BIOSECURITY TASMANIA

GUIDELINES FOR THE SAFE MANUFACTURE OF SMOKED FISH: FOCUS ON LISTERIA MANAGEMENT

Department of Primary Industries, Parks, Water and Environment

Tasmanian Government

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Table of Contents

Glossary of Terms and Abbreviations

ABS	Australian Bureau of Statistics
Ambient temperature	Temperature of the air around you or the product
Anaerobic	The absence of oxygen, a state which can exist in canned and vacuum-packed products
Biofilm	A mixture of microorganisms and their excretory products that builds up in food processing equipment
CCP	Critical Control Point. A point, procedure, operation or stage in a process at which a hazard is prevented, eliminated or reduced to an acceptable level
CFU	Colony Forming Unit, an estimate of viable number of bacteria
CL	Critical Limit – a criterion which separates acceptability from unacceptability
Cold chain	The process of maintaining foods under refrigeration, in either a chilled or frozen state, during storage, distribution and marketing
Contaminant	Something that may make food unsafe or unwholesome. Examples of contaminants are microorganisms, chemical residues or foreign matter
Controlling Authority	The Commonwealth, State or Territory authority that is responsible for the enforcement of food safety standards for smoked fish. The controlling authority in Tasmania is the Biosecurity Tasmania Division of the Department of Primary Industries, Parks, Water and Environment.
CP	Control Point
CSS	Cold smoked salmon
FSANZ	Food Standards Australia New Zealand
FSP	Food Safety Program
FSSP	Food Spoilage and Safety Predictor
GMP	Good Manufacturing Practice
HACCP	Hazard Analysis Critical Control Point is the system that identifies and controls those hazards which pose a significant risk to food safety
Hazard	A biological, chemical or physical agent which may compromise or affect food safety
LAB	Lactic acid bacteria
Log	Logarithm — used to express microbial counts e.g. log 2 is 100, log 3 is 1,000
HOG	Head on gutted
HSS	Hot smoked salmon

Microbial count	The number of microorganisms living in or on a food product
Microbiological limits	The maximum number of microorganisms specified for a food product
Microorganisms	Viruses, yeasts, moulds and bacteria
MAP	Modified Atmosphere Packaging. Enclosure of product in high gas barrier film, in which the gas environment around the product has been changed by removing all the air from pack and flushing it with a gas mixture of varying concentrations of oxygen, carbon dioxide and nitrogen. Vacuum packaging (VP) where, most of the air is removed before sealing the pack, is sometimes included in MAP
NATA	National Association of Testing Authorities
NSWFA	New South Wales Food Authority
Pathogen	A microorganism which causes illness
pH	A measure of acidity or alkalinity
PPPS 4.2.1	Primary Production and Processing Standard for Seafood
PRP	Pre-requisite program
QCP	Quality Control Points
QUAT	Quaternary ammonium compounds
RCP	Regulatory Control Point
RTE	Ready to eat
Shelf life	Length of time that a commodity may be stored without becoming unfit for use or consumption, due to loss of quality, the presence of undesirable chemicals, toxins, or growth of pathogens.
Spoilage bacteria	Bacteria which limit the shelf life of foods by producing objectionable odours, colours or slime
SSOP	Sanitation Standard Operating Procedure
Toxin	A chemical that can cause illness. Toxins may be produced in food by bacteria and moulds
Validate, validation	The process of obtaining evidence to demonstrate that hazards in a food process are controlled
Verify, verification	Means applying methods, procedures, tests and other evaluations in addition to monitoring to determine whether a requirement is complied with or a matter is met
WPS	Water phase salt

Foreword

Manufacture of smoked fish products involves many risk factors, summed up by the Food Standards Australia New Zealand (FSANZ) *Risk Ranking of Seafood in Australia* (FSANZ, 2005), which found that*:*

1. Contamination of cold smoked products with *Listeria monocytogenes* (*Lm*) at levels representing a health risk to the general population is considered unlikely. However, this rises to '*likely*' where there is insufficient management of risk through the food chain and for susceptible sub-populations and rises further to '*very likely*' when both conditions apply.
2. Other than scrupulous factory hygiene, there was no critical control point (CCP) available at the time to prevent contamination of ready-to-eat (RTE) cold smoked seafood products. Hot smoking can eliminate *Lm* on the product, but post-processing contamination can occur.

In 2019, there were several cases of listeriosis in Australia in which Tasmanian smoked salmon was implicated and two people from a vulnerable population died. Although no breaches of the Tasmanian *Primary Produce Safety Act 2011*, *Primary Produce Safety (Seafood) Regulations 2014* and applicable national food safety standards were identified by Biosecurity Tasmania (the regulator and controlling authority) in relation to this product, sufficient attention was drawn to the matter nationally to indicate that smoked salmon could continue to be scrutinised by health authorities and the public in terms of its safety.

Accordingly, the Tasmanian Department of Primary Industries, Parks, Water and Environment (DPIPWE) commissioned a review of its regulatory system as applied to monitor *Listeria* management and implement appropriate controls across Tasmania's smoked salmon producers. The scope of this review encompassed the operations of each accredited producer to assess the effectiveness of industry's *Listeria* management.

Consequently, in these Guidelines we aim to provide risk-based tools for food safety and *Listeria* management in the production of smoked salmon (and trout) with clear scientific underpinning for industry practices. By incorporating the Guidelines into Tasmania's Primary Produce Safety regulatory framework, we can facilitate compliance and mitigate potential non-compliance with the *Australia New Zealand Food Standards Code*.

The Guidelines were drafted by Dr John Sumner in cooperation with DPIPWE staff and benefitted greatly from the technical inputs of the Tasmanian salmon industry.

Purpose

The Guidelines for the Safe Manufacture of Smoked Fish: Focus on *Listeria* Management (the Guidelines) are intended as a reference for businesses and also for the controlling authority and food safety auditors.

The Guidelines explain the basic problem confronting processors of smoked fish – that fish entering the processing plant bring with them *Lm*, a significant pathogen that will make its way through to the final product unless:

- A CCP is employed that prevents, eliminates or reduces *Lm* to an acceptable level, together with
- A slicing and packing environment in which the presence of *Listeria* is minimised

A series of checklists is provided that will enable a business to assess its standard of *Listeria* management.

Then follow a series of appendices:
Appendix 1: CPs, RCPs, CCPs and GMPs in smoked fish processing
Appendix 2: Keeping the plant clean
Appendix 3: Microbiological testing
Appendix 4: Target bacteria and how to control them
Appendix 5: Target hazard *L. monocytogenes*

The International Commission on Microbiological Specifications for Foods (ICMSF, 2002) classifies *Lm* as a 'Severe' hazard – the highest category, and in Appendix 5 is a Hazard Sheet setting out important aspects of the pathogen.

What you need to do after reading these Guidelines

Review your food safety program: its processes, work instructions and monitoring forms. Only you can do this for your individual operation and for approval by the controlling authority.

If starting from scratch, set out how to meet all the provisions of the *Australia New Zealand Food Standards Code* and other relevant standards. You need to do this before the controlling authority will approve your food safety plan.

These include:
Standard 1.2 - Labeling (ingredients, allergens, date marking)
Standard 1.3.1 - Food Additives
Standard 1.3.3 - Processing Aids
Standard 1.6.1 - Microbiological Limits for Food (Schedule 27)
Standard 3.1.1 - Interpretation and Application
Standard 3.2.1 - Food Safety Programs
Standard 3.2.2 - Food Safety Practices and General Requirements
Standard 3.2.3 - Food Premises and Equipment
Standard 4.2.1 – Primary Production and Processing Standard for Seafood

The Standards can be downloaded from the Food Standards Australia New Zealand (FSANZ) website, www.foodstandards.gov.au.

1. Hazards and risks in smoked seafoods

Purpose

You have a Food Safety Program (FSP) in which you identify and manage hazards and risks in your HACCP plan.
In this section we:

- Identify a major hazard that confronts you in your process: *Lm*
- Spell out how *Lm* in raw fish progresses to final products
- Identify how to minimise the likelihood that final products will be contaminated

The impact of *Lm* in smoked seafoods in general, and from salmon in particular can be measured from two sources of information:

i. Recalls of smoked seafood in Australia for presence of *Lm* – this information is available on the FSANZ website.
ii. Food illness caused by *Lm* in smoked fish in Australia and in other countries.

Product recalls of smoked seafoods

As well as biological hazards, chemical and physical hazards can also cause injury and, if they are found to be present, result in a recall of all affected product.

