

LISTERIA MONOCYTOGENES **IN COOKED PRAWNS**

A Microbiological Survey Report

TECHNICAL REPORT SERIES NO. 26

FOOD STANDARDS AUSTRALIA NEW ZEALAND
2002

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Food Standards Australia New Zealand

AUSTRALIA
PO Box 7186
Canberra BC ACT 2610
Australia
Tel +61 2 6271 2241
Fax +61 2 6271 2278
email info@foodstandards.gov.au

NEW ZEALAND
PO Box 10599
Wellington
New Zealand
Tel +64 4 473 9942
Tax +64 4 473 9855
email info@foodstandards.govt.nz

Survey of *Listeria monocytogenes* in Cooked Prawns

2002

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Summary

A retail microbiological survey of 380 cooked prawn samples was coordinated by Food Standards Australia New Zealand (FSANZ) for the purpose of determining the frequency and level of contamination of *Listeria monocytogenes*. Peeled and unpeeled, imported and domestic cooked prawns were sampled and the retail temperature of the prawns was determined. Sample collection was conducted by State Health Departments, and food analysis was conducted by the Australian Government Analytical Laboratories (AGAL) and the Western Australia Centre for Pathology and Medical Research.

The microbiological status of prawns was assessed through testing for Standard Plate Count (SPC) and *Listeria monocytogenes*. *Listeria monocytogenes* was detected in 3% of cooked prawns and the population densities, when detected, were less than 50 cfu/g. The SPCs ranged from less than 10^3 to greater than 10^7 cfu/g and the temperatures of cooked prawns at retail display varied from -6°C to 12.8°C . There was no apparent association between high SPCs and high display temperatures recorded for cooked prawns.

1. Introduction

1.1 Rationale of Survey

In November 2000, the then Australia New Zealand Food Standards Council requested that Food Standards Australia New Zealand (FSANZ) review the microbiological criteria for *Listeria monocytogenes* in crustacea contained in the Australia New Zealand *Food Standards Code* (FSC). Standard 1.6.1 – Microbiological Limits for Foods, of the FSC (Volume 2), required a “nil detect” limit for *Listeria monocytogenes* in all five 25g samples from a batch of cooked crustacea at any stage during its stated shelf life.

As part of this review, FSANZ performed a microbiological risk assessment to estimate the public health risk posed to Australian and New Zealand consumers from the consumption of cooked crustacea contaminated with *Listeria monocytogenes* (FSANZ, 2002). The risk assessment undertaken was predominantly qualitative with some quantitative elements, and followed the principles of the Codex Alimentarius Commission for Microbiological Risk assessments (Codex Alimentarius Commission, 1999). The preliminary findings of the microbiological risk assessment concluded that there was the potential for cooked crustacea to be contaminated with *Listeria monocytogenes* (FSANZ, 2002).

An important outcome of the preliminary risk assessment findings was that there were insufficient data on the occurrence of *Listeria monocytogenes* contamination in cooked crustacea in Australia to undertake a comprehensive evaluation of the risk to public health and safety. As a result, a survey of *Listeria monocytogenes* in cooked prawns was commissioned by FSANZ to fill the information gap.

The data gathered from the survey were used in the final microbiological risk assessment. The assessment utilised a probabilistic model to estimate predicted levels of listeriosis in the population that may result from the consumption of contaminated cooked crustacea.

1.2 Supply of Cooked Crustacea in Australia

Cooked crustacea is distributed to retail traders as either chilled or frozen product. The majority of cooked crustacea is frozen at wholesale, with only a small percentage (approximately 10%) being chilled. Frozen cooked crustacea are marketed as frozen and are also often thawed before retail display. Cooked prawns are sold both peeled and unpeeled. Peeled prawns are typically cooked before the shell is removed. Peeling of prawns is increasingly done by machine, but hand-peeling still accounts for much of the world trade in prawns, particularly from non-industrialised countries (ICMSF, 1998). Cooked crustacea sourced in Australia are typically sold unpackaged. Imported cooked crustacea are individually quick frozen, glazed and then thawed prior to sale.

