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A survey of ready-to-eat hot and cold smoked salmon available at retail in New Zealand

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A Survey of Ready to Eat Hot and Cold Smoked Salmon Available at Retail in New Zealand

Between January and December 2010, 1,202 samples of packaged ready-to-eat smoked fish were sampled from retail outlets or directly from the processor including telephone, on-line or direct sale. The samples were selected according to the different food regulatory regime that they were processed under and market share. In total, 1,212 ready-to-eat smoked salmon samples were sampled from 19 operators, of which 598 were cold and 614 hot ready-to-eat smoked salmon.

The ready-to-eat smoked fish were analysed at the end of the processor's stated shelf life for the incidence and concentration of *L. monocytogenes*. *L. monocytogenes* was detected in a total of 8/1,212 samples of ready-to-eat smoked fish analysed (0.7%; 95% confidence interval: 0.3-1.3%). This can be further broken down; *L. monocytogenes* not detected in any of the hot ready-to-eat smoked fish samples (0/614 positive; 95% confidence interval: 0.0-0.6%) and *L. monocytogenes* was detected in 8/598 (1.3% positive; 95% confidence interval: 0.6-1.6%). Three of these samples exceeded the microbiological limits (>100 cfu/g) for processed finfish in the Food Standards Code, Standard 1.6.1 and the appropriate follow-up action was taken.

This survey is the largest microbiological survey of ready-to-eat smoked fish in New Zealand, and is indicative of lower levels of *L. monocytogenes* in New Zealand produced smoked salmon than in similar surveys conducted internationally.

Client Report FW11035

A survey of ready-to-eat hot and cold smoked salmon available at retail in New Zealand

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SUM	MARY		II
1	INTRO	DUCTION	4
2	MATEF	RIALS AND METHODS	7
2.1 2.2 2.3		Sample collection Analytical methodology Data analysis	7
3	RESUL	TS AND DISCUSSION	8
3.1 3.2 3.3		Prevalence apportioned to regulatory regime Quantitative data Comparison with international data	10
4	CONCL	LUSIONS	12
5	REFER	ENCES	13
APPF	ENDIX 1	Regulatory regime, producer, type and number of smo samples tested, and numbers positive for <i>L. monocytog</i>	

TABLE OF CONTENTS

INDEX OF TABLES

Table 1.	Recent international prevalence studies of <i>L. monocytogenes</i> on smoked salmon
Table 2.	Summary of smoked salmon producers, regulatory regime and sampling schedule
Table 3.	Prevalence of <i>Listeria monocytogenes</i> in 25 g smoked salmon samples9
Table 4.	L. monocytogenes enumeration results for smoked salmon samples10
Table 5.	Distribution of <i>L. monocytogenes</i> positive cold smoked salmon samples by product cut and regulatory regime, combination of Tranche 1 and 2 results

SUMMARY

To inform the strategy launched by the Ministry of Agriculture and Forestry (MAF) to reduce the levels of foodborne illness in New Zealand, an understanding of the exposure to various pathogens from different food groups is required. MAF recognises the significance of human listeriosis, especially for vulnerable consumers in New Zealand and the contribution that food and, in particular, ready-to-eat foods that are stored and chilled, make to this. MAF has therefore recognised and prioritised the need to minimise disease attributed to *Listeria monocytogenes* with a stated performance target outlined in the *Listeria monocytogenes* Risk Management Strategy 2008 – 2013, of "no increase in reported incidence of foodborne listeriosis after five years".

Ready-to-eat seafood has been associated with outbreaks of foodborne illness from *L. monocytogenes* infections. This microorganism is a significant hazard, especially in cold smoked seafood where there is no physical microbial kill step, and has been detected in both hot and cold smoked product at manufacture and at retail (Chitlapilly Dass *et al.*, 2010; Jørgensen and Huss, 1998). MAF therefore requested a point-in-time quantitative microbiological survey of *L. monocytogenes* in ready-to-eat (RTE) hot and cold smoked seafood at retail in New Zealand.

Seafood operators in New Zealand must currently operate either under the Animal Products Act (APA) 1999 and, as part of their Risk Management Programme (RMP) implement a *Listeria* monitoring programme, or conform with the Food Act 1981. This has two regulatory arms, the Food Hygiene Regulations (FHR) 1974 and Food Safety Programmes. All producers must comply with the Microbiological Limits for Food contained in Standard 1.6.1 of the Australia New Zealand Food Standards Code.

The objective of this project was to identify any differences in the effectiveness of *L. monocytogenes* control between RTE hot and cold smoked salmon producers operating under the APA (1999) and the Food Act 1981, as reflected in compliance with the Food Standards Code (FSC) 1.6.1.

