Joint FAO/WHO Expert meeting on microbiological risk assessment of *Listeria monocytogenes* in foods

FAO HQ, Rome, Italy: 24 - 28 October 2022

SUMMARY AND CONCLUSIONS

Issued in November 2022

The Joint FAO/WHO Expert meeting on microbiological risk assessment of *Listeria monocytogenes* in foods was convened in response to a request by the Codex Committee on Food Hygiene (CCFH) at its fifty-second session¹ to undertake a full farm to table risk assessment on *Listeria monocytogenes* in food in order to inform a possible future revision of the Guidelines on the Application of General Principles of Food Hygiene to the Control of *Listeria monocytogenes* in Foods (CXG 61- 2007)².

This document summarizes the conclusions and recommendations for the development of microbiological risk assessment of *L. monocytogenes* in specific foods; namely, leafy greens, frozen vegetables, cantaloupe melon and ready-to-eat (RTE) seafood, in the light of new data and approaches. This document has been prepared to facilitate the deliberations of the upcoming CCFH. The full report will be published as part of the Food and Agriculture Organization (FAO) and World Health Organization (WHO) Microbiological Risk Assessment (MRA) Series.

The meeting participants are listed in Annex 1 of this summary report. Dr Laurent Guillier served as Chairperson and Dr Jovana Kovacevic as Rapporteur.

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More information on this work is available at:


and

https://www.who.int/foodsafety/en/

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Scope and objectives

In response to a request from the 52nd Session of the Codex Committee on Food Hygiene (CCFH), the FAO/WHO Joint Expert Meeting on Microbiological Risk Assessment (JEMRA) convened a meeting in Rome, Italy, from 24 to 28 October 2022, with the objective of developing formal full risk assessment models for *Listeria monocytogenes* in selected foods (FAO and WHO, 2022). In the light of the available data and current risk assessment approaches, the expert group aimed to collectively ascertain the stages from primary production to consumption to be represented in the model, including approaches that accommodate the testing of scenarios, interventions and sampling schemes that could reduce the risk of listeriosis. The expert group also suggested possible future revisions of the three annexes of Codex Guidelines on the application of general principles of food hygiene to the control of *Listeria monocytogenes in foods* (CAC/GL 61 - 2007) (FAO and WHO, 2007) based on available evidence (Annex 2).

The expert group agreed that:

- Within each food group, the following food commodities were considered in the models: cut and packaged leafy greens (RTE lettuce), whole leafy greens (whole lettuce), RTE diced cantaloupe, whole cantaloupe, smoked fish and gravad fish (RTE seafood) and non-RTE blanched frozen vegetables (frozen vegetables).
- A full primary production to consumption risk assessment model should be applied to the food commodities utilizing a modular approach, ideally flexible to be reused between them and for other similar food commodities (e.g. frozen beans, fresh herbs). The different modules included in the risk assessment models for the four food groups proposed by the expert group will be implemented with open-source software.
- The primary production (pre-harvest) module should enable the assessment of the introduction of the pathogen in the raw materials, taking into account, if possible, the effects of season, agrifood practices, and climate change.
- The latent possibility of cross-contamination from the primary production to consumption should be considered.
- The dose-response model will be adapted from existing models that consider variability in pathogen virulence and consumer susceptibility and be common to all food commodities.
- Inclusion of whole genome sequencing (WGS) and other -omics data on *L. monocytogenes* may inform the risk assessment.
- The different (sub)modules of the models that describe the relevant stages of a food chain can be used to evaluate the impact of the stages on the risk in “what-if” scenarios.
- An uncertainty and sensitivity analysis should be performed to identify which model inputs and assumptions have the greatest impact on the model outputs (such as dose and risk for the consumer), which will help identify relevant “what-if” scenarios and data needs.
- There is value in gathering more data on *L. monocytogenes* in the food chain from different sampling and testing schemes to inform risk assessment.
Conclusions
The expert consultation concluded the following:

Leafy greens

- The assessment of the risk of listeriosis due to the consumption of leafy greens should be undertaken through a full primary production to consumption approach. Existing data can support a modular approach.
- The production activities including irrigation, fertilization and other on-farm management practices have an impact on the occurrence of L. monocytogenes on farm and could be modelled in a primary production (pre-harvest) module.
- Season has been recognized as an important factor driving the microbial kinetics in soil and on the leaves, and as such, it should be introduced in the model. Seasonal environmental conditions as an input may be altered in “what-if” scenarios created to assess the potential impacts of climate change.
- RTE and whole leafy greens (non-RTE) should be addressed in the model and should share a common primary production module. Lettuce was proposed as a representative commodity for the leafy greens model.
- Relevant stages to be represented in the risk assessment were identified, including: growth of leafy greens (field, controlled environment, hydroponics), harvesting, cooling, washing, sanitizing, cutting, packaging, multiple transportation steps, display at retail, and consumer practices.
- The model should be structured to measure the effectiveness of the prevention of contamination by soil/irrigation water, the efficacy of washing with or without sanitizers, the prevention of cross-contamination events along the production chain, the application of good hygiene practices during processing, the efficacy of sampling schemes, and the impact of different consumers’ practices related to handling and storage.
- The risk assessment should consider the possibility of cross-contamination and/or recontamination; e.g. cross-contamination at processing facilities and/or at home.
- The model should have the flexibility to assess climate change impact; e.g. scenarios that include extreme weather events and seasonal effects on the occurrent of L. monocytogenes in the environment.

Frozen vegetables

- The model should pertain to the risk of L. monocytogenes in non-RTE frozen vegetables.
- The assessment of the risk of listeriosis should span processing to consumption, as the main factors driving the risk have been identified in the stages of processing and consumer handling. L. monocytogenes can contaminate vegetables post-blanching; the model should consider the possibility of contamination events post-blanching.
- Relevant stages to be represented in the risk assessment were identified, including: cleaning, washing, blanching, freezing, packaging, display at retail, consumer practices such as defrosting, cooking, as well as common practices relating to non-intended use.
- The risk assessment should be structured to measure the effectiveness of blanching or other inactivation steps pre- or post-packaging, the prevention of contamination post-blanching, and the efficacy of sampling schemes at the end of processing.
- The risk assessment should evaluate the impact of different consumer practices related to storage and cooking, and the impact of shifting consumer practices to increase compliance with cooking of non-RTE frozen foods (i.e. through better labelling and targeted messaging for more susceptible groups to cook frozen vegetables).

### Cantaloupe
- The model should pertain to the risk of *L. monocytogenes* in whole and RTE diced cantaloupe.
- The assessment of the risk of listeriosis due to the consumption of cantaloupe should be undertaken through a full primary production to consumption approach. Existing data can support a modular approach.
- Relevant stages to be represented in the risk assessment were identified, including: cantaloupe growing on field, harvesting, cooling, washing, sanitizing, dicing, display at retail, consumer practices and multiple transportation steps.
- Flexibility to account for the impact of farming practices, extreme weather events, climate change, as well as for diverse market practices should be included in the risk assessment.
- The risk assessment should consider cross-contamination events at pre-harvest (e.g. irrigation water, soil, fertilizer), at processing (e.g. pooling of fruits from other producers, food contact surfaces/equipment and/or dicing), retail (e.g. market practices), and at consumer level (e.g. contamination from rind to flesh during slicing/dicing).
- The risk assessment should be structured to measure the effectiveness of the prevention of contamination during pre-harvest (e.g. soil, irrigation), the application of on-farm preventive measures (e.g. cantaloupes grown on soil barriers), the effect of processing stages, such as cleaning/washing, sanitization and removal of bruised parts, and the impact of consumers practices.
- The effect of the time/temperature profiles throughout the food supply chain (e.g. processing, transport, retail and consumer homes) is important to include in the risk assessment.