The FSANZ website records all food recalls and in Table 1 is a list of smoked seafood recalls from March 1998 - July 2019. Smoked salmon has been recalled on three occasions for presence of *Lm*, one of which (2004) was an imported product from Denmark, indicating that the bacterium is a global problem in smoked seafoods, as is seen in the next section on illness caused by *Lm*.

Table 1: Recalls of smoked seafood in Australia 1998-2019

Date	Product	Origin	Cause of recall
2018	Smoked salmon	Tasmania	Mislabelling
2017	Hot smoked salmon	Tasmania	*L. monocytogenes*
2013	Smoked mussels	New South Wales	*L. monocytogenes*
2011	Salmon terrine	Tasmania	*L. monocytogenes*
2011	Smoked salmon	Tasmania	*L. monocytogenes*
2011	Smoked trout dip	New South Wales	*L. monocytogenes*
2004	Smoked salmon	Denmark	*L. monocytogenes*
1999	Smoked mussels	Tasmania	*L. monocytogenes*

The scale of the *Lm* problem is significant, and a web search for "smoked fish Listeria" reveals numerous recent recalls of smoked salmon in USA, Canada, Europe and South America.

Listeria illness from smoked seafoods

In Table 2 are some of the outbreaks of food poisoning caused by consuming smoked seafoods containing *Lm*.
In Australasia there have been three seafood-related outbreaks of listeriosis:

- Three healthy people aged 83, 37 and 10 years in Tasmania became ill with symptoms limited to the gastrointestinal tract. The illnesses, in 1991, followed consumption of New Zealand smoked mussels which had been illegally repackaged with use-by dates over 3 months beyond their original and had *Lm* >1,000,000/g (Misrachi *et al*., 1991; Mitchell, 1991; Eyles, 1994).

- Listeriosis involving smoked mussels occurred in New Zealand in 1992, when newborn twin babies died as a result of *Listeria* infection (Eyles, 1994). The mother had received medical advice to consume smoked mussels to increase her iron count, as she was somewhat anaemic and it is possible that her consumption rate was unusually high (Andrews & Young, 1993).
- In 2019, several elderly people with underlying health issues contracted listeriosis in Queensland, NSW and Victoria linked with consumption of smoked salmon manufactured in Tasmania; two died.
- Internationally, there have been several outbreaks of listeriosis from smoked fish.

Table 2: Selected cases of foodborne listeriosis associated with smoked seafood

Location (year)	Cases (deaths)	Food
Tasmania (1991)	3	Smoked mussels
New Zealand (1992)	3 (2)	Smoked mussels
Sweden (1994/95)	9 (2)	Smoked rainbow trout
Finland (1999-2000)	23 (4)	Vacuum packed cold-smoked trout
Denmark (2014)	6	Vacuum packed cold-smoked trout
Europe (2015-18)	12 (4)	Vacuum packed cold-smoked salmon
Denmark (2017)	5	Vacuum packed cold-smoked salmon
Qld, Victoria, NSW (2019)	4 (2)	Vacuum packed cold smoked salmon

2. What's the problem?

The problem for the industry is that *Lm* is common in the environment: in soil and in water, both marine and freshwater.
This means when you harvest fish, they will bring *Lm* with them into the processing plant and the pathogen is potentially able to end up in final product.
Surveys indicate how often *Lm* appears in smoked fish in the retail sector in Australia and UK, and these are summarised in Table 3.

Table 3: Prevalence of *L. monocytogenes* in Australian and UK seafood

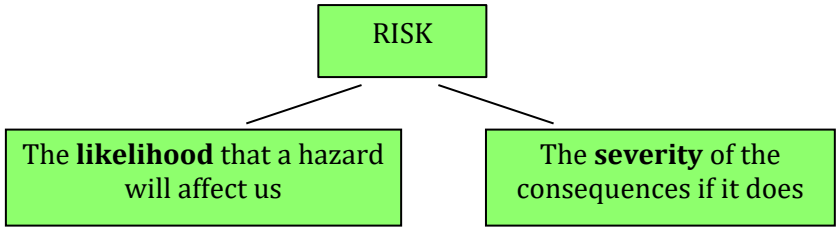
	Positive samples (%)	Reference
Smoked salmon fillets and slices in Tasmania	0.4	Garland (1995); Garland & Mellefont (1996)
Retail smoked fish and in Canberra	4.1	Rockliff & Millard (1996)
Retail smoked fish in Victoria	10	Dunn, Son & Stone (1998)
Retail smoked salmon in NSW	17.9	Arnold & Coble (1995)
Retail cold smoked fish in UK	17.4	Cited by NSWFA, 2009
Retail hot smoked fish in UK	3.4	Cited by NSWFA, 2009

As can be seen from Tables 1-3, *Lm* in CSS products is a global problem. In the Danish industry, Mejlholm *et al*. (2015) state: *"Initial cell concentrations of L. monocytogenes in naturally contaminated products are usually low, typically below the detection limit … with concentrations in the range of 7-10 cfu/g."*
Unfortunately it is possible that product with a low concentration of *Lm* leaving the factory will progress to an infective dose for vulnerable consumers if it is abused either by elevated temperature in the supply chain, or by extending shelf life past the use-by date.
Danish researchers (Mejlholm *et al*. 2010) have developed a tool called the Food Safety and Spoilage Predictor (FSSP) that can predict growth of *Lm* through the supply chain. The tool is accepted by FSANZ (2014) as a means for validating the ability of *Lm* to grow in CSS and HSS and is now in common use in the RTE food sector.

There are numerous inputs to the FSSP that need validation and these will be listed, together with the experimental details required In Section 6.

3. What's the risk?

Risk is made up of two parts: the likelihood that something bad (a hazard) will happen, plus the severity the hazard will have on us.



Who's most at risk?

If you produce a batch of product that contains *Lm*, how vulnerable are your customers? Fortunately, the majority of us are relatively resistant to pathogens because we have a range of natural body defence systems that:

- Kill pathogens in the stomach, where the acidity is high, a similar pH as battery acid
- Prevent pathogens attaching to the intestine, where they can set up home and damage us
- Reduce the likelihood that an infection will progress to become an illness

However, as shown in Table 4 many Australians are not so fortunate and are vulnerable to food infections because they have lowered immune systems: these are the young, old, pregnant and immune-compromised (YOPIs).

Table 4: Susceptible populations and proportions in Australian society (ABS data)

Population	Individuals	Percentage
Pregnancies	300,000	1.3
Neonates	330,000	1.4
Children 1-5	1,500,000	6.3
Elderly >65	3,500,000	14.6
Diabetes	1,700,000	7.1
Cancer Patients	400,000	1.7

Put all these categories together and you can see that vulnerable people make up more than 20% of your customers, and they are at the highest risk from consuming one of your products if it contains a hazard.

CSIRO tool

A very useful risk assessment tool was developed by Food Science Australia (FSA, 2000) where answering six questions will generate a qualitative risk assessment. When you put all the answers together you can make a risk rating – based on knowing about your process and about the hazards you're trying to manage.

Here's one way to go about getting the answers to the questions in the tool:

1. **Severity of the hazard.** The International Commission on Microbiological Specifications for Foods (ICMSF) describes the severity of illness according to three categories: Moderate, Serious and Severe, and classifies *Lm* as Severe.
2. **Likelihood** that the hazard will be present in the final product can be determined from your own microbiological testing or from surveys undertaken in Australia in Table 3.
3. **Growth required** to reach an infective dose is determined by assessing whether the storage conditions will allow growth. For example, *Listeria* can grow under refrigeration and if the product has long shelf life, the pathogen may reach an infective dose.
4. **Effect of processing** takes into account the extent to which product is heated. Hot smoked fish receives the *Listeria* cook (a CCP), designed to reduce 1,000,000 *Lm* in the raw fish to less than one in the smoked product. Cold smoking on the other hand will actually allow growth of *Lm*.
5. **Consumer cooking step** – smoked salmon is almost always consumed RTE so no further kill step is involved.
6. **Epidemiological links** – has the pathogen:product pairing caused outbreaks of food poisoning in Australia, or anywhere else in the world? You can see from Table 2 that it has caused illness and deaths from consumption of smoked fish.