Representative sampling of RTE smoked salmon products manufactured by food businesses operating under the APA (1999) or the Food Act regime (FHR and FSP) was performed over a 12 month period in two Tranches, obtaining approximately 200 samples from each regulatory regime and type of process (hot and cold smoking); in total 1212 RTE smoked salmon samples were analysed. Initial analysis was for the presence or absence of *L. monocytogenes* in a 25g sample. If *L. monocytogenes* was detected, enumeration was performed and positive samples confirmed by conventional biochemical assays, gene typed by Pulsed Field Gel Electrophoresis (PFGE) and serotyping.

Smoked salmon samples from 19 producers were analysed; encompassing 18 hot smoking facilities and 12 cold smoking facilities (some manufacturers produce both cold and hot smoked salmon and have multiple brands). There were 15 manufacturers operating under the Food Act 1981; 10 complying with the FHR 1974, and six with FSPs (one manufacturer changed programme from FHR to FSP after Tranche 1). Four processors operated under the APA (1999) having RMPs. These facilities also represented all regions throughout New Zealand.

There were eight samples positive for *L. monocytogenes* (0.7%), all of which were cold smoked salmon. Four were obtained from premises operating under the Food Act (1981) and the FHR (1974), three premises operating under FSPs and one under the APA (1999). However, calculated confidence intervals indicate that there is overlap in the probability of detecting similar numbers of positive samples between these regimes, suggesting a lack of statistical significance in the differences detected between the prevalences recorded for the three regimes.

All samples that were positive for *L. monocytogenes* were re-sampled for enumeration. Only three samples (all from one producer, FHR regime) gave counts greater than the lower limit of detection (5.0×10^0) , giving concentrations of 4.0×10^3 , 9.5×10^4 and 3.6×10^4 *L. monocytogenes* g⁻¹. The Food Standards Code 1.6.1 allows for 1 sample in 5 to have a maximum number of 1.0×10^2 *L. monocytogenes* g⁻¹; and although five samples were not obtained from each producer for each type of product, the concentrations in three of the samples were above the maximum allowable limit. This producer changed from a FHR programme to a FSP, and when subsequently tested in Tranche 2, the two samples tested were negative.

This survey is the largest of its kind performed in New Zealand (n=1212), with a *L. monocytogenes* incidence rate of 0.7% (8/1212 total samples). Previous studies investigating the presence of *L. monocytogenes* in New Zealand smoked salmon were small, and while having a much higher incidence (8/12 and 10/16), the numbers examined do not allow for accurate comparisons to be made. No regulatory regime could be defined as being better than another at controlling *L. monocytogenes* contamination.

1 INTRODUCTION

Listeria monocytogenes is a cold tolerant microaerophilic ubiquitous micro-organism, found commonly in soil, water and on plant material. Ingestion of this microorganism may result in invasive listeriosis, with symptoms including meningitis, septicaemia, endocarditis, encephalitis, conjunctivitis and flu-like illness, often resulting in hospitalisation, and fatality rates approximately 20-30% (WHO, 2004). Listeriosis remains an uncommon infection in New Zealand. In 2010 the incidence rate was 0.5 per 100 000 population (ESR, 2011), with a case fatality rate of 30.4%. Deaths nearly always occur in at-risk individuals, e.g. immunocompromised, pregnant women, neonates and those over 60 years of age.

The World Health Organisation has concluded that the primary route of transmission to humans is via foods contaminated during production (WHO, 1988). Epidemiological investigations are in accord with this conclusion, showing that during the last 20 years, both epidemic and sporadic cases of listeriosis are primarily foodborne.

One food group of concern is ready-to-eat smoked fish. This food is produced by salting, smoking, trimming, or slicing the fish, and then vacuum-packaging the final product. The fish or fillets are smoked in a smoke chamber at 20 to 30°C for 2 to 4 d (cold smoking) or at >60°C for 6 to 10 h (hot smoking). Listeria monocytogenes is capable of growth on smoked salmon under refrigeration temperatures (Guyer and Jemmi, 1991; Hudson and Mott, 1993; Hwang, 2007; Midelet-Bourdin et al., 2010), as well as proliferating rapidly at cold smoking chamber temperatures (Junttila et al., 1988; Seeliger and Jones, 1986) therefore the major control measure against L. monocytogenes are the addition of salt and phenolic compounds (Cornu et al., 2006). Cold smoking processes do not offer sufficient listericidal capacity to render the product safe if initially contaminated (Hwang, 2007; Jemmi and Keusch, 1992; Poysky et al., 1997). Reports on the listericidal effect of the hot smoking process are mixed, and it appears to depend on the correct combination of time, temperature, salt content and smoke ingredients (Jemmi and Keusch, 1992; Poysky et al., 1997; Rørvik, 2000). However, the processing environment itself has been shown to be a source of contamination (Chitlapilly Dass et al., 2010; Dauphin et al., 2001; Di Pinto et al., 2010; Gudmundsdóttir et al., 2005; Hoffman et al., 2003; Hu et al., 2006; Vogel et al., 2001). This indicates that both hot and cold smoked salmon are RTE foods that have the potential for post processing contamination, and offer conditions that will support high growth, therefore adhering to Good Manufacturing Practice is an important step in minimising contamination risks.