The microflora of raw crustacea largely reflects the quality of the water from which the crustacea are harvested, and is also affected by the on-ship and in-plant environment, handling practices, and the duration and type of refrigerated storage. Inactivation studies indicate that cooking may not destroy all bacteria if initial levels are high. There are reports of bacterial counts of 4.5×10^4 cfu/g on tropical prawns after boiling for 3 minutes (ICMSF, 1998). However, contamination is more often associated with cross-contamination of raw and cooked crustacea coupled with inadequate cold storage (ICMSF, 1998). Chilled or thawed cooked crustacea generally have a shelf-life of 10 days at -1°C (FSANZ, 2002), giving limited opportunity for microbial growth to occur before the food is consumed.

2. Objective of the survey

The main objective of the survey was to determine:

- the frequency and level of *Listeria monocytogenes* contamination in both domestically produced and imported cooked prawns at the retail level.

In addition, the standard plate count (SPC) and internal temperature of the prawns was measured in retail samples of cooked prawns to provide information on the handling and storage of this type of product.

3. Design and methods

3.1 Sampling design

Australian and imported unpackaged cooked prawns were sampled for this survey. Samples included peeled and whole prawns that were frozen and chilled, from a range of species and a range of sizes (e.g. prawns marketed as small, medium and large). Priority was given to the most frequently sold products/species and sizes at each sampling location. Both wild caught and farmed prawns were sampled.

The survey took place over a 4 week period during July and August 2002. A total of 380 samples were collected from four locations: Perth, Brisbane, Melbourne and Sydney. Environmental health officers purchased the cooked prawns according to a detailed sampling plan.

Cooked prawn samples were purchased from a range of retail outlets in each city including major supermarkets, specialist seafood retailers and fish markets. The number of samples from each retailer was limited to three. The sampling plan for each location is listed in Table 1.

Table 1: Sampling plan for prawns by time and location

Retail Location	Number of Samples				
	Week 1	Week 2	Week 3	Week 4	Total
Major Supermarkets	13	12	13	12	50
Specialist Seafood Retailers and Fish Markets	12	13	12	13	50
TOTAL	25	25	25	25	100

3.2 Methods

3.2.1 General methods

For each sample, either 100 g of peeled prawns or 300 g of unpeeled prawns was chosen for microbiological analysis. One sample was required for frozen prawns whereas chilled prawns required an additional sample of 50-100g for temperature measurement.

Cooked prawns that were subject to microbiological analysis were sampled aseptically, placed in insulated containers with chiller packs and delivered to the laboratory in each city on the day they were purchased.

For chilled prawns, the internal temperature was measured in three individual prawns and an average temperature recorded. Where individual prawn size made it unpractical to measure the internal temperature, the temperature was measured at three different locations in the pile from which the prawns were purchased. The temperature measurement was not taken for prawns that were visibly frozen; in this case it was recorded as “frozen”.

Officers purchasing samples completed a questionnaire (Appendix 1) at the time of sampling, which provided details of the product and retail establishment including the conditions of storage and display. Information was recorded on the history and details of the product including best-before dates if available. Samples were held at 0-5°C until microbiological analysis commenced, and laboratories were instructed to commence analysis within 24 hours of receiving samples.

3.2.2 Microbiological analysis

The microbiological analyses were conducted by the Australian Government Analytical Laboratory (AGAL) and the Western Australian Centre for Pathology and Medical Research. All cooked prawns were analysed for *Listeria monocytogenes* and SPC using standard methods (Appendix 2).

The enumeration of *Listeria monocytogenes* was conducted in tandem with the presence/absence test for *Listeria monocytogenes*. The limit of detection for enumerating *Listeria monocytogenes* was 50 cfu/gram.

A dilution range of 10^{-3} – 10^{-5} was used to determine the standard plate count for each sample (sub-sample). This gave a detection range of 10^4 – 10^7 cfu/g.

4. Results

A total of 380 samples of cooked prawns were collected from Brisbane, Sydney, Melbourne and Perth. Eighty-nine samples (23%) were peeled, 290 samples (76%) were unpeeled and one sample was not recorded as either peeled or unpeeled. Fifty-one percent of samples were collected from supermarkets and 49% from specialist seafood retailers or fish markets. Table 1 gives a breakdown of prawns by frequency and type.