In the Tables 5 and 6, a qualitative risk assessment is made for vulnerable consumers consuming cold and hot smoked salmon:

Table 5 : Risk of contracting listeriosis in vulnerable consumers from CSS

Product	**Cold smoked salmon**
Hazard	***L. monocytogenes***
Severity	Severe
Likelihood	Moderate *L. monocytogenes* will probably be on raw fish entering the process
Growth required to reach infective dose	Yes, but this may not be high for vulnerable consumers (ca 5000 cells)
Effect of processing	Freezing may reduce the concentration of *Listeria*. Chilled storage over 35-day shelf life allows growth to an infective level
Consumer cooking step	None
Epidemiological links	Yes. There are documented cases of listeriosis from CSS
Risk rating	**High**

Table 6: Risk of contracting listeriosis in vulnerable consumers from HSS

Product	**Hot smoked salmon**
Hazard	***L. monocytogenes***
Severity	Severe
Likelihood	Moderate *L. monocytogenes* will probably be on raw fish entering the process
Growth required to reach infective dose	Yes, and this may reach a high count (> 1000 cfu/g) because of reduced competition
Effect of processing	*Listeria* cook will eliminate the pathogen but recontamination is possible during slicing and packing. Freezing may reduce the concentration of *Listeria*. Storage over 35-day shelf life allows growth to an infective level
Consumer cooking step	None
Epidemiological links	Yes. There are documented cases of listeriosis from HSS
Risk rating	**High**

4. What's the solution?

The solution comes in several parts:

1. Identify and operate a Critical Control Point (CCP) for CSS by ensuring it remains at a very low level through the storage life.
2. Arrange product flow so CSS is not able to contact HSS during slicing and packing, thereby reducing the likelihood of recontamination.
3. Operate a *Listeria* cook as a CCP for HSS to eliminate the pathogen.
4. Arrange product flow so that HSS is not re-contaminated with *Lm* during slicing and packing.

Section 5: *What does a good smoked fish smoking process look like?* contains a series of checklists that can be used to assess how well your business conforms with a high standard of *Listeria* control.

The intention of the checklists is to present elements of process control that businesses, the controlling authority and auditors can use as opportunities for improvement.

5. What does a good fish smoking process look like?

There are many prerequisites for a good fish smoking process and we list them here under five main headings:

1. Skills and knowledge of the managers and operators
2. Properly constructed premises and good manufacturing practices
3. Effective cleaning system
4. A validated Hazard Analysis Critical Control Point (HACCP) system
5. Test results on product and premises to verify that the business is operating safely

Within each of these headings are numerous criteria that indicate how completely the requirements are being met and, by difference, any weak points in the food safety system.

These criteria have been developed in the aftermath of large outbreaks of listeriosis in the United States in 1999 and 2000 where technologist, Bruce Tompkin developed a series of criteria for evaluating how well a business is set up to manage the pathogen (Tompkin, 1999, 2002).

Following food poisoning outbreaks from smallgoods in South Australia, these criteria were refined for use in that State (Sumner, 2004; 2005) and have been further refined here for use in smoked fish businesses.

i. Skills and knowledge of the managers and operators

Regulatory requirement

The Food Standards Code (FSC) and the Primary Production and Processing Standard (PPPS) for seafood, Standard 4.2.1, and Standard 3.2.2 (Food Safety Practices and General Requirements) state that a seafood business must ensure that seafood handlers have:

a) *Skills in food safety and food hygiene; and*
b) *Knowledge of food safety and food hygiene matters commensurate with their work activities*

You should document the skills and knowledge base of your staff to check whether they are appropriate for a high-risk RTE food like smoked fish. The format of Table 7 is one way to document staff training.

Table 7: Typical description of skills and knowledge base of the company

	Person	Position	Qualifications	Experience
Technical	(name)	Technical Manager	BSc (UTas) Micro	15 years food business
QA	(name)	QA Manager	BSc (Melb) Zoology	8 years QA with us
	(name)	Lab Manager	BSc (UTas) Chemistry	2 years with us
	(name)	Lab Assistant	Certificate 3	14 years in QA
Supervisor	(name)	Filleting room	HACCP course	10 years with us
Operators	(name)	Slice/pack room	HACCP course	8 years with us
	(name)	Freezer store	HACCP course	4 years with us
	(name)	operators	In-house training	

ii. Properly constructed premises and good manufacturing practices

Regulatory requirement

The Primary Production and Processing Standard (PPPS) for seafood, Standard 4.2.1, states that:
(1) A seafood business must ensure that seafood premises, including live seafood premises, and equipment used in the primary production of seafood are –
(a) so far as is reasonably necessary, kept clean; and
(b) designed, constructed, maintained and operated such that the safety or suitability of the seafood will not be adversely affected.
Food Standards Code (FSC) Standard 3.2.3 *(Food Premises and Equipment)* provides considerable detail on all aspects of construction and hygienic operation.

You're manufacturing RTE foods and the premises need to be constructed and operated as far as possible to maintain a *Listeria* free section after any kill step that eliminates the organism, such as hot smoking.
The checklist prompts you to assess how well your business aligns with the requirements of the Food Standards Code under three headings:

- Factory layout and operation
- Entry points
- Packing room construction and operation

iii. Effective cleaning system

Regulatory requirement

Food Standards Code (FSC) Standard 3.2.2 (Food Safety Practices and General Requirements) states that:

(1) A food business must ensure the following equipment is in a clean and sanitary condition set out below –
  a) eating and drinking utensils – immediately before each use; and
  b) the food contact surfaces of equipment – whenever food that will come into contact with the surface is likely to be contaminated
(2) In subclause (1) a 'clean and sanitary condition' means, in relation to a surface or utensil, the condition of a surface or utensil where it –
  a) is clean; and
  b) has had applied to it heat or chemicals, heat and chemicals, or other processes, so that the number of micro-organisms on the surface or utensil has been reduced to a level that –
    I. does not compromise the safety of the food with which it may come into contact; and
    II. does not permit the transmission of infectious disease.

The checklist will prompt you to assess how well your business aligns with the requirements of the Food Standards Code under three headings:

- Written program – are instructions clearly written for the cleaning crew?
- Specific cleaning programs – high risk areas like drains, coolrooms and air conditioning units can be persistent sources of *Listeria* and require specific programs
- Monitoring of cleaning

There are three elements to a successful Food Safety Program (FSP), two of which (GMPs and SSOPs) must be implemented before a HACCP plan can be devised and operated.
Completing checklists for the previous three aspects will indicate whether your business has the solid foundation to underpin HACCP.

iv. A validated Hazard Analysis Critical Control Point (HACCP) system

Regulatory requirement

Food Standards Code (FSC) Standard 3.2.1 (Food Safety Programs) sets out how a FSP must be developed in accord with HACCP principles, and audited.
There are three CCPs that the business must first validate, and then monitor for each batch of production:

- **Hot smoking (Temperature:time of the process)**

In hot smoking the process must be designed so that, if 1,000,000 *Lm* were present at the site of microbiological concern, smoking would reduce the population to less than one. This is called a 6-D process and times and temperatures to deliver this for *Lm* are shown in Table 8. The CCP is a stage that eliminates the hazard.

Table 8: Temperature:time combinations that eliminate *Lm* from smoked fish

Temperature (°C)	Time (min)
60	44
61	33
62	24
63	18
64	13
65	10
66	7
67	6
68	4
69	3
70-72	2

You can validate your process by inserting a probe at the slowest heating point of the largest piece of fish and putting that piece at the slowest heating part of your smoker oven.

- **Cold smoking (addition of acidity regulator)**

It has been known for some time that organic acids (such as lactic and acetic) inhibit the growth of *Lm* and they are in common use in the RTE meat industry in products such as vacuum packed sliced ham.
Organic acids have also been shown to inhibit *Lm* in vacuum packed smoked fish without adverse effect on its sensory quality (Vogel *et al*. 2006). In a challenge test on fillets seeded with Lm, the researchers injected a proprietary brand of lactate and diacetate into the fillets, which were then cold smoked and stored over 42 days at 10°C; no growth of Lm occurred.
In Scandinavian countries, the use of organic acid (usually lactate and/or diacetate) at the curing stage of salmon processing has been commercialised as indicated in Figure 1.

Figure 1: Use of Acidity Regulator E 262 (Sodium diacetate) in Danish smoked salmon on retail sale in Australia.

The effectiveness of these ingredients can be assessed using the FSSP; if used at the correct concentration they inhibit growth of *Lm* throughout the supply chain, so if 10 *Lm* were able to enter a retail pack, there would still be only 10 at the end of shelf life.

- **Cold smoking (Option 2: Use of biopreservative cultures)**

In 1998, Danish researchers showed that the background bacterial flora inhibited the growth of *Lm* in CSS (Paludan-Muller *et al.* 1998; Huss *et al*. 1988). These are the "good" bacteria that are in large numbers in yoghurt, and may have a probiotic effect in our intestines.
The same research team at the Danish Institute for Fisheries Research then showed that Lactic Acid Bacteria (LABs) could inhibit growth of *Lm* and also reduce its numbers on CSS (Nilsson *et al*. 1999; 2004).
In Europe, the technology is used by a small proportion of RTE meat and smoked fish businesses. However, use of bioprotective cultures or any other technology in the Australian industry would require rigorous R&D to validate that growth of *Lm* is prevented on products sliced on high-speed machinery.

