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How to meet an FSO – Control of *Listeria monocytogenes* in the smoked fish industry*

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Introduction

Listeriosis is a food-borne disease which primarily affects people in particular risk-groups (immuno-suppressed, elderly, neonates) in which lethality is as high as 25 %. The disease does occur in out-breaks but most cases appear as sporadic cases. The disease is primarily associated with ready-to-eat (RTE) food products with extended (refrigerated) shelf life. Outbreaks have mostly been caused by dairy and meat products, however, the disease has also been traced to lightly-preserved fish products such as smoked mussels and cold-smoked fish (trout).

Determining infectious dose is difficult due to lack of good animal infection models and the risk associated with different doses of the organism has therefore been evaluated by comparing disease rates with prevalence with models of growth of the organism (1). The quantitative risk assessments conducted by FDA/FSIS (2) and WHO/FAO (3) have used a similar approach and have also compared a range of dose response curves from different studies. Using such approaches, all studies conclude that the risk of listeriosis is higher when consuming food products with high numbers of the organism than when consuming food products with low levels (figure 1 (3)). However, a minimum infectious dose cannot be determined. The WHO/FAO team concluded as part of an expert consultation that 99 % of all listeriosis cases would be eliminated if levels of *L. monocytogenes* (*L.m.*) were kept below 1000 cfu *L.m.*/g at point of consumption assuming a consumption pattern equivalent to that of the USA.

Listeriosis from lightly preserved fish products?

The risk assessments by WHO/FAO and FDA/FSIS cover listeriosis from read-to-eat products, in general, and does not cover all the individual products. However, an attempt is made to rank different food commodities in terms of relative risk

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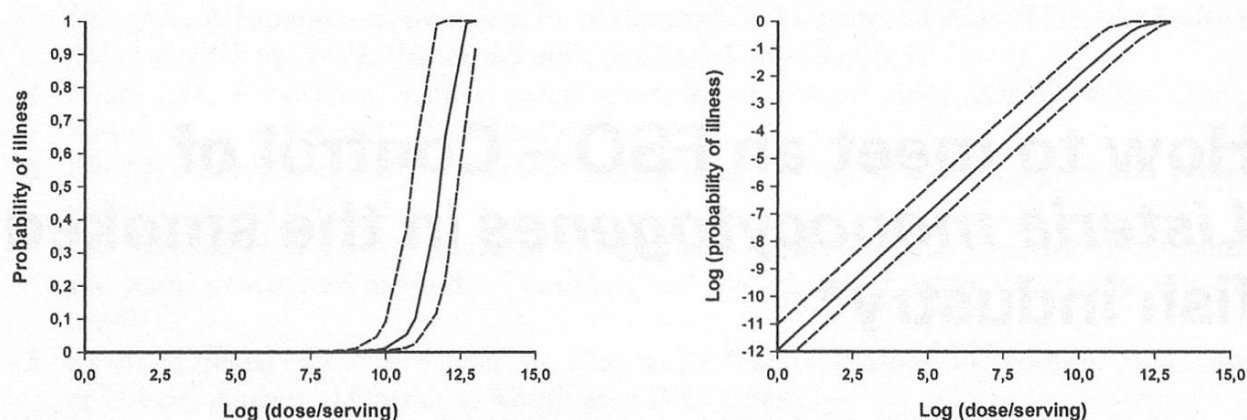


Figure 1 Dose response curve (left: log-linear and right: log-log) for listeriosis in susceptible individuals consuming ready-to-eat food products

(table 1 (4)). These estimates are based on a number of assumptions (serving size (60 g), number of servings per year (1–18), levels of the organism, risk per serving 2.1×10^{-8} etc.) and should only be taken as rough estimates. The FDA/FSIS risk assessment should be consulted for these assumptions. Using the estimates of this assessment, roughly 1% of listeriosis cases is believed to be caused by cold-smoked fish.