Table 1: Prawns sampled by frequency and type

No. of samples	Production method		Origin		Source of sample		Display		Form	
	Farmed	Wild caught	Imported	Domestic	Super-market	Seafood retailer /fish market	Frozen	Chilled	Peeled	Unpeeled
	69	165	139	230	195	185	67	313	89	290
Total	234		380		380		380		379	

4.1 Temperature at retail

Cooked prawns were sampled either as ‘frozen’ (i.e. they were displayed at retail in freezer cabinets) or as ‘chilled’ (displayed at retail under chilled conditions). There were 67 samples (18%) recorded as ‘frozen’ compared to 313 chilled samples (82%). The average temperatures recorded for chilled cooked prawn samples at retail varied from -5.5°C to 12.8°C . Of these, 113 samples (36%) had average temperatures exceeding 5°C . One hundred and eighty-three samples (59%) had average temperatures between 0°C and 5°C , and 17 samples (5%) had average temperatures less than 0°C . Figure 1 shows the average temperature of cooked prawn samples, excluding samples recorded as frozen.

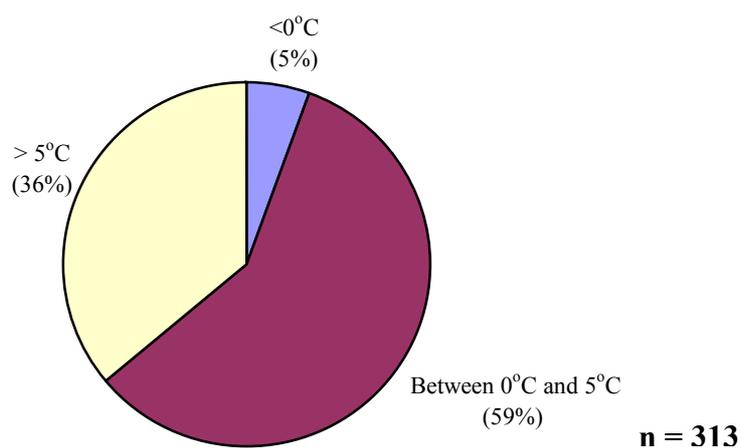


Figure 1: Average temperature of cooked prawn samples at retail (excluding samples recorded as ‘frozen’)

4.2 Standard Plate Count

4.2.1 Frozen versus chilled prawns

Figure 2 shows the range of SPCs obtained for both frozen and chilled cooked prawns. The SPC ranged from less than 10^3 to greater than 10^7 cfu/g.

Cooked prawns displayed in freezer cabinets ('frozen') all had SPCs of less than 10^6 cfu/g. Fifty-seven percent of 'frozen' samples had mean SPCs of less than 10^3 cfu/g, 27% had SPCs between 10^3 and 10^4 cfu/g, 9% had SPCs between 10^4 and 10^5 cfu/g and 7% had SPCs between 10^5 and 10^6 cfu/g.

Cooked prawns displayed under chilled conditions had SPCs ranging from less than 10^3 to greater than 10^7 cfu/g. Ten percent of chilled cooked prawns had mean SPCs less than 10^3 cfu/g, 21% had SPCs between 10^3 and 10^4 cfu/g, 30% had SPCs between 10^4 and 10^5 cfu/g, 21% had SPCs between 10^5 and 10^6 cfu/g, 14% had SPCs between 10^6 and 10^7 cfu/g and 5% had SPCs greater than 10^7 cfu/g.

The SPCs for frozen cooked prawns were generally lower than for chilled cooked prawns.

Although the best-before date was recorded on the questionnaire when available, there was insufficient information to associate the high SPCs with best-before dates.

