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Validation of Ready-to-Eat Foods for Changing the Classification of a Category 1 into a Category 2A or 2B Food

in relation to Health Canada's *Policy on Listeria monocytogenes in Ready-to-Eat Foods (2011)*

Date of implementation: November 29, 2012

Bureau of Microbial Hazards
Food Directorate
Health Products and Food Branch

Canada

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Disclaimer: Certain companies may be producing ready-to-eat (RTE) foods under Hazard Analysis Critical Control Points (HACCP) plans. The management and maintenance of effective HACCP plans require validation and verification. The aim of this present document **is not** to replace validation provisions as set forth by HACCP plans. If the intent is to change the classification of a Category 1 into a Category 2A or 2B RTE food (in relation to Health Canada's *Policy on Listeria monocytogenes in Ready-to-Eat Foods (2011)*), product-specific validation must be performed by processors/importers, as described in this document, but this validation **is not** a condition of sale.

1. Objectives and context of this document

This document has been prepared to assist processors and importers of RTE foods with the validation of the classification of their RTE products into either Category 2A or Category 2B, as defined in Health Canada's *Policy on Listeria monocytogenes in Ready-to-Eat Foods* (Health Canada, 2011)[1, 2]:

- **Category 2A:** RTE food products in which limited growth of *Listeria monocytogenes* to levels not greater than 100 colony forming units (CFU)/gram (g) can occur throughout the stated shelf-life[3]. This category of RTE food includes:
    - RTE foods which are known to occasionally contain low levels of *L. monocytogenes* **and** do not have a kill step

    and/or
    - RTE refrigerated foods with a stated shelf-life of ≤ 5 days.

- **Category 2B:** RTE food products in which the growth of *L. monocytogenes* cannot occur throughout the stated shelf-life (in other words, a RTE food in which *L. monocytogenes* does not increase in numbers by 0.5 log CFU/g or greater throughout the stated shelf-life).

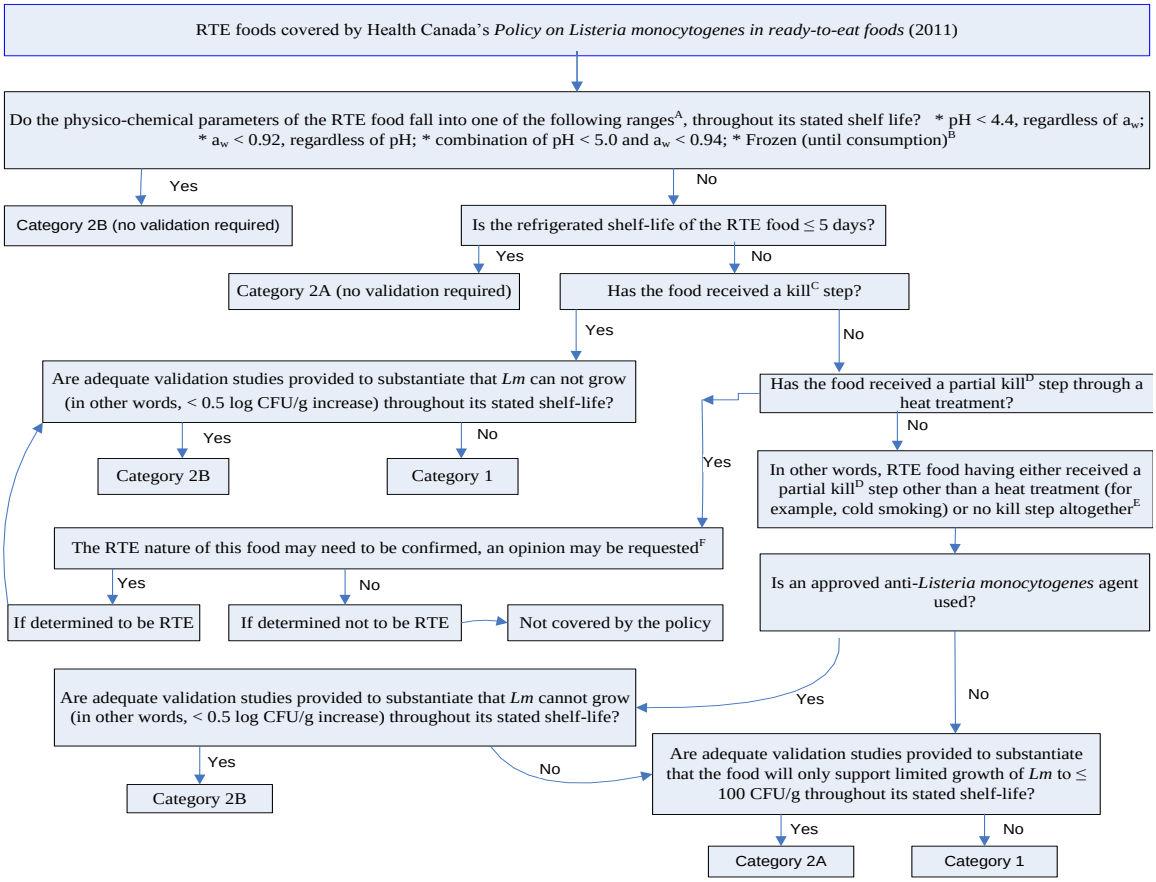
Processors and importers should follow the flowchart presented in Figure 1 – Classification of RTE foods – to determine if a RTE product requires validation to determine if it prevents or limits the growth of *L. monocytogenes*, based on its categorization.