Table 1

Estimates of relative risk of listeriosis from ready-to-eat food products

Food	Cases of listeriosis per 100 000 people per year	Cases of listeriosis per 10^6 serving per year
Milk	0.091	0.005
Ice cream	0.00012	0.000014
Fermented meats	0.0000066	0.0000025
Smoked fish	0.0046	0.021
"all" RTE foods	0.5	

Recent US data have shown that 4–5 % of smoked fish samples were positive for *L. monocytogenes* (5). A Danish study (6) indicated great plant-to-plant variation in contamination rate, thus from some plants all product samples were positive whereas other plants produced products where *L. monocytogenes* was not detected (6). *L. monocytogenes* typically occurs at levels of < 10 cfu *L.m.*/gram, but is sporadically isolated at higher levels (5, 6). In the recent study of Gombas *et al.* (5), smoked fish was the only product with contained samples with *L. monocytogenes* levels between 10^4 and 10^6 cfu/g.

L. monocytogenes may grow in vacuum-packed, cold-smoked salmon stored at chill temperatures (4–5 °C). Some trials (typically inoculated packs) report several log increases in 3–4 weeks whereas other studies report slower or no growth (7).

Setting a food safety objective for *L. monocytogenes* in ready-to-eat products is a societal decision and to date no government has officially announced a food safety objective for the organism. Considering the common occurrence and consumption of low levels, ICMSF has used 100 cfu/g as an example of an FSO (8).

There are no documented (published) cases of listeriosis from cold-smoked salmon; however, cold-smoked trout and other lightly preserved fish products have caused listeriosis (9, 10, 11, 12).

Risk Management Options

The sections below build on earlier lectures at this SGLH-symposium and readers can consult these texts for definitions of terms and explanations.

Listeria monocytogenes can be controlled, but probably not eliminated from the cold-smoked salmon production. Low levels of *L. monocytogenes* are consumed daily in a variety of ready-to-eat cold-smoked fish, including cold-smoked salmon without major adverse effects, and there are few documented incidents of listeriosis linked to these products. Hence elimination is not required to meet public health goals, but stringent control of increase to hazardous levels is.

Following the concepts introduced by ICMSF (13), one can summarize microbiological control measures as follows:

- Control initial level (H_0)
- Reduce levels during production (R)
- Prevent re-contamination (I)
- Prevent growth (I)
- Reduce levels just before consumption (R)

The Food Safety Objective is expressed as: $H_0 - \Sigma R + \Sigma I \leq \text{FSO}$
(See text of Martin Cole in this journal.)

Initial levels (H_0) are typically low. Contamination rates of raw fish vary depending on geographical region. As part of this example, an initial level of 1 cfu *L.m./g* is assumed as H_0 .

Cold smoked fish is salted to 3–6 % NaCl in the water phase and subsequently cold-smoked which takes place at 22–28 °C. These procedures will not *per se* reduce levels of *L. monocytogenes* therefore R is not present in the equation. It should be noted that the number of positive samples often is significantly lower immediately following cold-smoking (14) than when sampled just before smoking indicating that in practice some reduction takes place.

During processing, contamination rates increase, and an I resulting in 1 cfu *L.m./g* is estimated. Typically the I denoting (re)contamination is an absolute figure such as 1 or 10 cfu *L.m./g*, whereas the I denoting growth describes an increase. One must carefully evaluate where in the process a potential re-contamination occurs when inserting numbers to the equation. Growth during storage may vary. Some investigations point to only marginal growth (e.g. 1 log unit) during storage (6) whereas other studies see sporadic high levels (5). Therefore, for some products a

value for I of 1–2 log units increase may be valid whereas a value of 5–6 log units increase may apply to other products.

Assuming that the fish is consumed “raw” by the consumer (i.e. without cooking), there will be no reduction.

Clearly, in the cases where significant growth occurs, control measures need to be implemented. Reducing H_0 will not ensure that the FSO is met, as long as the recontamination remains at the 1 cfu *L.m./g* level. Therefore GHP and HACCP have to reduce I; i.e. prevent/limit (re)contamination and/or prevent/limit growth.