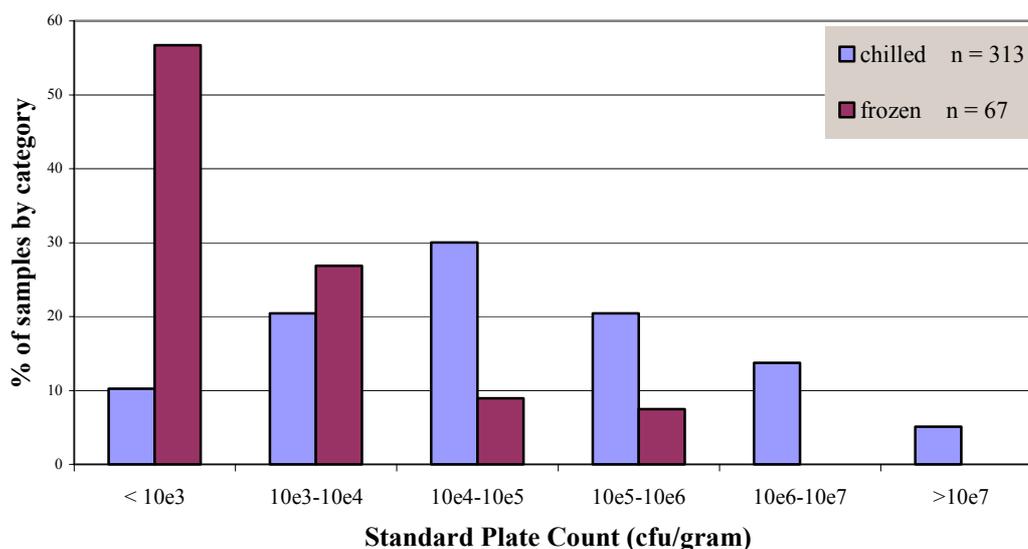


Figure 2: The range of mean SPC for both frozen and chilled cooked prawns

4.2.2 Peeled versus unpeeled prawns

Figure 3 shows the range of SPCs obtained for both unpeeled and peeled cooked prawns expressed as a percentage of the total number of samples for each category. There is no apparent difference in SPCs for unpeeled and peeled prawns.

There was no association between high SPCs and whether the prawns were peeled or unpeeled, imported or domestic, or the different types of retail outlets.

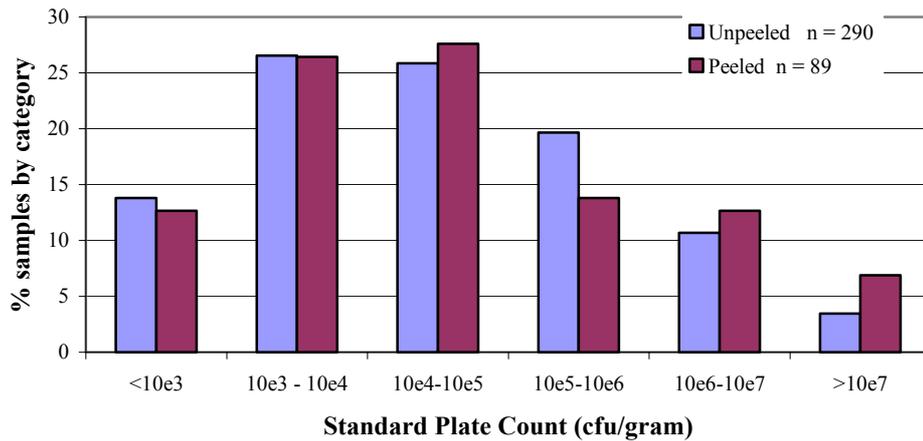


Figure 3: SPC for peeled and unpeeled cooked prawns expressed as a percentage of each category of product

4.2.3 Farmed versus wild caught prawns

A comparison of SPCs for farmed and wild caught cooked prawns showed that wild caught prawns had a relatively higher percentage of SPCs in the upper range, when compared to farmed prawns (Figure 4).

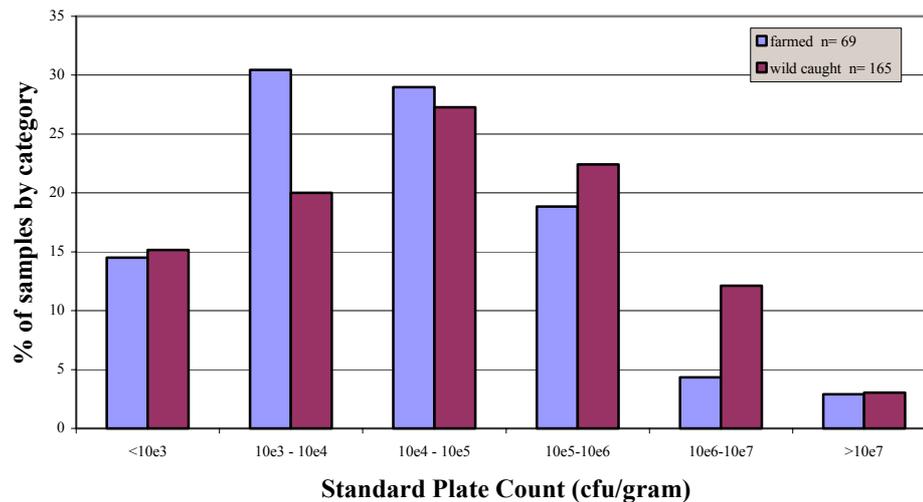


Figure 4: SPC for farmed and wild caught cooked prawns expressed as a percentage of each category

4.3 *Listeria monocytogenes*

Table 2 provides a breakdown of the total number of samples positive for *Listeria monocytogenes* according to origin (imported or domestic product) and type (peeled or unpeeled).