- **Cooling of smoked product**

As shown in Table 9, Food Standard 3.2.2 specifies a two-stage cooling process, making cooling an RCP and a CCP. It can be validated by inserting a probe at the site of microbiological concern (the slowest cooling spot).

Table 9: Two-stage cooling process (Standard 3.2.2)

	Temperature range (°C)	Time (hours)
Stage 1	60-21	2
Stage 2	21-5	4

Test results on product and premises that verify the business is operating safely

Regulatory requirement

Standard 3.2.2 (Food Safety Practices and General Requirements) states that the food safety program: *Provides for appropriate records to be made and kept by the food business demonstrating action taken in relation to, or compliance with, the food safety program.*

Appendix 3 provides background information on what you should test, the frequency of testing and the bacteria to be monitored.
In summary, a business should build a database of indicator bacteria at three stages:

- Incoming raw material (fish, fillets)
- Sliced product
- Final packaged product – chilled and frozen

After the initial database has been established, testing frequency can be adjusted according to the volume of production, which may involve consultation with your controlling authority.

6. Checklists for assessing standard of *Listeria* management

Note that these checklists are intended to assist in process and product improvement, they are not meant to be prescriptive.

Checklist for skills and knowledge

What to check	Yes	No	Comment
1. In a large operation is there a technical person qualified at degree level?			
2. Are key managers (QA, Operations) experienced?			
3. Have operators and supervisors completed HACCP training?			
4. Are operators enrolled in Certificate level training?			
5. Has the cleaning crew been trained?			
6. Is there a training record?			

Checklist for premises and their operation

What to check	Yes	No	Comment
Factory layout and operation			
1. Is there a one-way flow from raw to cooked product?			
2. Are cooked areas separated from raw areas?			
3. Is the demarcation clear e.g. different coloured flooring?			
4. Are operators confined to their work areas?			
5. Do operators have distinguishing uniforms/head coverings?			
Entry points			
1. Are there effective hygiene barriers at each entry point?			

2. Is entry to packing room guarded by a deep sanitiser footbath that cannot be skirted?
3. Is the sanitiser level monitored and maintained at the required level?
4. Does the entry room to clean area have hand washing/sanitising facilities?
5. Do operators don clean protective clothing on every occasion they enter packing room?
6. Do maintenance personnel observe same entry protocol as operators?
7. Do tools used by maintenance personnel remain in cooked area?

Packing room construction and operation

1. Is the packing area dry?
2. Is condensation from shrink tunnels removed effectively?
3. Is there positive air pressure in the packing room?
4. Are walls and floors free of cracks?
5. Are there open drains?
6. Do drains flow from the cooked to the raw side?
7. Is back siphoning of drains prevented?
8. Are overhead structures that might harbour dust and bacteria absent?
9. Are air conditioning units positioned away from packing tables?
10. Are forklifts absent from the packing room?
11. Do conveyors have solid rollers?
12. Do conveyors have belts of impermeable materials?
13. Is equipment with rusting or hollow framework absent?
14. Is damp insulation absent?
15. Are door seals in good condition?

16. Are on/off switches capable of being cleaned?
17. Are motor housings capable of being cleaned?
18. Are listericidal sanitiser granules used on floors?

Checklist for the cleaning program and its monitoring

What to check	Yes	No	Comment

Written program

1. Is there a written cleaning program that is appropriate?
2. Does the program specify how each part of the building and piece of equipment is cleaned?
3. Is the frequency of cleaning stipulated: daily and periodic cleaning?
4. Has the cleaning team been trained and the training program documented?

Specific cleaning programs

1. Drains - specific program in addition to daily clean
2. Linings (floors, walls, overheads) - specific program in addition to daily clean
3. Cool rooms - specific program in addition to daily clean
4. Air conditioning units – weekly clean and fogging with QUAT
5. Containers - post-process clean
6. Slicers - post-process clean
7. Slicers – clean during work breaks
8. Packing machines - post-process clean
9. Packing machines – clean during work breaks
10. Tables - post-process clean

11. Conveyors - post-process clean
12. Racks - post-process clean
13. Trolleys – post-process clean

Cleaning chemicals and their use

1. Is equipment for applying and rinsing cleaning solutions adequate?
2. In large plants is there a ring main and drop points at appropriate locations?
3. Are low pressure:low volume foaming systems used?
4. Are cleaning chemicals purchased from a reputable supplier?
5. Is detergent appropriate for the soils generated – a chlorinated alkali?
6. Is the sanitiser listericidal – a QUAT?
7. Are correct concentrations used – clear operator instructions?
8. In small plants is the measurement of cleaning fluids appropriate - marked jugs etc?
9. Is cleaning equipment properly stored - separate room, chemicals locked and bunded?
10. Is advice sought from the cleaning specialist?
11. Is the advice documented?

Breaking the *Listeria* cycle

1. Is there a contingency plan for heating slicing machines to inactivate persistent colonisation?
2. Is there a plan for eliminating *Listeria* from chillers by heating them?

Monitoring of cleaning

1. Are packing room surfaces tested for presence of *Listeria*?
2. Are cleaned working surfaces tested for residual contamination?
3. Is the result assessed against the Australian Standard for cleaning effectiveness

4. Are there corrective actions that target positive detection of *Listeria*?

Checklist for a validated Hazard Analysis Critical Control Point (HACCP) system

What to check	Yes	No	Comment

Identification and validation of CCPs

1. Has the business identified CCPs using the Codex definition (prevent, eliminate or reduce the hazard to an acceptable level)?
2. Has the business identified a CCP for hot smoking?
3. Is the CCP based on a 6D *Listeria* inactivation of 65°C/10 minutes or equivalent lethality at an alternative temperature and time?
4. Was the validation done correctly – temperature:time recorded at the site of microbiological concern – the thermal centre?
5. Has the cooling process been validated against the FSC standard: cool from 60°C to 21°C within 2 hours, then cool to 5°C within a further 4 hours?
6. Are there records for cooling of every batch?
7. Is the validation documented?
8. Has the business identified a CCP for cold smoking?
9. Is the CCP based on the combined hurdle effects in the product of:
   - Storage temperature
   - Storage time
   - pH
   - Salt level
   - Organic acid level
   - Phenol level

or

- Use of bioprotective cultures

10. Was the CCP validated by use of the Food Safety and Spoilage Predictor (FSSP)?

Monitoring of CCPs

1. Is the thermal process monitored and documented for every batch of HSS?
2. Is the corrective action appropriate for an inadequate thermal process?
3. Is the cooling process monitored and documented for every batch of HSS?
4. Is the corrective action appropriate for an inadequate cooling process?
5. For each batch of CSS are the following parameters monitored:
   - Salt
   - Organic acid
   - pH
   - Phenol (if used in the validation)
6. Is the corrective action appropriate if the chemical composition of the product allows the growth of *Listeria*?

Checklist for monitoring and test results

What to check	Yes	No	Comment

Product testing

1. Is there a microbiological database for finished products (HSS and CSS, chilled and frozen)?
2. Does the database include testing at intervals over the shelf life?
3. Do the organisms include Total bacterial count and Lactic acid bacteria count?
4. Is the incubation temperature and time appropriate for a long shelf life chilled product (25°C for 96 hours)?
5. Does the business test end-of-shelf life product for *Listeria*?

7. If *Listeria* is detected is the concentration established?

Testing of food contact surfaces

1. Are slicing and packing machines tested during production for the presence of *Listeria*?
2. Is corrective action effective for dealing with HSS products processed through machines found to be *Listeria*-positive?

7. How to develop a CCP for cold smoked fish

In 2014, Australia's standards-setting body, FSANZ provided manufacturers with the option to change formulations for RTE foods to prevent growth occurring if *Lm* contaminates the final product, for example during slicing and packing.
The decision gives you two options:

- Option 1: If your product allows the growth of *Lm* then the recall limit is *"not detected in a 25g sample".*
- Option 2: If *Lm* can't grow over the product's shelf life, the recall level is at 100/g – that's 2,500 times more leeway from the manufacturer's viewpoint and reflects the reduced risk from products that don't allow growth of *Lm*.