Good Hygiene Practice

Listeria monocytogenes is capable of colonising food processing environments, and product contamination is often contamination during processing rather than by survivors from the raw material. Hence each smoke-house may harbour its own unique type of *L. monocytogenes* (15, 16). *L. monocytogenes* may hide in brines, colonise slicers and have its harbouring niches in drains and on floors (14, 17, 18, 19). Therefore, the GMP/GHP program of a food processing plant with *L. monocytogenes* as an identified hazard, must focus specific actions on doing environmental sampling for the organism with the ultimate aim being its elimination from the food processing environment, more specifically, food contact surfaces. It should be noted that the contamination rate of the raw fish may vary between different slaughterhouses and the raw fish may be the original source of *L. monocytogenes* introducing it into the factory environment.

HACCP

A hazard analysis of *L. monocytogenes* in cold-smoked salmon production, distribution and use reveals that with current processing and storage practices, no listericidal step (CCP₁) exists. Thus the organism survives processing, recontamination will occur and the typical storage conditions [vacuum-packed, chill-stored (5 °C), NaCl at 3–6 % (water phase salt) and pH of approx. 6.2] may not prevent growth to hazardous levels. Sporadically, high levels of the organism are detected. However, it must be emphasised that control measures that prevent or limit growth after production can be introduced, e.g. by frozen storage or by limiting shelf-life.

Several studies have documented that *L. monocytogenes* grows well in cold-smoked salmon. Such studies are typically using inoculated packs to evaluate growth potential. Growth in naturally contaminated products is mostly not nearly as pronounced (6). This may be explained by the inhibition of growth by naturally occurring lactic acid bacteria (17). Although cold-smoked salmon is regarded as “one” product, it actually covers a diverse range of products where NaCl levels range from as low as 1.5 % NaCl to almost 7 %. Also, smoking procedures vary and some types of smoking may be inhibitory to growth of the organism. Further studies must evaluate the inhibitory properties of NaCl, smoke and lactic acid bacteria to determine if a CCP actually may be designed.

Several measures that can prevent growth (hence constituting a CCP) could be implemented:

- The use of freezing for storage/distribution;
- >10 % NaCl (WPS) or the addition of other inhibitory factors;
- Lactates with or without a CO₂-atmosphere;
- Lactic acid bacteria and/or their bacteriocins;
- Selected salt-smoke combinations, paying special attention to the phenol content of the smoke;
- Eliminating *L.m.* after packaging using processes such as ultra-high pressure, irradiation, etc.

Performance Objectives and Criteria

Following the ICMSF (13), a performance criterion is the required outcome of a step, or a combination of steps that contribute to assuring a food safety objective is met. This may be expressed for instance as a kill step, e.g. a 12D reduction of proteolytic *Cl. botulinum* or as a particular frequency of contamination in raw material. More recently, the term "performance objective" has been introduced to describe an equivalent of the FSO further back the food processing chain. Hence, the term performance criteria (PC) is then describing the outcome (e.g. 12D reduction). As stated previously, if growth of *Listeria monocytogenes* is possible/likely during storage and distribution, the FSO must be translated to a performance objective (PO) to compensate for the amount of growth expected between sampling and consumption. Thus, it has been demonstrated that in naturally-contaminated cold-smoked salmon stored at 5 °C, an ca. 1 log increase occurs during a 3-week storage period occurs (6). Therefore, if a shelf life limit of less than three weeks (at 5 °C) is used, the PC of 10 cfu *L.m.*/g at the end of the processing line will allow the FSO to be met. Most processors will set a PC of <10 cfu *L.m.*/g to build in safety margins. However, at present, there is no consensus on what this safety margin should be.

Product and process criteria

The preservation and safety of cold-smoked salmon depends on the use of appropriate raw materials, limitation of recontamination and combinations of salt and low temperature after processing to limit growth. Since no listericidal step is included in the processing no process criteria can be set. Moreover, intrinsic growth control measures specific for *L. monocytogenes* are normally not applied, thus product criteria cannot be set. It should be noted, that on-going work on control measures such as lactate-diacetate, lactic acid bacteria or specific smoke/NaCl combinations may result in development of product criteria that may control the growth of the organism.