Twelve of the 380 samples (3%) were positive for *Listeria monocytogenes* although there were no counts greater than 50 cfu/g. Seven peeled prawn samples were positive for *Listeria monocytogenes*, compared to 5 unpeeled samples. This difference was statistically significant (*Fishers Exact*, $p=0.009$). Of the positive samples, 7 (5%) were imported product and 4 (2%) were domestic product. There was no significant difference in the contamination level between imported and domestic prawns (*Fishers Exact*). One positive sample was of unknown origin.

Table 2: Comparison between number of samples positive for *Listeria monocytogenes* and the total number of samples

Type of Prawn	Number Positive for <i>Listeria monocytogenes</i>			Total
	Imported	Domestic	Unknown	
Peeled	6/77(8%)	0/8	1/4	7/89(8%)
Unpeeled	1/62(2%)	4/221 (2%)	0/7	5/290(2%)
Unknown		0/1		0/1
Total	7/139 (5%)	4/230(2%)	1/11	12/380(3.2%)

5. Discussion

Contamination with *Listeria monocytogenes* can arise through a variety of sources such as contaminated cooling water, the use of contaminated containers for cooked product, and inadequate separation of raw and cooked product. Once in the marketplace, unsealed product and product that has been removed from its packaging for pre-sale thawing and/or display may be cross-contaminated through contact with ice, utensils, surfaces, handlers and other food products.

Once processed, cooked crustacea can be marketed chilled or frozen. Frozen product may be stored in bulk and thawed at the retail level for display and sale. While *Listeria monocytogenes* cannot grow in frozen product, chilled product presents opportunities for bacterial growth during distribution and retail display. High display temperatures can favour bacterial growth. The current survey, however, does not confirm whether or not there is a correlation between the retail display temperatures recorded for cooked prawns and the incidence of positive samples for *Listeria monocytogenes*.

Of the 380 samples tested for *Listeria monocytogenes*, only 12 samples (3%) tested positive. Peeled prawns had a higher contamination rate compared to unpeeled prawns (8% and 2% respectively), possibly reflecting more extensive handling. This was statistically significant (*Fishers Exact*, $p=0.009$). There were insufficient domestic peeled prawn samples to compare the rate of contamination with imported peeled prawns. The contamination rates for unpeeled prawns were similar for both imported and domestic products.

Probabilistic modelling was undertaken by Food Science Australia for the FSANZ microbiological risk assessment of *Listeria monocytogenes* in Cooked Crustacea (FSANZ, 2002). Using the results from this survey, the modelling estimated that, assuming there was no growth of the organism during domestic storage, one case of listeriosis every 1,600 years may arise from the consumption of contaminated crustacea in Australia. Modelling of a worst-case scenario, which assumed that the cooked prawns would be stored in a domestic refrigerator for a maximum of three days after purchase (allowing growth of *Listeria monocytogenes*), indicated that there could be one case of listeriosis caused by cooked crustacea every 2.5 years in Australia. However, the significant difference reported in this survey between contamination rates of cooked peeled versus unpeeled prawns (8% and 2%), indicated that there may be a higher exposure of individuals to *Listeria monocytogenes* where those individuals have consumed cooked peeled product versus other forms of crustacea. The risk assessment nevertheless concluded that *Listeria monocytogenes* in cooked crustacea poses a low risk to the Australian population. The review of the microbiological limits for cooked crustacea also concluded that there was no

justification for retaining a *Listeria monocytogenes* standard for cooked crustacea in the FSC. However, further education is needed to alert at risk populations¹ to avoid cooked peeled crustacea.

It is difficult to relate the level of *Listeria monocytogenes* contamination found in cooked prawns in this survey (<50 cfu/g) to other published data as little quantitative work has been conducted on the levels of *Listeria monocytogenes* in cooked crustacea and seafood products generally. However, the *Listeria monocytogenes* contamination rates in this survey are comparable with surveys published in the international literature where rates in retail samples are frequently below 10% (Adesiyun, 1993; Gecan *et al.*, 1994; Hartemink *et al.*, 1991; Hofer *et al.*, 1990; McLauchlin and Nichols, 1994). As a comparison, contamination rates of smoked salmon with *Listeria monocytogenes* are much higher and generally range from 10 to 20% at retail, with some recorded contamination rates of up to 40% (FSANZ, 2002). It was not possible to determine the source of contamination for prawns in the current survey.