There have been many studies internationally on the incidence of *L. monocytogenes* in smoked salmon, giving prevalence rates from 0 to 100%, although most report prevalences between 0 and 50% (Table 1).

Two studies investigating the prevalence of *L. monocytogenes* in smoked salmon produced in New Zealand have been reported to the authors' knowledge. The first by Hudson *et al* (1992) found 75% (34.9% - 90.1%: 95% CI) of smoked salmon samples (8 of 12 tested) contained *L. monocytogenes*, while the second study performed by Fletcher *et al* (1994) found 0 of 3 hot smoked salmon, and 10 of 13 (77%) (46.2% - 5.0%: 95% CI) of cold smoked salmon positive for *L. monocytogenes*, giving a combined (hot and cold smoked salmon) positive rate of 62.5% (35.4% - 84.8%; 95% CI). Despite this seemingly high prevalence, there are no documented cases of listeriosis following from smoked salmon consumption in New Zealand; however smoked trout and other fish products such as mussels have caused listeriosis, both in New Zealand (Brett *et al.*, 1998) and overseas (Miettinen *et al.*, 1999; Mitchell, 1991).

500000	-			
Year	Country	Sample Description	No. +ve /	Reference
			No. samples	
2001	France	Cold smoked salmon Plant 1	2/21	(Dauphin et al.,
		Cold smoked salmon Plant 2	1/2	2001)
		Cold smoked salmon Plant 3	11/11	_
			14/34	
2004	Spain	Smoked salmon	28/100	(Vitas and Garcia-
				Jalon, 2004)
2006	Austria	Cold smoked salmon	27/202	(Suppin et al.,
				2006)
2006	USA	Cold smoked salmon	28/300	(Hu et al., 2006)
2007	France	Cold smoked salmon Plant 1	7/78	(Beaufort et al.,
		Cold smoked salmon Plant 2	6/78	2007)
		Cold smoked salmon Plant 3	2/86	
		Cold smoked salmon Plant 4	2/86	
		Cold smoked salmon Plant 5	0/82	
		Cold smoked salmon Plant 6	0/86	
		Cold smoked salmon Plant 7	16/66	
		Cold smoked salmon Plant 8	8/64	
			41/626	-
2007	Italy	Smoked salmon	11/104	(Latorre <i>et al.</i> , 2007)
2008	Spain	Smoked salmon	7/89	(Cabedo <i>et al.</i> , 2008)
2010	Italy	Smoked salmon	45/132	(Pinto et al., 2010)
2010	Italy	Smoked salmon	0/19	(Pesavento <i>et al.</i> , 2010)
2011	Republic of Ireland	Smoked salmon	26/120	(Chitlapilly Dass <i>et al.</i> , 2011)

Table 1.	Recent international prevalence studies of <i>L. monocytogenes</i> on smoked
salmon	

In New Zealand, hot and cold smoked salmon producers must operate either under the Animal Products Act (APA) 1999 or the Food Act 1981. The APA 1999 requires the producer to adhere to a Risk Management Programme (RMP), while the Food Act 1981 is regulated by the Food Hygiene Regulations (FHR) 1974. In 1997, the Food Act was modified to facilitate a voluntary transition from compliance with the FHR to the adoption by the food industry of Food Safety Programmes (FSPs). Whichever regulatory regime a producer subscribes to, they must also comply with Standard 1.6.1 of the Australia New Zealand Food Standards Code (FSC). This standard states that for ready to eat processed finfish, 1 sample in a batch of 5 (minimum number of sample units to be tested) may contain no more than $1.0 \times 10^2 L$. monocytogenes g⁻¹.

5

This study was performed to determine the prevalence and level of *L. monocytogenes* in New Zealand produced RTE hot and cold smoked salmon and to determine if *L. monocytogenes* concentrations, and compliance with the FSC 1.6.1, differed depending upon which regulatory regime the producer operated under.

2 MATERIALS AND METHODS

2.1 Sample collection

Five hundred and ninety eight cold ready to eat smoked salmon packs, and six hundred and fourteen hot smoked ready to eat salmon samples were purchased between January and December 2010. Samples were purchased from retail outlets, or directly from processors; either by on-line purchase, in person or by telephone. The number of samples analysed for each processor was chosen based on market share, how many processors in each regulatory regime and the availability of samples. A breakdown of processors operating under different food safety regimes and numbers of samples tested from each processor is detailed in Appendix 1 and summarised in Table 2. For retail samples, individual packs ranging between 50 - 200 g were purchased and were immediately placed in cold storage for transportation back to the laboratory. Packs purchased on-line were sent to ESR by the packaging and chilling arrangement the producer normally uses. Temperatures of each sample were recorded on arrival at the laboratory. Samples were then grouped according to use by date and stored in a 4°C chiller until sampling. All samples were tested within (±) two days of the manufacturer's stated use-by-date, or at an agreed use-by-date with MAF if the use-by-date was not stated by the manufacturer on the packaging.