### RTE seafood
- A full primary production (harvest and farming) to consumption risk assessment for the risk of *L. monocytogenes* should be developed for RTE seafood. Hot- and cold-smoked fish and gravad fish were proposed as a representative food category for the RTE seafood model.
- The risk assessment should be flexible to accommodate other RTE fish products in the future (e.g. sashimi, ceviche).
- Relevant stages to be represented in the risk assessment were identified, including: growth of fish (open sea, aquaculture), harvesting, evisceration and head cutting, filleting, different smoking steps, gravad fish steps, freezing, slicing, packaging, multiple transportation steps, retail and consumer handling.
- Cross-contamination should be considered in the model because RTE fish products are produced in several steps, sometimes within one facility, in other cases in different facilities in different countries.
- The effect of added lactic acid bacteria cultures for biocontrol of *L. monocytogenes* should be evaluated in the risk assessment.
- Whole genome sequencing/strain typing data, when available, could be used to assess dominant strains at different production stages to inform cross-contamination module.
The risk assessment should consider raw materials coming in with different levels of contamination. The effect of the time/temperature profiles throughout the food supply chain (e.g. processing, transport, retail and consumer homes) is important to include in the risk assessment.

Dose-response

Based on a review of the published dose-response models for *L. monocytogenes*, the expert group proposed the use of existing models based on susceptible populations with underlying conditions that increase risk of listeriosis (or different risk of illness in different age-gender groups). The proposed dose-response model considers consumer susceptibility and virulence characterization based on the genomic data that are currently available.

- An updated set of parameters of this model that account for three classes of virulence of *L. monocytogenes* should be put forward, incorporating current data on specific virulence profiles associated with sequence types (ST) and/or clonal complexes (CC).
- There is a need for additional data on *L. monocytogenes* in the food chain to better inform *L. monocytogenes* occurrence, virulence and dose response, so that a risk assessment for different ST/CCs of *L. monocytogenes* can be performed.

WGS

- The global prevalence of *L. monocytogenes* in the exposure assessment part can be replaced by the specific prevalence for groups of ST/CCs when WGS data have demonstrated that these groups of ST/CC are overrepresented in a specific commodity.
- The three classes of virulence proposed in MRA 38 should be used in the model. Scientific literature should be used to compile a list of ST/CCs belonging to different virulence groups.
- At this time, genetic biomarkers for enhanced robustness or increased fitness are not sufficiently conclusive for subgrouping *L. monocytogenes* with respect to difference in behaviour to be incorporated in the exposure assessment part.
- The uncertainty between phenotypic and genotypic profiles of *L. monocytogenes* should be considered and carefully examined before being used in risk assessment.

The expert group highlighted several paragraphs in the three annexes in the *Guidelines on the application of general principles of food hygiene to the control of Listeria monocytogenes in foods* (CAC/GL 61-2007) that could benefit from an update (Annex 2).

In conclusion, the expert group elaborated formal models for the risk assessment of *L. monocytogenes* for lettuce, cantaloupe, frozen vegetables and RTE fish. As a next step, these models should be programmed, tested and reviewed. The review should verify that the models are flexible to run different scenarios and incorporate new data to account for national, regional and international context. Following the review, the models should be made publicly available. These novel models will advance the state of knowledge on *L. monocytogenes* risks by incorporating advances in next generation technologies and by spanning primary production to consumption.
Annex 1: List of participants

EXPERTS

Hamzah Al-Qadiri, University of Jordan, Jordan

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RESOURCES PERSONS

Lingping Zhang, Joint FAO/WHO Food Standards Programme, Italy

SECRETARIAT

Ursula A. Gonzales-Barron, Instituto Politécnico de Bragança, Portugal

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Christine Kopko, FAO, Italy

Jeffrey LeJeune, FAO, Italy

Juliana de Oliveira Mota, WHO, Switzerland

Moez Sanaa, WHO, Switzerland

Kang Zhou, FAO, Italy
### Annex 2: Recommended revisions to the Annexes from the Guidelines on the application of general principles of food hygiene to the control of *Listeria monocytogenes* in foods (CAC/GL 61-2007)