Microbiological Criteria

Sampling and testing and using microbiological criteria may in some cases be used as control measure. When the establishment of microbiological criteria is

chosen as a risk management option, such criteria should be based on the FSO of <100 cfu *L.m./g* or a PC derived from this level. They may be used as acceptance criteria in situations where the history of the product is not known, at points such as at port-of-entry or at certain retail outlets. It should be considered in each product/hazard combination if other acceptance criteria will provide a larger degree of safety assurance. It is fairly evident then that, in practice, for cold-smoked salmon, the only way one can achieve an FSO of 100 cfu *L.m./g* for a product with a 3 week shelf-life, is to prevent or limit growth of the organism to 1 or 2 logs.

Evaluation of sampling plans

Using the cases introduced by ICMSF (13) to determine sampling plans, case 10 applies when the number is reduced before consumption, case 11 applies to products where growth will not occur and case 12 applies to products where growth may occur. In both situations ICMSF has set the microbiological limit at <100 cfu *L.m./g* (21). An important part of evaluating if sampling/testing is a valid control measure is to determine the performance of the sampling plan. Guidance for this has been provided in a spreadsheet available from <http://www.dfst.csiro.au/icmsf/samplingplans.htm>. Using this spreadsheet and assuming a log-normal distribution with a standard deviation of 0.8 logs, the following conclusions can be drawn:

If growth does not occur, case 11 ($n=10$, $m=100$ cfu *L.m./g* (equivalent to FSO) and $c=0$) is used. This sampling plan will give a $\geq 95\%$ probability of rejecting lots if their mean *L.m.* count is ≥ 30 cfu *L.m./g*.

If limited growth occurs, case 12 ($n=10$, $m=1$ cfu *L.m./g* and $c=0$) is used. This sampling plan will give a $\geq 95\%$ probability of rejecting lots if their mean *L.m.* count is ≥ 0.135 cfu *L.m./g*.

If extensive growth does occur, case 12 ($n=20$, $m=\text{"absence in 25 g"}$ and $c=0$) is used. This sampling plan will give a $\geq 95\%$ probability of rejecting lots if their mean *L.m.* count is ≥ 0.00537 cfu *L.m./g*.

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Summary

This text discusses options for controlling *Listeria monocytogenes* contamination and growth in cold-smoked fish processing. It arrives at a suggested value for a so-called food safety objective (FSO) by referring to national and international quantitative risk assessments as well as earlier studies of prevalence and growth of the bacteria. Especially process-contamination and growth during storage are issues that must be dealt with to control the organism, as current day processing does not

include a critical control point (CCP). Finally, possible product criteria (chemical or microbiological criteria) are outlined.

Zusammenfassung

Dieser Beitrag diskutiert die Optionen bei der Kontrolle von Kontaminationen durch *Listeria monocytogenes* und deren Wachstum während der Kalträucherung von Fisch. Die Diskussion mündet in einem suggerierten Wert, dem so genannten FSO (Food Safety Objective), in Anlehnung an die nationalen und internationalen Risikobewertungen und an frühere Studien über Prävalenz und Wachstum dieses Bakteriums. Um diesen Organismus unter Kontrolle halten zu können, muss insbesondere auf die Kontamination während des Prozesses und auf ein Wachstum während des Vertriebs geachtet werden. Dies ist besonders wichtig, da heute dieser Prozess keinen kritischen Lenkungspunkt (CCP) beinhaltet. Zum Schluss werden mögliche Kriterien (chemische oder mikrobiologische) diskutiert.

Résumé

Cette contribution discute les options disponibles pour maîtriser les contaminations par *Listeria monocytogenes* et leur croissance durant le fumage à froid de poisson. Les discussions débouchent sur une valeur suggérée, le FSO (Food Safety Objective), ceci sur la base d'évaluations nationales et internationales des risques ainsi que sur des données d'études existantes sur la prévalence et la croissance de cet organisme. Il faut être en particulier attentif à la recontamination durant le processus de fabrication et à la croissance durant la distribution pour pouvoir maîtriser ce germe. Ceci est particulièrement important car il n'existe pas, à l'heure actuelle, de point critique de maîtrise (CCP). Pour terminer, des critères possibles chimiques ou microbiologiques, sont discutés.

Key words

Listeria monocytogenes, Food Safety Objective, HACCP, microbiological criteria, process criteria

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