Regulatory Regime		Sam	ples Tested	
regulatory regime	Processor	Tranche 1	Tranche 2	Total
Food Act - FHR	А	27		27
	В	11	0	11
	С	2	0	2
	D	55	61	116
	E	48	62	110
	F	16	17	33
	G	7	9	16
	Н	15	18	33
	Ι	19	19	38
	J	0	16	16
Food Act - FSP	Κ	47	47	94
	А		8	8
	L	54	60	114
	Μ	42	36	78
	Ν	18	16	34
	0	39	30	69
Animal Products Act	Р	10	10	20
	Q	0	1	1
	R	99	100	199
	S	96	97	193
Totals		605	607	1212

Table 2.Summary of smoked salmon producers, regulatory regime andsampling schedule

2.2 Analytical methodology

The outside surfaces of the smoked salmon packs were sanitised by wiping with 70% alcohol. Contents were sampled aseptically through an excised window on the plastic wrap. A 25 g sample was placed in a sterile stomacher bag and 225 mL half Fraser Broth

7

(Fort Richard, Auckland, NZ) was added and stomached for 2 min before incubating at 30° C for 24 h ± 2 h. Remaining samples were placed into labelled sterile bags and stored at 4°C for later enumeration if the sample was positive for *L. monocytogenes*. Following enrichment, 0.1 mL was plated onto RAPID'*L. mono* agar (Bio-Rad, Steenvoorde, France) and incubated at 37°C for 24 h ± 2 h. Plates were examined for typical *L. monocytogenes* colony morphology (blue), and further identified by re-streaking the presumptive positive colony onto Trypticase soy agar with 0.6% yeast extract (TSAYE), incubating overnight and performing a CAMP test (Christie *et al.*, 1944).

Samples confirmed to be positive for *L. monocytogenes* were further quantified for CFU g⁻¹. From the stored sample (approximately 2 days at 4°C), a 25 g sample was aseptically prepared in a sterile stomacher bag and 225 mL buffered peptone water added before stomaching for 2 min. This initial (10⁻¹) suspension was allowed to stand at room temperature for 1 h ± 5 min to resuscitate stressed microorganisms, before serial dilutions were made to 10⁻³ and volumes plated onto Agar Listeria Ottavani & Agosti (ALOA; Fort Richard, Auckland, NZ), and incubated at 35°C for 24 – 48 h. Typical *Listeria* spp. colonies (blue/green colonies with opaque halo) were counted and a full confirmation of up to five colonies per plate was performed by restreaking onto TSAYE agar, incubating at 35°C for 18 h to 24 h or until growth was satisfactory. From well separated colonies on TSAYE agar, catalase, Gram stain, haemolysis, carbohydrate utilisation, CAMP test and motility tests were performed to confirm the presence of *L. monocytogenes*. Purified colonies were also genotyped by PFGE and serotyped on the basis of somatic (O) antigens. If no growth was observed on ALOA plates, results were recorded as <5 CFU g⁻¹.

2.3 Data analysis

Data were analysed to determine if the smoked salmon products were in compliance with Food Standards Code 1.6.1, which states that for ready to eat processed finfish, 1 sample in a batch of 5 (minimum number of sample units) may contain no more than 1.0×10^2 *L. monocytogenes* g⁻¹. Although smoked salmon samples were not obtained in batches of 5 for this survey, the guidelines in FSC 1.6.1 were still applied. 95% confidence intervals around the prevalence values for the proportion of smoked salmon products likely to be contaminated given the samples analysed were calculated using the method for calculating confidence limits for population proportions given in Zar (1999).

3 RESULTS AND DISCUSSION

1212 cold and hot smoked salmon samples from 19 New Zealand producers, operating under the Food Act 1981, or the APA 1999, were purchased over a 12 month period, and analysed for the presence of *L. monocytogenes*. The number of samples analysed for each processor was chosen based on market share, how many processors in each regulatory regime and the availability of samples. Those samples found to be positive were further enumerated and confirmed. ESR has provided MAF with a spreadsheet containing the raw data for this survey. Summaries of these data for each regulatory regime are shown in Appendix 1. This spreadsheet was used in the analysis that follows. All presumptive *L. monocytogenes* results were confirmed by conventional assays, PFGE and serotyping.