FSANZ has established regulatory limits for SPC in both cooked and raw crustacea and limits are set out in Standard 1.6.1 of the FSC. The survey showed that approximately 15% of samples (58/380) exceeded the SPC limit of 10⁶ cfu/g for cooked crustacea. However, high SPCs were not necessarily correlated with the highest temperatures recorded at retail display. High SPCs can be indicative of poor process control as a result of inadequate cooking and/or cross-contamination, and/or inappropriate temperature control during storage and display. In addition, samples with high SPC levels may indicate that, if pathogens were present in low numbers, there may have been an opportunity for their growth before chilled product reaches consumers.

Initial bacterial loads of prawns at the processing plant can range from 10³ to 10⁷ cfu/g (ICMSF, 1998). Prawns cooked by boiling or steaming for a few minutes either before or after peeling show an approximate 100- to 10,000- fold reduction in mesophilic and psychrotrophic counts (Ridely and Slabyj, 1978). However, there tends to be a rapid return to bacterial levels similar to that before cooking, as the product is held and handled during inspection and grading (ICMSF, 1998).

Results of the mean SPCs in cooked prawns from this survey (Figure 2) indicated that the range of levels followed a normal distribution, and suggested that there were no gross failures in processing, handling or cross-contamination. These data on SPC levels could be used in the future to allow refinement and/or assessment of the appropriateness of the limit set in the FSC. However, further surveys are needed to confirm these results. The lower SPCs for frozen product were expected, as microorganisms do not grow in frozen product.

A relatively higher percentage of wild caught prawns had high SPCs compared to farmed prawns. A possible explanation for this is that wild crustacea, if cooked on board the vessel, result in a greater potential for post-cooking contamination as a result of the extended time of storage prior to retail sale. It is more difficult to ensure good hygienic practice at sea. However, not all wild caught prawns are cooked at sea with some cooked in the factory. As it was not possible to determine how many of the wild caught prawns were cooked at sea in this survey, this hypothesis cannot be confirmed.

¹ at risk populations include: pregnant women and their foetuses, neonates, the elderly, and the immuno-compromised

Standard 3.2.2 of the FSC requires food businesses to store potentially hazardous foods under temperature control. While the majority (64%) of cooked prawn samples (excluding 'frozen' product) were stored at or below 5°C, 36% of cooked prawn samples in this survey recorded temperatures above 5°C at the time of sampling. Some individual prawn samples displayed under refrigeration had recorded temperatures as high as 12°C. The high temperatures indicate that a number of retail outlets need to improve the temperature control of displayed cooked prawns. It is also an issue about which food businesses may require further education.

6. Conclusions

Results from this survey indicate that both the contamination rate and the level of contamination in cooked prawns with *Listeria monocytogenes* in Australia is low (3% and <50 cfu/g respectively) and presents a low public health and safety risk. Notwithstanding this, at risk sub-populations should avoid eating cooked peeled prawns and further education is needed for these groups.

The FSC sets a SPC regulatory limit of 10⁶ cfu/g for cooked crustacea. All frozen cooked prawns met this standard, but 19% of chilled cooked prawns exceeded this limit. These results indicate the need for improvements in the way these products are handled and/or stored.

The majority (64%) of chilled cooked prawns recorded temperatures at retail display of less than or equal to 5°C. However, 36% of chilled cooked prawn samples had temperatures exceeding 5°C. This indicates a need for some retail outlets to improve the temperature control of displayed cooked prawns and education of food businesses may be warranted in this area.