As indicated previously, CSS as currently manufactured by Tasmanian businesses does not have a CCP for *Lm* and, given its long shelf life, CSS will support the growth of *Lm* during storage, retailing and home consumption.
If you wish to make your CSS "bullet proof" to the growth of *Lm* you will need to prove this to the controlling authority. You can do this by collecting data (see the list 1-7 below) and inserting them into a spreadsheet tool.
The tool was developed by scientists in Denmark, with U Tas playing an important role, and is an alternative to challenge testing. It's a piece of software into which you enter a number of key parameters about your product and it predicts how long the growth of *Lm* is prevented and slowed.
This tool is part of the Food Spoilage and Safety Predictor (*FSSP)* software package (Mejlholm & Dalgaard, 2009). The *Lm* Growth Model has also been peer reviewed by a team of international experts (Mejlholm *et al.* 2010). The *FSSP* is currently used in the seafood industry, and in other industries where *Lm* affects safety of RTE products.

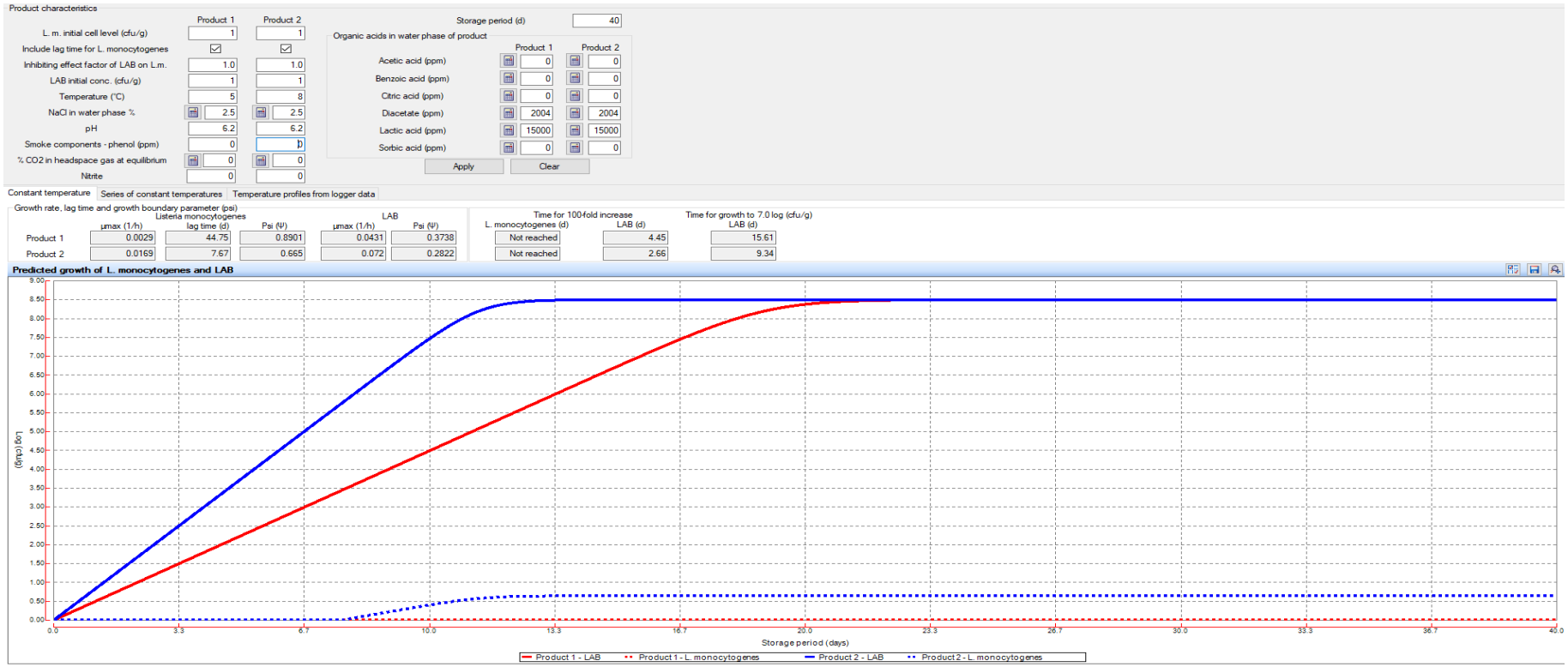
Validating that CSS will not support the growth of *Lm*

As you use the FSSP, the image on your computer screen (Figure 2) shows you what you need to know about your product:

1. Storage temperature
2. Shelf life on your label
3. Salt content in the water phase
4. pH of the product
5. Lactic acid in the water phase
6. Diacetate in the water phase
7. The number of lactic acid bacteria in product at beginning of retail

You'll see some actual data entered into the tool from two batches of vacuum packed CSS packed in modified atmosphere (MAP). Both batches have the same formulation (2.5% salt in the water phase and some organic acids added at salting.). The difference is that one batch was stored at 5°C and the other at the mildly abusive temperature of 8°C. The Predictor was set at an initial level of one *Lm* cell/g of CSS. At 5°C (red lines) there was no growth of *Lm* over 40 days and at 8°C (blue lines) growth of *Lm* was about 4 cfu/g – a level that is highly unlikely to cause listeriosis even in vulnerable consumers. While organic acid has had a significant effect on *Lm*, you'll also see huge growth of Lactic acid bacteria, which also have an inhibitory effect on *Lm*.
Clearly, these parameters give a very safe product, even when stored at 8°C. However, you'll need to prove that your process delivers the correct salt in water, lactate and diacetate concentrations. You can do this by sending samples (five is a good number) of the thickest part of the fillet to a lab that is able to analyse for these parameters.

Figure 2: Growth of *L. monocytogenes* at 5°C and 8°C in CSS to which a proprietary brand of Potassium lactate and Sodium diacetate has been added



Appendix 1: CPs, RCPs, CCPs and GMPs in smoked fish processing

There are a number of unit operations in smoked fish processing that are Control Points, Regulatory Control Points or Critical Control Points, or that rely on Good Manufacturing Practices (GMPs):

1. Gilling and gutting
2. Active chilling of fish and chilled storage until filleting
3. Filleting and bone removal
4. Salting
5. Smoking
6. Chilling
7. Slicing
8. Packing
9. Freezing
10. Thawing and chilled storage

Let's be sure of the definitions for RCPs, CCP, and CPs.

A CCP must achieve one of the following:

1. Prevent the hazard, or
2. Eliminate the hazard from the product, or
3. Reduce the hazard to an acceptable level

A Regulatory Control Points (RCPs), are "must do" controls specified in Standards and Regulations e.g. receiving fish at no warmer than 5°C.

A Control Points (CP) reduces the impact of the pathogen but doesn't do the job as completely as a CCP.

Individual businesses may have different unit operations, so let's consider each of the above operations thinking only in terms of food safety – quality is also important but food safety is the main game here.

1. Gilling and gutting

In large operations this is done mechanically, and is a CP for correct removal of specified organs such as gills, gut and head) without damaging the edible portion of the fish. This may be done either at the harvest site or at the processing plant.

2. Active chilling and chilled storage

Active chilling is when refrigeration is applied to remove heat from a harvested fish and reduces their temperature to 5°C or below as specified by PPPS 4.2.1. Active chilling of fish bodies is therefore an RCP.

3. Filleting and bone removal

Whether done by hand, filleting and pin bone removal aims to result in two fillets devoid of bones. Bones are a physical hazard and are treated by processors as a CP.

4. Salting

Salt is added either by dry salting, brine injection or immersion in brine. Salt dissolves in the water phase of the fillet. Water phase salt (WPS) around 4% is inhibitory to some bacteria, but not to *Lm*, which is salt tolerant, making salting a CP.

5. Smoking

Fillets are smoked either on racks, or hanging in an atmosphere of gases liberated by burning of wood chips. Fillets take up flavours and odours from the smoke, as well as phenols, which are antimicrobial.
The temperature of smoking varies:

- In cold smoking fillets are held at 25-30°C and is a CP
- In hot smoking fillets are held at a temperature:time regime that delivers a "*Listeria* cook" (reduction of 1,000,000 cells of *Lm* to less than one cell) and is a CCP.

6. Pre-slice chilling

After smoking fillets are moved to a pre-slice chiller in order to reduce the temperature according to Standard 3.2.2; the process is an RCP and a CCP.

7. Slicing

Chilled fillets are sliced in various ways to produce final products that range from thick steaks to thin slices.
The process is a GMP.

8. Packing

Packing involves sliding product onto a backing plate and vacuum sealing it within a bag.
The process is a GMP.

9. Freezing

When water in the product is changed to ice crystals the WPS increases, causing osmotic shock and inactivation of *Lm*. Freezing has been shown reduce the population of Lm in CSS by >90%. (Yoon *et al*. 2004) but since *Lm* may not be eliminated or reduced to an acceptable level the freezing stage is a CP.