3.1 Prevalence apportioned to regulatory regime

The *L. monocytogenes* isolation data (Table 3) show a small range of prevalence values, from 0% in all hot smoked salmon samples, irrespective of regulatory regime the producer

8

subscribed to, to 2.0% in cold smoked salmon produced under the Food Hygiene Regulations (FHR) 1974 of the Food Act 1981. However, the confidence intervals (CI) given for the positive samples obtained under the Food Safety Programme (FSP), as well as those operating under a Risk Management Programme (RMP) as required by the APA 1999, are 0.3 to 4.4 and 0 to 2.8, giving an overlap despite the very low numbers, indicating that there is no statistical difference between the regimes.

There were five out of 19 operators whose products gave at least one positive result; two operating under FHR 1974 / Food Act 1981, two under FSP / Food Act 1981, and one under a RMP / APA 1999.

Regulatory Regime / Programme	Hot or cold smoked	n	No. (%) +ve	95% Confidence Intervals (%)
Food Act 1981	Cold	201	4 (2.0)	0.5-5.0
/ FHR (1974)	Hot	201	0	0.0-1.8
			4 (1.0)	0.3-2.5 (overall)
Food Act 1981	Cold	197	3 (1.5)	0.3-4.4
/ FSP	Hot	200	0	0.0-1.8
			3 (0.8)	0.2-2.2 (overall)
Animal	Cold	200	1 (0.50)	0.0-2.8
products Act 1999 / RMP	Hot	213	0	0.0-1.7
			1 (0.2)	0.0-1.3 (overall)
Total		1212	8 (0.7)	0.3-1.3

 Table 3.
 Prevalence of Listeria monocytogenes in 25 g smoked salmon samples

Note: All isolates tested were confirmed as *L. monocytogenes* by biochemical assays, PFGE and serotyping.

The initial agreement was to analyse 100 samples of cold and hot smoked salmon from each regulatory regime, per tranche (200 samples from each category, 1200 samples in total). However, due to unexpected circumstances (Canterbury earthquake, 04/09/2010), only 197 cold smoked salmon samples originating from premises with a FSP (under the Food Act 1981) were tested. Because the probability of a positive sample is so low it is not anticipated that the three missing samples would alter the overall conclusions. If the three missing samples had tested positive, the prevalence would increase to 1.7% (95% CI: 0.8-3.0) in cold smoked salmon samples and 0.9% (95% CI: 0.5-1.6) overall.

The Risk Management and Food Safety Programmes offer alternative risk-based approaches to food safety and to hazard control, as opposed to the specific requirements and measures outlined in the FHR (1974). Despite the introduction of FSPs in 1997, intended to supersede the FHR (1974) by voluntary transition, there are many smoked salmon producers still operating under the FHR regime in New Zealand.

3.2 Quantitative data

In this study, the presence or absence of *L. monocytogenes* on the salmon samples was first determined, and if present, enumeration was undertaken by plating serial dilutions onto a selective agar. Results are shown in Table 4.

Positive Sample	Tranche	Confirmed by CAMP	CFU g ⁻¹	Confirmation of 5 colonies	PFGE Type	Somatic Serotype
		test				
1	1	Y	4.0×10^{3}	Y	Asc0095/Apa0085	1/2
2	1	Y	9.5×10^4	Y	Asc0095/Apa0085	1/2
3	1	Y	3.6×10 ⁴	Y	Asc0095/Apa0085	1/2
4	1	Y	$< 5.0 \times 10^{0}$	Y	Asc0002/Apa0002	1/2
5	1	Y	$< 5.0 \times 10^{0}$	Y	Asc0048/Apa0041	1/2
6	1	Y	$< 5.0 \times 10^{0}$	Y	Asc0096/Apa0086	Untypable
7	2	Y	$< 5.0 \times 10^{0}$	Y	Asc0045/Apa0013	4
8	2	Y	$< 5.0 \times 10^{0}$	Y	Asc0035/Apa0027	1/2

 Table 4.
 L. monocytogenes enumeration results for smoked salmon samples

Grey shaded area = previously seen clinical isolates

One smoked salmon producer (A) produced three of the eight positive samples, those with countable *L. monocytogenes*, all from Tranche 1 (samples 1-3). Although only individual packs from each producer were sampled as per the agreed sampling plan, and not five as outlined in FSC 1.6.1, the concentrations obtained were above the acceptable limit. MAF were notified and action was taken to decontaminate the plant. Production was therefore temporarily halted and the regulatory regime changed from FHR to FSP within the Food Act. Due to the temporary closure of the processing plant, only two samples were able to be included in Tranche 2, however, both tested negative for *L. monocytogenes*.

The three positive *L. monocytogenes* samples from Producer A all had identical PFGE Types and serotype (Table 4). However, the samples could not be distinguished by lot or batch number from their packaging, and the use-by date was identical for all three, despite being purchased from two different locations, over three dates. Therefore the source of contamination can not be determined to be either from the fish itself, or from a single source of contamination within the process/premises.