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- Food Safety Victoria
- Health Department of Western Australia
- NSW Health
- Queensland Health

Analytical testing

- Australian Government analytical Laboratories
- Queensland Health Scientific Services
- Western Australia Centre for Pathology and Medical Research

8. References

- ANZFA (1999) P178 – Review of Microbiological Standards – Full Assessment Report, ANZFA, Canberra
- Adesiyun (1993) Prevalence of *Listeria* spp., *Campylobacter* spp. *Salmonella* spp. *Yersinia* spp. and toxigenic *Escherichia coli* on meat and seafoods in Trinidad, *Food Microbiology*, 10, 395 – 403
- Codex Alimentarius Commission (1999) Principles and Guidelines for the Conduct of Microbiological Risk Assessment, 23rd Session, Rome, ftp://ftp.fao.org/codex/alinorm99/cac23rep_complet.pdf (accessed July 2001)
- European Commission Health & Consumer Protection Directorate-General (1999) Opinion of the Scientific Committee on Veterinary Measures Relating To Public Health on *Listeria monocytogenes*.
- Farber and Peterkin (1999) Incidence and Behavior of *Listeria monocytogenes* in Meat Products, in Marth and Ryser (eds) *Listeria, Listeriosis and Food Safety* 2nd Edition, Marcel Dekker Inc., New York
- FAO (1999) FAO Fisheries Report No. 604, FAO Expert Consultation on the trade impact of *Listeria* in fish products Amherst USA 17-20 May 1999, FAO, Rome (<http://www.fao.org/icalog/inter-e.htm>) (report accessed February 2002)
- FSANZ (2002) Final Assessment Report, Proposal P239, *Listeria* risk assessment and risk management strategy, FSANZ, Canberra
- Gecan J, Bandler R and Staruszkiewicz W (1994) Fresh and frozen shrimp: a profile of filth, microbiological contamination, and decomposition, *Journal of Food Protection*, 57 (2) 154 - 158
- Harrison, M.A., Huang, Y.W., Chao, C.-H. and Shineman, T. (1991) Fate of *Listeria monocytogenes* on packaged, refrigerated and frozen seafood. *Journal of Food Protection*, 54, 524-7
- Hartemink R and Georgsson F (1991) Incidence of *Listeria* species in seafood and seafood salads, *International Journal of Food Microbiology*, 12, 189 - 196
- Hasell (2000) Food safety - are we winning? *New Zealand Food Journal*, Vol 30, 34-35
- Hofer E and Ribeiro R (1990) Ocorrência de espécies de *Listeria* em camarão industrializado, *Revista de microbiologia*, 21 (2), 207 – 208
- ICMSF (1996) *Microorganisms in Food 5: Microbiological Specifications of Food Pathogens*, Blackie Academic and Professional, London
- ICMSF (1998) *Microorganisms in Foods. 6. Microbial ecology of food commodities*. Blackie Academic and Professional, Great Britain.

Marth (1988) Disease Characteristics of *Listeria monocytogenes*, *Food Technology*, Vol 42, No 4. 165-168

McLauchlin J and Nichols G (1994) *Listeria* and seafood, *PHLS Microbiology Digest*, 11 (3), 151 - 154

McLauchlin (1995) What is the infective dose for human listeriosis? in Proceedings of the XII International Symposium on Problems of *Listeria* in Perth, Promaco Conventions Pty Ltd Canning Bridge Western Australia

National Advisory Committee on Microbiological Criteria for foods (1991) The Human Infective Dose of *Listeria monocytogenes*, *International Journal of Food Microbiology*, Vol 14, 216-219

Ridley, S.C. and Slabyj, B>M> (1978) microbiological evaluation of shrimp (*Pandalus borealis*) processing. *Journal of Food Protection*, 41, 40-3

Rocourt and Brosch (1992) Human Listeriosis 1990, WHO Collaborating Centre for Foodborne Listeriosis

Sutherland and Porritt (1997) *Listeria monocytogenes*, in Hocking (editor-in-chief), *Foodborne Microorganisms of Public Health Significance* 5th Edition, Australian Institute of Food Science and Technology Inc., Sydney

United States Food and Drug Administration (January 19 2001) Relative Risk to Public Health from Foodborne *Listeria monocytogenes* among selected categories of ready-to-eat foods, Draft risk assessment, Federal Register 66 no 13

Appendix 1: Survey Questionnaire

Food Standards Australia New Zealand *Listeria monocytogenes* in Cooked Prawns Survey 2002

Sampling and Analysis Details

Section A of this form is to be completed at the time of sampling.

Section B is to be completed by the representative of the laboratory conducting the analysis. This form should then be forwarded to the survey co-ordinator together with the completed "Sampling and Analysis Details" forms for the other samples.