10. Thawing and chilled storage

A proportion of frozen, vacuum-packed fish is thawed and retailed in chilled retail display. Any surviving Lm will now be able to increase their population because they can grow under the low oxygen levels present in vacuum packs.
Growth is dictated by temperature of storage and the length of time prior to consumption but the Food Standards Code requires storage no warmer than 5°C making storage an RCP.

Summary

Operation	CCP, RCP or CP
Gilling and gutting	CP
Active chilling of fish, and chilled storage until filleting	RCP
Filleting and bone removal	CP
Salting	CP
Cold smoking	CP
Hot smoking	CCP
Chilling	RCP/CCP
Slicing	GMP
Packing	GMP
Freezing and frozen storage	CP
Thawing and chilled storage	RCP

Appendix 2: Keeping the plant clean

How you keep your operation clean during the processing day, and how you clean it at the end of processing is an integral part of your Food Safety Plan – without a clean plant you can't operate your HACCP plan effectively.

Elements of a cleaning program

There are two distinct parts:

1. Cleaning - removal of all soils from equipment and working surfaces
2. Sanitising – inactivation of any microbes remaining on or in equipment

How to clean the seafood plant

There are three ways to do this depending on the size of your establishment:

1. Dry cleaning

This is obviously done without the hose – by scraping, brushing, wiping or vacuuming product and soils from floors and equipment. It can be done throughout the processing day (part of "clean-as-you-go") and is the first stage in end-of-day cleaning.
Equipment needed: Brushes, scrapers, dustpans, squeegees, plus a safe, clean place to locate them. Colour coding is useful, and equipment needs to be stored safely in a specific place.

2. Manual cleaning

Some equipment needs taking to pieces before you can clean it. After disassembly you'll need to remove soils in a bath of cleaning solution.
Equipment needed: In small-scale plants this is "bucket-and-brush" cleaning but in medium and large plants mobile spray units are used to deliver cleaning solution as a foam which clings to the equipment and facilitates soil removal by brushing.

3. Cleaning-in-place (CIP)

Some pieces of equipment can be cleaned by passing cleaning solutions through the internals of pieces of equipment such as brine tanks and lines involving little disassembly.
Equipment needed: Bulk cleaning solutions, often with a permanent ring main system to link with individual pieces of equipment, coupled with software programs to deliver validated cleaning times and solution temperatures. Cleaning is enhanced by spray balls and other equipment designed to deliver agitation to hard-to-get parts of equipment.

Soils in the seafood plant and cleaning solutions needed to remove them

Protein and fat are components of fish that form soils on equipment during the processing day, and for each of which there is a chemical that can dissolve and remove them. The most commonly used cleaning solution (detergent) is a chlorinated alkali based on caustic soda and sodium hypochlorite (hypo), which will remove fat and protein. Contact time and temperature are important - very hot solutions can cause "bake-on" and cleaning solutions are usually delivered at warm (40-50°C) temperatures. Slime is also present on fish as they enter the filleting process and this is removed from equipment by using chlorinated alkali detergent.

If you live in a hard water area your supplier may recommend additions to your detergent to improve its effectiveness such as surfactants, builders and water conditioners. And water isn't cheap, so the less you use the better.

Sanitisers

A sanitiser is a chemical that reduces the level of microbial contamination on the surfaces of food equipment. To qualify as a sanitiser the chemical must kill 99.999% of *E. coli* and *S. aureus* in 30 seconds at 25°C – this is done as a laboratory test.
Sanitisers used in cleaning seafood plants include:

1. Chlorine (usually Sodium Hypochlorite – Hypo)
2. Quaternary Ammonium Compounds (QUATs)
3. Acid sanitisers
4. Organic acids (such as Acetic Acid)
5. Peroxyacetic acid

They all have advantages and disadvantages that you need to consider in discussion with your cleaning supplier.

Delivering cleaning solutions

You need to follow five steps when you clean the plant at end of processing:

1. Dry clean – remove as much soil as possible from equipment, floors and walls
2. Rinse equipment and surfaces with cold water
3. Deliver detergent solution either with manual and/or CIP to equipment
4. Rinse with water
5. Deliver sanitiser solution either with manual and/or CIP to equipment

Applying cleaning solutions is usually done with low pressure and low volume foaming wands - high-pressure pumps only blast soils and solutions all over the plant, creating aerosols and cross contaminating premises. Typically, detergents are foamed onto surfaces and left for around 15 minutes (contact time) while the chemical reactions take place so that all the soil reacts with the detergent. Sanitisers are also foamed and left for the correct contact time needed for bacterial inactivation.

Having a cleaning plan and people trained to follow it

Your cleaners need a written plan, plus training on how to carry it out and to have sufficient time for the job.
Chemical safety is also important:

- For large operations chemicals need to be stored in a lockable room or caged area that is protected by bunds (low walls) to contain leaks; small to medium food businesses will need a lockable room or cabinet.
- Operators require training on using cleaning chemicals safely and what to do if they have an accident.
- They also need personal protection equipment and to have the material data safety sheet (MSDS) available.

Choosing systems and cleaning solutions

Reputable suppliers of cleaning chemicals are as much concerned with setting their customer's business up properly as they are with selling drums of soap. Producers can expect a number of 'add-ons' from chemical suppliers such as:

- Training the cleaning crew, both in technique and Work Health & Safety (WH&S) (concentrated cleaning chemicals are dangerous)
- Trialling cleaning solutions and reporting on their effectiveness
- Providing work instructions on how to clean different equipment and areas
- Working out a cleaning budget

- Assessing the effectiveness of the cleandown

These services can be valuable where you don't have resources to deliver staff training, and where you haven't the capacity to verify the effectiveness of the cleaning regime. The additional service may also provide the information you need to be confident your chemical usage is appropriate for your needs.

Some do's and don'ts

- Don't use porous and absorbent items like rags or wooden handled tools - they harbour bacteria.
- Do use separate brushes for product and non-product surfaces - colour-coded is good e.g. red means only use for floor waste, green is used for surfaces that may come into contact with product.
- Do sanitise brushes and store them correctly between use.
- Do use low pressure:low volume cleaning systems to minimise splashing and aerosols.
- Do store hoses on reels or racks.
- Do examine the blowers in the cool room to ensure they are not dusty or dripping water.
- Always do a 'pre-op' inspection before work is started. Check whether surfaces and equipment are clean and, if they aren't, do a clean down and sanitise. This will slow operations and you'll want to find out why it wasn't done properly first time.

How well did we do?

You've cleaned your plant and you've paid for your cleaners, cleaning chemicals and water, now you need to know whether the job's been done right.
Here's how:

1. Walk around and look for any remaining soils
2. Look at stainless equipment: if you see a rainbow-like sheen this tells you protein hasn't been removed – you're not delivering the right chemicals
3. Assess whether there's any organic material left – if there is, the job's not done properly. You can assess this in real time by investing in:
   - Swabs which tell you whether protein remains
   - An ATP bioluminescence kit which tells you whether microbes, fat, protein or sugar remains
4. You can also swab surfaces and send the swab to a laboratory for a bacterial count and wait two or more days for the result.
5. When you get the result make sure you document it so your auditor can assess the effectiveness of your cleaning system, and you also give your cleaners feedback on how well they went.

Prerequisites for a successful cleaning program include having a plant that is properly designed for cleaning and product flow, and then monitoring the plant environment to determine whether your cleaners have done the job properly.

Environmental monitoring

As well as ensuring your premises are clean, you are required to monitor whether you are keeping them free of pathogens, especially *Lm*. As part of your FSP you will need to agree with the controlling authority a regime for testing your premises – frequency and location of testing.
You also divide your premises into zones:

Zone A: Product contact surfaces e.g. slicing machines, tables on which you cut fillets into portions

Zone B: Surfaces that don't contact product, but are close to them e.g. switches, handles
Zone C: Distant surfaces from product contact e.g. drains, air conditioning units, walls and floors
Zone D: Surfaces outside the processing area e.g. corridors from smoking ovens
If your monitoring program detects *Lm* in Zone A, such as on slicing or packing equipment for HSS, you're into Corrective Action that will include intensive cleaning of the equipment and a clearance program for any product affected by the monitoring result.
Contamination in the other zones will involve Corrective Actions such as additional cleaning and testing to confirm the zone has been freed of *Lm*.