The PFGE types found in this study were compared to the PFGE databank held by ESR. Four isolates (1, 2, 3 and 6) had unique pulsotypes not correlating with any other isolates in the database, while samples 4, 5, 7 and 8 were common with previous human (clinical) isolates; isolates 4 and 8 were also pulsotypes isolated previously from seafood.

Cold smoked salmon had a higher prevalence of *L. monocytogenes* contamination than hot smoked salmon (1.3% positive; 95% CI: 0.6-1.6 vs 0% positive; 95% CI: 0.0-0.6 respectively). This may indicate that hot smoking process does offer additional listericidal properties to render the product safe if initially contaminated.

The cold smoked salmon analysed by the cut type is shown in Table 5. Positive samples were observed across three of the four types of cuts and across the regulatory regimes.

	Type of Cut					
Regulatory	Pieces	Portions	Shavings	Slices		
Regime						
Food Act 1981 /	-	3/42	-	1/159		
FHR (1974)						
Food Act 1981 /	0/12	1/36	-	2/149		
FSP						
Animal	0/31	-	1/26	0/143		
products Act						
1999 / RMP						
Total	0/43	4/78	1/26	3/451		

Table 5.Distribution of L. monocytogenes positive cold smoked salmon samplesby product cut and regulatory regime, combination of Tranche 1 and 2 results

Salmon products from five producers returned positive *L. monocytogenes* results. Two of the five producers where facilities manufactured salmon only products were positive; two of the ten facilities where additional seafood products were processed were positive, and one of the four processors where additional species including poultry, red meats and vegetables were processed was positive. The additional products processed at these facilities were not tested for *L. monocytogenes* contamination; therefore it is unknown whether the additional foods processed in these environments contributed to the positive results found in this survey.

3.3 Comparison with international data

Surveys and studies performed internationally since 2000, have given a large range of prevalence of *L. monocytogenes* in cold smoked salmon (no studies were specified to be on hot smoked salmon), ranging from 0% (Pesavento *et al.*, 2010) to 100% (Dauphin *et al.*, 2001). Table 1 outlines recent surveys, countries and prevalence rates.

Compared to the recent surveys performed overseas this survey represents a dramatically lower incidence rate of *L. monocytogenes* in smoked salmon samples produced in New Zealand than generally found internationally (Table 1), providing an indication of the overall successful performance of this sector. Increased industry awareness of the potential risks of *L. monocytogenes* in RTE foods as well as efficient recall, decontamination and re-testing systems may also be a factor in the very low prevalence rate found.

This study is the first in New Zealand that has investigated the prevalence of *L. monocytogenes* in hot smoked salmon. With a 0% (95% CI: 0.0-0.6) prevalence rate, this could intimate that the hot smoking process as carried out in New Zealand, combined with the stringent regulatory requirements observed by these processors, may be sufficient to effectively manage *L. monocytogenes* contamination of this product.

4 CONCLUSIONS

Contamination of RTE hot or cold smoked salmon by *L. monocytogenes* was detected in a total of eight of 1212 samples tested (0.7%; 95% CI: 0.3-1.3). This figure can be broken down into hot smoked samples (0% positive; 95% CI: 0.0-0.6)) and cold smoked salmon (1.3% positive; 95% CI: 0.6-1.6). The cold smoked salmon producers can be further differentiated by the regulatory regime they operate under. Those samples obtained from premises operating under the Food Act 1981, complying with FHR (1974) resulted in a 2% positive rate, while those with FSPs had 1.5%, and those samples from producers operating under the APA (1999) with RMPs had the lowest incidence rate of 0.5%. The confidence intervals were such that there is no statistical difference between the results obtained from each of the regulatory regimes studied.

Three samples of the eight positive for *L. monocytogenes* had concentrations higher than that allowed under the FSC 1.6.1. These samples were all from one premises, which was informed and decontaminated. In addition, the premises changed the regulatory regime under which it operates from the FHR (1974) to an FSP under the Food Act (1981). Operating an FSP obliges the operator to think about, clearly understand and document the hazard controls necessary for its products. Following the change, product was re-sampled in Tranche 2 and tested negative. However, while this is a good result, the small number of samples taken are insufficient to attribute any improvement to the change in regulatory regime.

This study is the largest of its kind to be performed in New Zealand, and indicates that the level of *L. monocytogenes* contamination in ready to eat smoked salmon is significantly lower than found in similar studies internationally. Very limited surveys undertaken in New Zealand during the 1990s indicated prevalence rates of up to 75%, however the small sample sizes do not allow comparisons to be drawn.