Sample Identification Code:

(see Guide to Sampling)

e.g. N-SM-1-05-ABC

	-			-		-		-				
--	---	--	--	---	--	---	--	---	--	--	--	--

Section A – Sampling Details

1. Name of sampler

2. Name of retail outlet

3. Address of premises

4. Type of retail outlet

Supermarket

Fish Market

Specialist Retailer Seafood

5. Date sample taken / /2002

6. Time sample taken hours (24 hour time)

7. Type of prawn

- | | |
|-------------------------------------|---|
| <input type="checkbox"/> Banana | <input type="checkbox"/> Royal Red |
| <input type="checkbox"/> Bay | <input type="checkbox"/> School |
| <input type="checkbox"/> Coral | <input type="checkbox"/> Tiger |
| <input type="checkbox"/> Endeavour | <input type="checkbox"/> Unspecified |
| <input type="checkbox"/> Freshwater | <input type="checkbox"/> Other (please specify) |
| <input type="checkbox"/> King | _____ |

8. Size of prawn (as advertised at point of sale)

- | | |
|---------------------------------|---|
| <input type="checkbox"/> Small | <input type="checkbox"/> Unspecified |
| <input type="checkbox"/> Medium | <input type="checkbox"/> Other (please specify) |
| <input type="checkbox"/> Large | _____ |

9. Peeled/Whole Prawns

- Unpeeled (whole) prawns
- Peeled prawns

10. Origin of prawns

- Imported Country of Origin: _____
- Domestic Region of Origin: _____
(Australian)

11. Wild caught or farmed

- Wild caught
- Farmed
- Unknown

12. Point of sale storage conditions

- | | |
|---|---|
| <input type="checkbox"/> Refrigerated cabinet | <input type="checkbox"/> Cold air flow |
| <input type="checkbox"/> Freezer | <input type="checkbox"/> On ice |
| <input type="checkbox"/> Walk in cool room | <input type="checkbox"/> Other (please specify) |
-

13. Chilled or frozen prawns

- | | |
|----------------------------------|---|
| <input type="checkbox"/> Chilled | <input type="checkbox"/> Frozen (go to Q15) |
|----------------------------------|---|

14. Record the internal temperature of the 3 of the prawns (or, for very small prawns, at 3 locations in the pile of prawns) from **Sub-sample B** within 5 minutes of purchase (indicate + or -)

Prawn 1:

--	--	--

 °C

Prawn 2:

--	--	--

 °C

Prawn 3:

--	--	--

 °C

15. Brand Name of the Cooked Prawns (if available):

--

16. Batch Code (if available):

--

17. Best-Before-Date (if available):

--

18. Other comments

19. Signature of sampler

20. Date

Section B – Recording of Results (For Laboratory Use Only)

General

21. Date of sample received / /2002

22. Time sample received hours
(24 hour time)

23. Temperature of sample on arrival (indicate + or -)
 °C or Frozen

24. Time and Date testing of sample commenced
 Hrs on / /2002

Results of analysis

25. Presence of *Listeria monocytogenes* in 25 grams

Detected

Not Detected

26. Record the results of *L. monocytogenes* enumeration

Plate 1: . _____ c.f.u per gram

Plate 2: . _____ c.f.u per gram

27. Standard Plate Count per gram

_____ c.f.u. per gram

Appendix 2 Methods of Analysis

Tests Performed and Methods of Analysis

Test	Analysis Method
<i>Listeria monocytogenes</i> – Presence/Absence	Australian S 1766.2.16.1 1998
<i>Listeria monocytogenes</i> enumeration*	ISO 11920-2:1998
Standard Plate Count	AS 1766.2.1

AS Australian standard, ISO International standard organisation

* The following variations to the ISO 11290-2: 1998 method for *Listeria monocytogenes* were employed:

- At Step 9.2.1, 0.2 ml of a 10⁻¹ dilution was transferred by sterile pipette to each of the 2 dishes of PALCAM agar; and
- At Step 9.7, a DNA probe (such as Geneprobe) was used for *Listeria monocytogenes* species confirmation.

Appendix 3 Microbiological Terms

Standard Plate Count (SPC)

A standard plate count is used for determining the total number of viable mesophilic colony forming units in a food product. Total counts denote the microbiological load in a food and may be used to indicate unsatisfactory handling practices, process failure or improper storage.

Colony forming units (cfu)

Food samples are diluted and plated onto suitable agar medium. Any viable microbial cells present can multiply and form visible colonies. Each of these colonies is counted as a colony forming unit.