Rev 2. 2000.

Ericsson H, Eklow A, Danielsson-Tham ML, Loncarevic S, Mentzing LO, Persson I, Unnerstad H, Tham W. 1997. An outbreak of listeriosis suspected to have been caused by rainbow trout. J Clin Microbiol 35(11): 2904–2907.

Farber JM. 1991. *Listeria monocytogenes* in fish products. J Food Protect 54(12):922–924.

Farber JM, Peterkin PI. 1991. *Listeria monocytogenes*, a food-borne pathogen. Microbiol Rev 55(3):476–511.

FDA/FSIS. 2003. Quantitative assessment of the relative risk to public health from foodborne *Listeria monocytogenes* among selected categories of ready-to-eat foods. In: US Food and Drug Administration/Food Safety and Inspection (USDA).

Fonnesbech VB, Huss HH, Ojeniyi B, Ahrens P, Gram L. 2001. Elucidation of *Listeria monocytogenes* contamination routes in cold-smoked salmon processing plants detected by DNA-based typing methods. Appl Environ Microbiol 67(6):2586–2595.

Gellin BG, Broome CV. 1989. Listeriosis. Jama-J Am Med Assoc 261(9):1313–1320.

Gendel SM, Ulaszek J. 2000. Ribotype analysis of strain distribution in *Listeria monocytogenes*. J Food Protect 63(2):179–185.

Gombas DE, Chen Y, Clavero RS, Scott VN. 2003. Survey of *Listeria monocytogenes* in ready-to-eat foods. J Food Protect 66(4):559–569.

Gray MJ, Zadoks RN, Fortes ED, Dogan B, Cai S, Chen Y, Scott VN, Gombas DE, Boor KJ, Wiedmann M. 2004. *Listeria monocytogenes* isolates from foods and humans form distinct but overlapping populations. Appl Environ Microbiol 70(10):5833–5841.

Hoffman AD, Gall KL, Norton DM, Wiedmann M. 2003. *Listeria monocytogenes* contamination patterns for the smoked fish processing environment and for raw fish. J Food Protect 66(1):52–60.

Hood SK, Zottola EA. 1997. Adherence to stainless steel by foodborne microorganisms during growth in model food systems. Int J Food Microbiol 37(2–3):145–153.

Jacquet C, Rocourt J, Reynaud A. 1993. Study of *Listeria monocytogenes* contamination in a dairy plant and characterization of the strains isolated. Int J Food Microbiol 20(1):13–22.

Kalmokoff ML, Austin JW, Wan XD, Sanders G, Banerjee S, Farber JM. 2001. Adsorption, attachment and biofilm formation among isolates of *Listeria monocytogenes* using model conditions. J Appl Microbiol 91(4):725–734.

Kathariou S. 2002. *Listeria monocytogenes* virulence and pathogenicity, a food safety perspective. J Food Protect 65(11):1811–1829.

Klæboe H, Rosef O, Sæbø M. 2005. Longitudinal studies on *Listeria monocytogenes* and other Listeria species in two salmon processing plants. Int J Environ Health Res 15(2):71–77.

Lappi VR, Thimothe J, Nightingale KK, Gall K, Scott VN, Wiedmann M. 2004. Longitudinal studies on Listeria in smoked fish plants: Impact of intervention strategies on contamination patterns. J Food Protect 67(11):2500–2514.

Louie M, Jayaratne P, Luchsinger I, Devenish J, Yao J, Schlech W, Simor A. 1996. Comparison of ribotyping, arbitrarily primed PCR, and pulsed-field gel electrophoresis for molecular typing of *Listeria monocytogenes*. J Clin Microbiol 34(1):15–19.

Mafu AA, Roy D, Goulet J, Magny P. 1990. Attachment of *Listeria monocytogenes* to stainless-steel, glass, polypropylene, and rubber surfaces after short contact times. J Food Protect 53(9):742–746.

MSIS: Meldesystem for smittsomme sykdommer (MSIS Year report for infectious diseases). 2001. In: Oslo, Norway: Statens institutt for folkehelse (The National Institute for Public Health).

MSIS: Meldesystem for smittsomme sykdommer (MSIS Year report for infectious diseases). 2002. In: Statens institutt for folkehelse (The National Institute for Public Health).

Nightingale KK, Windham K, Wiedmann M. 2005. Evolution and molecular phylogeny of *Listeria monocytogenes* isolated from human and animal listeriosis cases and foods. J Bacteriol 187(16):5537–5551.

NMKL: Listeria monocytogenes, detection in foods. 1999. In: Nordisk metodekomite for næringsmidler (Nordic Commitee on Food Analysis). Veterinætinstituttet. Oslo, Norway.

NMKL: Listeria monocytogenes, detection in foods. 2004. In: Nordisk metodekomite for næringsmidler (Nordic Commitee on Food Analysis).

Rosef O, Djupvik J, Kolberg M. 2002. Listeria bakterier i to eksportslakterier for laks. Listeria in salmon slaughterhouses. NorVetTidsskr 114:293–296.

Rørvik LM, Caugant DA, Yndestad M. 1995. Contamination pattern of *Listeria monocytogenes* and other Listeria spp. in a salmon slaughterhouse and smoked salmon processing plant. Int J Food Microbiol 25(1):19–27.

Rørvik LM, Skjerve E, Knudsen BR, Yndestad M. 1997. Risk factors for contamination of smoked salmon with *Listeria monocytogenes* during processing. Int J Food Microbiol 37(2–3):215–219.

Sauders BD, Durak MZ, Fortes E, Windham K, Schukken Y, Lembo AJ Jr, Akey B, Nightingale KK, Wiedmann M. 2006. Molecular characterization of *Listeria monocytogenes* from natural and urban environments. J Food Protect 69(1):93–105.

Thimothe J, Nightingale KK, Gall K, Scott VN, Wiedmann M. 2004. Tracking of *Listeria monocytogenes* in smoked fish processing plants. J Food Protect 67(2):328–341.

Tompkin RB. 2002. Control of *Listeria monocytogenes* in the food-processing environment. J Food Protect 65(4): 709–725.

Tompkin RB, Scott VN, Bernard DT, Sveum WH, Gombas KS. 1999. Guidelines to prevent post-processing contamination from *Listeria monocytogenes*. Dairy Food Environ Sanit 19:551–562.

Wiedmann M. 2002. Molecular subtyping methods for *Listeria monocytogenes*. J AOAC Int 85(2):524–531.

Wiedmann M, Bruce JL, Keating C, Johnson AE, McDonough PL, Batt CA. 1997. Ribotypes and virulence gene polymorphisms suggest three distinct *Listeria monocytogenes* lineages with differences in pathogenic potential. Infect Immun 65(7):2707–2716.