<table>
<thead>
<tr>
<th>Para.</th>
<th>CAC/GL 61-2007</th>
<th>JEMRA recommendations</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Annex I: Recommendations for an environmental monitoring program for <em>Listeria monocytogenes</em> in processing areas</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Title</strong></td>
<td>Recommendations for an environmental monitoring program for <em>Listeria monocytogenes</em> in processing areas</td>
<td>• To consider including primary production (handling, packing, with or without water) to the title. This could be done in a separate Annex or by modifying existing Annex I to include primary production consideration.</td>
</tr>
<tr>
<td><strong>Introduction</strong></td>
<td>“…foods that support <em>L. monocytogenes</em> growth and that are not given…”</td>
<td>• To consider the inclusion of other foods that do not support <em>L. monocytogenes</em> growth (e.g. frozen foods – frozen fruits/vegetables, ice cream). • To consider including information on the purpose of the environmental monitoring program, as this will drive the sampling approach (i.e. verification of cleaning and sanitation, reaction to a positive sample, etc.).</td>
</tr>
<tr>
<td><strong>a)</strong> Type of product and process/operation</td>
<td></td>
<td>• To consider including history of <em>L. monocytogenes</em> in raw materials to this section and including primary production. • To consider products, equipment (e.g. forklifts) and employee flow within the processing environment.</td>
</tr>
<tr>
<td><strong>b)</strong> Type of samples</td>
<td></td>
<td>• To consider highlighting the importance of non-food contact surfaces (NFCS), and close to food contact surfaces (FCS), in specific operations (i.e. frozen foods, condensation). • To consider adding examples of other NFCS and FCS (workers, shoes, gloves and cleaning equipment).</td>
</tr>
<tr>
<td><strong>Time of sampling</strong></td>
<td></td>
<td>• To consider adding an additional bullet to capture the time of sampling during production or prior to the start of production (e.g. T = 0 and T = 3hr into production).</td>
</tr>
<tr>
<td><strong>g)</strong> Analytical methods</td>
<td></td>
<td>• To consider updating to include more relevant techniques (e.g. remove pulsed-field and ribotyping replace with WGS and MLST).</td>
</tr>
<tr>
<td><strong>i)</strong> Actions in case of positive results</td>
<td></td>
<td>• To consider including source tracing and isolate profiling to help understand transient or persistent nature of contamination.</td>
</tr>
</tbody>
</table>
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- To consider the addition of “Seek and destroy” approach.
- To consider micro-climate (e.g. air movement and condensation)
- To consider the addition of recommendation for root cause analysis.

### Annex II: Microbiological criteria for *Listeria monocytogenes* in ready-to-eat foods

<table>
<thead>
<tr>
<th>Section</th>
<th>Table</th>
<th>Note</th>
</tr>
</thead>
<tbody>
<tr>
<td>4.1 &amp; 4.2</td>
<td>Table 1 &amp; Table 2, footnote c</td>
<td>Delete the word “analytical standard deviation” and change to “standard deviation” and provide justification for the value used.</td>
</tr>
<tr>
<td>4.1 &amp; 4.2</td>
<td>Table 1 (heading)</td>
<td>To consider clarifying “m” by amending the text in the footnote to specify acceptable lots (≤m) from unacceptable lots (&gt;m).</td>
</tr>
</tbody>
</table>

### Annex III: Recommendations for the use of microbiological testing for environmental monitoring and process control verification by competent authorities as a means of verifying the effectiveness of HACCP and prerequisite programs for control of *Listeria monocytogenes* in ready-to-eat foods

#### Introduction

- To consider expanding the scope of the annex to include elaboration of the concepts from Section 3 of the main text of the Code, to capture primary production given that some foods (e.g. cantaloupe, leafy greens) can go directly from farm to sale and on-farm activities (e.g. harvest, cutting, washing, packing) can be a source of contamination with *L. monocytogenes*.

#### a) Environmental Monitoring

- To consider recommending storage of isolates for further characterization and sharing of data for risk assessment.

#### b) Process Control Verification

- To consider recommending corrective actions for FBOs for when microbiological criteria are exceeded or for when *Listeria* spp. is detected (NFCS and FCS). *Listeria* spp. or other indicators are some of the options to include in the criteria for corrective action.