5 **REFERENCES**

Beaufort A, Rudelle S, Gnanou-Besse N et al. (2007) Prevalence and growth of *Listeria monocytogenes* in naturally contaminated cold-smoked salmon. Letters in Applied Microbiology; 44(4): 406-11.

Brett MSY, Short P, McLauchlin J. (1998) A small outbreak of listeriosis associated with smoked mussels. International Journal of Food Microbiology; 43(3): 223-9.

Cabedo L, Picart I Barrot L, Teixidó I Canelles A. (2008) Prevalence of *Listeria monocytogenes* and *Salmonella* in ready-to-eat food in Catalonia, Spain. Journal of Food Protection; 71(4): 855-9.

Chitlapilly Dass S, Abu-Ghannam N, Antony-Babu S et al. (2010) Ecology and molecular typing of *L. monocytogenes* in a processing plant for cold-smoked salmon in the Republic of Ireland. Food Research International; 43(5): 1529-36.

Chitlapilly Dass S, Cummins EJ, Abu-Ghannam N. (2011) Prevalence and typing of *Listeria monocytogenes* strains in retail vacuum-packed cold-smoked salmon In the Republic of Ireland. Journal of Food Safety; 31(1): 21-7.

Christie R, Atkins NE, Munch-Petersen E. (1944) A note on lytic phenomenon shown by Group B streptococci. Australian Journal of Experimental Biology and Medical Science; 22: 197-200.

Cornu M, Beaufort A, Rudelle S et al. (2006) Effect of temperature, water-phase salt and phenolic contents on Listeria monocytogenes growth rates on cold-smoked salmon and evaluation of secondary models. International Journal of Food Microbiology; 106(2): 159-68.

Dauphin G, Ragimbeau C, Malle P. (2001) Use of PFGE typing for tracing contamination with *Listeria monocytogenes* in three cold-smoked salmon processing plants. International Journal of Food Microbiology; 64(1-2): 51-61.

Di Pinto A, Novello L, Montemurro F et al. (2010) Occurrence of *Listeria monocytogenes* in ready-to-eat foods from supermarkets in Southern Italy. New Microbiology; 33(3): 249-52.

ESR. (2011) Public Health Surveillance. Information for New Zealand public health action: Annual Surveillance Summary. Accessed at:

http://www.surv.esr.cri.nz/surveillance/annual_surveillance.php. Accessed: 14 April 2011.

Fletcher GC, Rogers ML, Wong RJ. (1994) Survey of *Listeria monocytogenes* in New Zealand seafood. Journal of Aquatic Food Product Technology; 3(2): 13-24.

Gudmundsdóttir S, Gudbjörnsdóttir B, Lauzon HL et al. (2005) Tracing *Listeria monocytogenes* isolates from cold-smoked salmon and its processing environment in Iceland using pulsed-field gel electrophoresis. International Journal of Food Microbiology; 101(1): 41-51.

Guyer S, Jemmi T. (1991) Behavior of Listeria monocytogenes during fabrication and storage of experimentally contaminated smoked salmon. Applied and Environmental Microbiology; 57(5): 1523-7.

Hoffman AD, Gall KL, Norton DM et al. (2003) *Listeria monocytogenes* contamination patterns for the smoked fish processing environment and for raw fish. Journal of Food Protection; 66(1): 52-60.

Hu Y, Gall K, Ho A et al. (2006) Daily variability of listeria contamination patterns in a cold-smoked salmon processing operation. Journal of Food Protection; 69(9): 2123-33.

Hudson JA, Mott SJ. (1993) Growth of *Listeria monocytogenes*, *Aeromonas hydrophila* and *Yersinia enterocolitica* on cold-smoked salmon under refrigeration and mild temperature abuse. Food Microbiology; 10(1): 61-8.

Hudson JA, Mott SJ, Delacy KM et al. (1992) Incidence and coincidence of *Listeria* spp., motile aeromonads and *Yersinia enterocolitica* on ready-to-eat fleshfoods. International Journal of Food Microbiology; 16(2): 99-108.

Hwang CA. (2007) Effect of salt, smoke compound, and storage temperature on the growth of Listeria monocytogenes in simulated smoked salmon. Journal of Food Protection; 70(10): 2321-8.

Jemmi T, Keusch A. (1992) Behavior of *Listeria monocytogenes* during processing and storage of experimentally contaminated hot-smoked trout. International Journal of Food Microbiology; 15(3-4): 339-46.

Jørgensen LV, Huss HH. (1998) Prevalence and growth of *Listeria monocytogenes* in naturally contaminated seafood. International Journal of Food Microbiology; 42(1-2): 127-31.

Junttila JR, Niemela SI, Hirn J. (1988) Minimum growth temperatures of *Listeria monocytogenes* and non-haemolytic *Listeria*. Journal of Applied Bacteriology; 65(4): 321-7.