Appendix 3: Microbiological testing

There are numerous bacteria – both spoilage and pathogenic (disease-causing) that need to be controlled in your business and there are several stages in your operation where you need to know you're controlling them:

1. On food contact surfaces after cleaning and sanitising the plant
2. In raw fish
3. On food contact surfaces during the process – on slicing machines that are in use
4. In finished products – after packing and frozen storage

In this section we focus on the testing needed to give you confidence that your incoming raw materials and process parameters will consistently result in finished products of acceptable hygienic quality.

Microbiological testing – pluses and minuses

Microbiological testing is a useful tool in monitoring your process because it can tell you whether certain bacteria are present and, in some cases, how many are in the product. The bad news is that micro testing is not cheap and you probably have a 2-3 day wait before you get the result. Also, counting bacteria in foods is not very accurate and most food microbiologists would consider a count of 10,000/g of product to be very similar to one of 50,000/g.

For these reasons we always look for information which is as real time as possible, and to use it to replace micro testing.

Sampling – lots to think about

In 1959 the Pillsbury Company received a call from the US Armed Forces asking: *"Can Pillsbury produce foods for consumption in the zero gravity of space capsules?"* The answer was yes, but to increase the certainty that no pathogens would be present, the sampling rate was so high that only a small portion of very expensive space food remained for the astronauts after the lab had done its job.

It's history now that HACCP was invented to cut down on micro testing and it also stimulated microbiologists to do the maths on how many samples are needed to pick up a pathogen like *Lm* in a food product – we'll cover this later.

How effective is end product testing for pathogens?

We worry about pathogens like *Listeria* in smoked fish and sometimes find it reassuring when the laboratory reports that they were not detected in the sample we submitted. We shouldn't be over confident however because pathogens are difficult to find as they are often not spread homogeneously through the lot, and the sample units you choose may not contain them.

Kornacki (2006) states: *"Finished product testing cannot be relied upon as the sole determinant of a Listeria-free product. No amount of finished product sampling and testing short of assaying the entire product with a perfect method can guarantee that the product is Listeria-free. Finding a problem through finished product testing is likely in situations where the incidence of product contamination is high."*

In the following table, Kornacki provides the sample numbers you need to take to be confident of finding the pathogen, if it is present in the lot of production.

Suppose you test a day's production of CSS for *Lm*, and let's assume that it's only present in a tiny proportion of the lot – let's say 0.1% is contaminated. You need over 4600 samples to be 99% certain of finding one *Lm* cell – you can't afford that. Ironically, if the product is more contaminated you need fewer samples to be confident of finding it.

Note that, if a customer or your controlling authority includes pathogens in their specifications, you'll need to sample according to their instructions - it's a cost of doing business.

Number of units in a batch to be tested to detect one or more positive *Lm* in the production lot

	Number of samples to be tested		
Percent positive	*90% confidence*	*95% confidence*	*99% confidence*
1	230	299	461
0.1	2303	2996	4605

How frequently should I test my products?

If you're a small manufacturer you and the controlling authority will want to have confidence in your products and here are some principles:

1. If you're starting up with a new product it makes sense to test a number of early batches (say the first 10 batches that you manufacture).
2. Then you must test at the minimum frequency that demonstrates ongoing process control, as set out by your controlling authority.
3. If, over time, you can demonstrate a very high standard of product hygiene you have strong supporting evidence to reduce the frequency of testing.

If you're a large manufacturer you'll probably test a sample from each product type you manufacture more frequently because of the increased exposure and therefore risk.

Sampling and your business's risk profile

Key questions for working out your testing program include:

1 How often should I test?
2 How many samples should I take?
3 Which organisms should I test for?

Earlier we did risk ratings for *Lm* in cold smoked and in hot smoked salmon so you know you're selling products with significant risk for vulnerable consumers. Another factor affecting risk is your volume of production.

> *Question: Who has the higher risk – a manufacturer making 5000kg/day or one that makes 50kg/week (all made on one day) and assuming both manufacturers have systems to minimise the opportunity for Listeria to get into the final product?*
> *Answer: The large manufacturer, because there are many more of their units in the market place leading to a greater chance that a vulnerable person will consume the product.*

So, a large manufacturer will need to have a much more intensive sampling program to:

1 Monitor the end-of-day cleandown
2 Monitor food contact surfaces during production
3 Test final product for *Lm*

This is an example of the way you need to think about testing and to develop a regime for the frequency of testing for each of your product types.

Which micro tests are useful for product testing?

Pathogen testing

You will need to do pathogen testing according to Food Standards Code Standard 1.6.1 at a frequency depending on your volume of production. You should consult your controlling authority for advice on frequency of testing.

Testing for Indicator bacteria

Indicator bacteria are useful organisms to have in a testing program because you get a result more quickly and more cheaply than testing for a pathogen. The most useful indicator test for smoked fish is the total bacterial count (also known as Total Plate Count, TPC; Total Viable Count, TVC; Standard Plate Count, SPC and Aerobic Plate Count, APC). This count is an indicator of the general population of bacteria in a product or on a food contact surface and the result is usually expressed as the number of colony forming units (cfu) per gram of product or per cm^2 of food contact surface.

What should I test to support process control?

There are three stages when some information is useful:

- Incoming raw material
- Sliced product in process
- Final product - chilled or frozen

Shelf life testing

To monitor the bacterial levels through the shelf life of your product, you can test for indicator bacteria via APC and for LABs. It is important to remember that because you've stored product under refrigeration for several weeks, the dominant microflora will be psychrotrophic bacteria.

Psychrotrophs generally have a temperature optimum of 15-25°C and a maximum growth temperature of 30-35°C (ICMSF, 1996) and it is logical to incubate cultures near their optimum (25°C) for sufficient time (4 days) so the colonies are clearly visible and therefore countable on the culture plate.

The importance of using the correct incubation temperature in monitoring shelf life studies was illustrated by Pothakos *et al.* (2012) who incubated plate counts of stored food samples at either 22°C/5 days or 30°C/3 days and found that counts from the former temperature were 0.5 – 3 log cfu/g higher.

The authors concluded*: "This study highlights the potential fallacy of the total aerobic mesophilic count as a reference shelf life parameter for chilled food products as it can often underestimate the contamination levels at the end of the shelf life."*

This demonstrates psychrotrophs are the cause of low temperature food spoilage. However, if a customer requires product to be incubated at 35°C/2 days for shelf life testing you will need to accede to their requirement.

Appendix 4: Target bacteria and how to control them

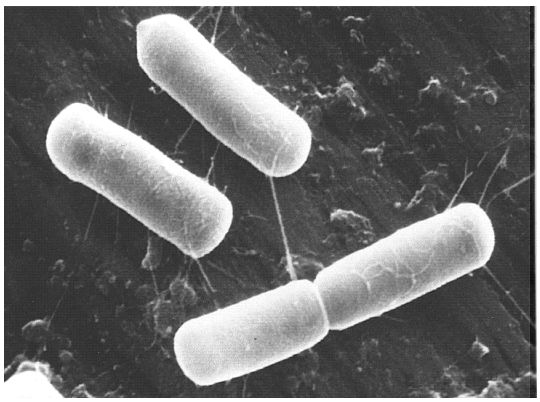
Bacteria are important in manufacture of RTE seafood for both the right and the wrong reasons – there are "good" and "bad" bacteria and they both play important roles in product safety.

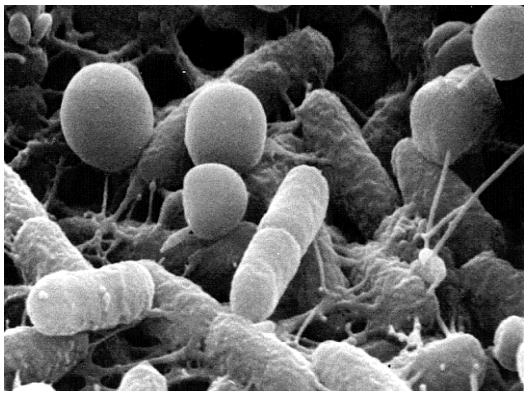
"Good" bacteria are starter cultures used in fermented smallgoods and cheese manufacture. The same bacteria become important in vacuum-packed CSS because they outgrow *Lm*, take away essential nutrients and release chemicals called bacteriocins, which limit the growth of *Lm*.

"Bad" bacteria are those that cause illness among consumers of RTE foods. They do this in two ways. Some bacteria produce chemicals called toxins that are poisonous to humans, and which they release into the food as they grow. The toxin causes food poisoning, particularly vomiting, usually between 2-6 hours after eating the product. For example, the *S. aureus* toxin is heat resistant and cannot be destroyed by cooking and can induce vomiting within 3 to 6 hours of eating a contaminated food.
Other bacteria cause illness by growing within our gut. It usually takes between 24 and 48 hours for these bacteria to grow to numbers high enough to cause illness.
Disease-causing bacteria are called pathogenic bacteria or pathogens. The pathogen of most importance in smoked fish is *Listeria monocytogenes* – it grows steadily in vacuum-packed, chilled conditions and it's salt tolerant.