Latorre L, Parisi A, Fraccalvieri R et al. (2007) Low prevalence of *Listeria monocytogenes* in foods from Italy. Journal of Food Protection; 70(6): 1507-12.

Midelet-Bourdin G, Copin S, Leleu G et al. (2010) Determination of Listeria monocytogenes growth potential on new fresh salmon preparations. Food Control; 21(10): 1415-8.

Miettinen MK, Siitonen A, Heiskanen P et al. (1999) Molecular epidemiology of an outbreak of febrile gastroenteritis caused by *Listeria monocytogenes* in cold-smoked rainbow trout. Journal of Clinical Microbiology; 37(7): 2358-60.

Mitchell DL. (1991) A case cluster of listeriosis in Tasmania. Communicable Disease Intelligence; 15: 427.

Pesavento G, Ducci B, Nieri D et al. (2010) Prevalence and antibiotic susceptibility of *Listeria* spp. isolated from raw meat and retail foods. Food Control; 21(5): 708-13.

Pinto AD, Novelleo L, Montemurro F et al. (2010) Occurrence of *Listeria monocytogenes* in ready-to-eat foods from supermarkets in Southern Italy. New Microbiologica; 33(3): 249-52.

Poysky FT, Paranjpye RN, Peterson ME et al. (1997) Inactivation of *Listeria monocytogenes* on hot-smoked salmon by the interaction of heat and smoke or liquid smoke. Journal of Food Protection; 60(6): 649-54.

Rørvik LM. (2000) *Listeria monocytogenes* in the smoked salmon industry. International Journal of Food Microbiology; 62(3): 183-90.

Seeliger HPR, Jones D, editors. Listeria Baltimore: Williams & Wilkins; 1986. 1235-45 p. (PHA Sneath; NS Mair; ME Sharpe et al. editors. Bergey's Manual of Systemic Bacteriology).

Suppin D, Safer M, Dabbass A et al. (2006) On the prevalence of *Listeria monocytogenes* in cold smoked fish: An overview. Wiener Tierarztliche Monatsschrift; 93(5-6): 145-9.

Vitas AI, Garcia-Jalon VAEI. (2004) Occurrence of *Listeria monocytogenes* in fresh and processed foods in Navarra (Spain). International Journal of Food Microbiology; 90(3): 349-56.

Vogel FB, Huss HH, Ojeniyi B et al. (2001) Elucidation of *Listeria monocytogenes* contamination routes in cold-smoked salmon processing plants detected by DNA-based typing methods. Applied and Environmental Microbiology; 67(6): 2586-95.

WHO. (1988) Foodborne Listeriosis. Bulletin of the World Health Organisation; 66(4): 421-8.

WHO. (2004) Risk assessment of *Listeria monocytogenes* in ready-to-eat foods, MRA Series 4 & 5. Accessed at: http://www.who.int/foodsafety/publications/micro/mra_listeria/en/index.html

Accessed: 15 April 2011.

Zar JH. (1999) Biostatistical Analysis. Upper Saddle River, New Jersey: Prentice Hall.

Regulatory Act / Processor	Type of Sample	Tranche No. Tested	1 No. positive	Tranche 2 No. Tested	No. positive	Total Number Tested	Total Positive
Food Act 1981							
Food Hygie	ne Regulations						
A	Cold smoked	20	3			27	3
	Hot smoked	7					
В	Cold smoked	8				11	0
	Hot smoked	3					
C D	Hot smoked	2				2	0
D	Cold smoked	38		45		116	0
	Hot smoked	17		16			
E	Cold smoked	34		40	1	110	1
	Hot smoked	14		22			
F	Hot smoked	16		17		33	0
G	Hot smoked	7		9		16	0
Η	Hot smoked	15		18		33	0
	Hot smoked	19		19		38	0
J	Cold smoked			16		16	0
					Tota	l 402	
Food Safety	Programme						
K	Cold smoked	24		24		94	0
	Hot smoked	23		23			
Α	Cold smoked			2		8	0
	Hot smoked			6			
L	Cold smoked	30	1	33		114	1
	Hot smoked	24		27			
N	Cold smoked	24		21		78	0
	Hot smoked	18		15			
N	Hot smoked	18		16		34	0
0	Cold smoked	22	1	17	1	69	2
	Hot smoked	17		13			
					Tota	l 397	

APPENDIX 1 Regulatory regime, producer, type and number of smoked salmon samples tested, and numbers positive for *L. monocytogenes*

Listeria monocytogenes in RTE Smoked Salmon (2009-10)

Animal Produ	ucts Act					
Р	Cold smoked	10	10		20	0
Q	Hot smoked		1		1	0
R	Cold smoked	49	48	1	199	1
	Hot smoked	50	52			
S	Cold smoked	41	42		193	0
	Hot smoked	55	55			
					Total 413	
				Grand	d Total 1212	