The problem

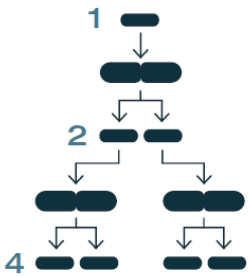
Bacteria are microscopic – if you lined them up in a queue you'd get about 1 million of them in a metre. This allows them to get into small crevices on working surfaces (left) and on food (right).

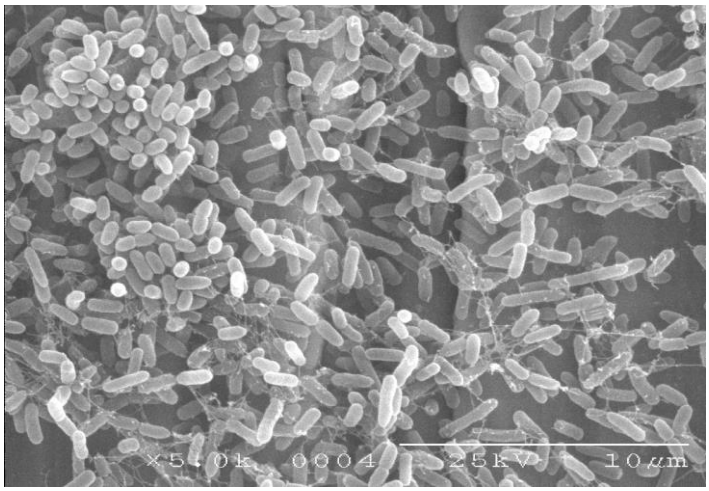




They may be small but they punch well above their weight because of the way they grow. They simply divide into two, and you can see some of them doing this in the images, above.

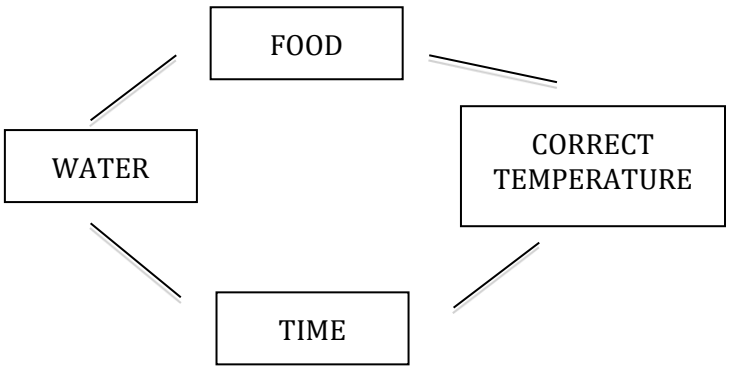
As you can see, this doubles the population and, given good conditions, one bacterium will multiply to 1,000,000 in less than 7 hours. They completely overgrow the surface of the food and generate off odours and make it slimy.





The result of bacterial growth – the food surface is overgrown and the population will be more than 10 million/cm² on the surface

What do bacteria need?



If bacteria have four factors (food, water, correct temperature and time) they'll grow quickly to big populations. Seafood contains protein, vitamins and other growth factors – and it's more than 70% moisture, so it's an excellent food for bacteria.
Temperature is a major control for bacteria and, in the seafood industry two groups are important depending on where they can grow.

	Growth range	Fastest growth	Target bacteria
Psychrotrophs	-5 to 30°C	15-25°C	*Listeria*
Mesophiles	7 to 45°C	25 to 37°C	*Salmonella, E. coli, Staphylococcus, Clostridium*

Controlling bacteria in your operation

The basis of most controls is simple - just take away whatever bacteria need in order to grow.
You're in the seafood business, so you can't deny them their food.
The other three factors are within your control and you use them widely in your everyday operations.

Temperature

In your business you probably use four temperature zones to control target bacteria:

- Freezing stops all growth because most of the water is removed as ice crystals.
- Chilling cooler than 7°C stops target bacteria that are mesophiles, but doesn't stop *Listeria,* which can grow steadily at chill temperatures.
- Cooking/smoking at 65°C for 10 minutes kills many pathogens, including *Lm*.
- Cooling smoked seafood rapidly through the Danger Zone (35-45°C) prevents any survivors growing to large numbers.

Water

Curing has been used for thousands of years and involves adding salt or sugar to make a brine. In brine, the water is "tied-up" by the salt and is not available to the bacteria.
There is a technical term called "water activity (a_w)" which describes how much water is tied up. Pure water has an $a_w = 1.0$ while cured fish has an $a_w < 0.95$.

Ingredients

Other controls which you use involve addition of chemicals which prevent bacterial growth. If you use liquid smoke or smoke generated from wood you're using chemicals which help to stop target bacteria from growing.

Vacuum packing

When you remove air from a pack by drawing a vacuum you're taking away the oxygen that spoilage bacteria need in order to grow. So vacuum packing improves shelf-life, but it has no effect on *L. monocytogenes*, which can grow in vacuum packs at chill temperatures.

Time

CSS and HSS have long shelf lives and *L. monocytogenes* grows steadily in the chiller. In fact, your chiller may be a permanent source of *Listeria* – growing in the door handles, door seals and refrigeration drip tray.

Appendix 5: Target hazard *L. monocytogenes*

L. monocytogenes has been known for over 70 years as a pathogen of small animals. It was named in recognition of Lord Lister who pioneered antiseptic surgery – methods for preventing wounds becoming infected during surgical operations. During the 1980s *Lm* became known as a food-borne pathogen as a result of several very large outbreaks, some involving scores of deaths. Among the species of *Listeria* the only pathogenic species is *Lm*.

Hazard severity

The International Commission on Microbiological Specifications for Foods (ICMSF), a gathering of the world's best microbiologists, describes the severity of illness according to three categories: Moderate, Serious and Severe; *Lm* is in the Severe category.

Severity	Description
Moderate	Not usually life threatening; no complications; normally short duration; symptoms are self-limiting; can have severe discomfort
Serious	Incapacitating but not life threatening; complications infrequent; moderate duration
Severe	Life threatening, substantial complications, or long duration

Impact and epidemiology

Lm has caused a number of serious outbreaks of food poisoning from smoked seafoods in several countries around the world (see Table 2 earlier in this document).

Infectious dose

For most people more than a million *Lm* must be swallowed before they become ill, with usually a 2-4 day bout of gastroenteritis. For vulnerable consumers, the infectious dose may be less than 5,000 organisms – you can get that from eating 50g of a product containing 100 *Lm* cells/g. In these consumers the illness starts with flu-like symptoms but can progress to meningitis (infection in the brain) or septicaemia (blood poisoning) and the illness may take several weeks to develop.

As shown in Table 2 a proportion (20-30%) of infected people die. These consumers are elderly people, pregnant women and their foetus or new-born baby, and people whose immune system is low or compromised e.g. because they had antibiotics or cancer treatment, post-transplant drug therapy, or because their liver is damaged.

Controlling authorities and food standards organisations have set a "zero-tolerance" for ready-to-eat foods because of the potential for it to grow in some foods to levels high enough to cause infection in susceptible consumers. Unfortunately zero tolerance is very difficult to achieve in practice because *Lm* is a robust organism that lives in food factories and also grows steadily in refrigerated foods with long shelf lives.

Growth of *L. monocytogenes*

Lm grows over a wide range of environmental conditions commonly found in seafood operations.

Conditions	Minimum	Optimum	Maximum
Temperature (°C)	-0.4	37	45
pH	4.4	7	9.4
a_w	0.92	0.99	>0.99

There are three reasons why *Lm* is a very robust organism in the seafood business:

- It can grow in refrigeration storage
- It is salt-tolerant
- It grows well in vacuum packs

It becomes a problem in products with the long shelf life (4-5 weeks) associated with smoked fish.

Killing *L. monocytogenes*

Lm is not very heat resistant and hot smoking will reduce high populations in product (> 1 million/g) to less than one/g.

Control in processing of smoked seafood

Lm is common in the environment and can enter food premises in many ways, including on the hands, clothing and boots of workers, for which reason factories making smoked seafoods have strict entry requirements.
The most critical situation for a food plant however, is if *Lm* sets up permanent residence in equipment which contacts product during slicing and packing. And if the organism sets up permanent residence in some hard-to-clean area, it may be impossible to prevent recontamination of product without radical action to break the *Listeria* cycle.

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