[1] Please see the "ready-to-eat food" definition presented in Appendix A of Health Canada's *Policy on Listeria monocytogenes in Ready-to-Eat Foods* (Health Canada, 2011) to confirm if the product under consideration is covered by the above policy, and hence the potential applicability of this document.

[2] Please note that the compliance criteria for RTE foods falling under Category 2A and 2B is ≤ 100 CFU/g throughout the stated shelf-life, as presented in Table 1 of Health Canada's *Policy on Listeria monocytogenes in Ready-to-Eat Foods* (Health Canada, 2011).

[3] The term "stated shelf-life", used throughout this document, means the durable life date, for example the "best before" date, shown on the label of the package (Government of Canada, 2012).

Figure 1 – Classification of RTE foods



[A] Some Category 2 products may be intended for use in Category 1 products, or some RTE foods may be targeted to persons at high-risk. A finding of *L. monocytogenes* would lead to follow-up action(s) and hence, a health risk assessment (HRA) may be required on a case-by-case basis, to be conducted by the Bureau of Microbial Hazards (BMH), in order to determine the compliance action to be taken. These Category 2 foods may be assessed to represent a Health Risk 1 concern.

[B] Some frozen RTE foods, otherwise considered as Category 2B, may be temperature-abused, causing them to thaw and thereby could potentially permit the growth of *L. monocytogenes*. A finding of *L. monocytogenes* would lead to follow-up action(s) and hence, an HRA may be required on a case-by-case basis, to be conducted by the BMH, in order to determine the compliance action to be taken. These Category 2 foods may be assessed to represent a Health Risk 1 concern.

[C] For the purpose of this document, a kill step represents a validated treatment that achieves a minimum 5-log reduction in numbers of *L. monocytogenes.*

[D] For the purpose of this document, a partial kill step represents a treatment achieving a < 5-log reduction in numbers of *L. monocytogenes.*

[E] In general, these RTE foods are known to occasionally contain low levels of *L. monocytogenes.*

[F] For example, this could be processed products which have a cooked appearance (but are not fully cooked). These may be considered RTE and thus may be subjected to the provisions of Health Canada's *Policy on Listeria monocytogenes in ready-to-eat foods* (Health Canada, 2011) if they only have microwave cooking instructions, or if the instructions are only to warm and serve.

RTE products can fall into one of the scenarios described below:

- Category 2B if its pH/water activity (a_w) values are such that they do not support the growth of *L. monocytogenes* (Codex Alimentarius Commission, 2009), in other words, pH < 4.4, regardless of a_w; a_w < 0.92, regardless of pH; combination of pH < 5.0 and a_w < 0.94. **Validation NOT required.**

- Category 2B when the RTE product is kept frozen (until consumption) (Codex Alimentarius Commission, 2009). **Validation NOT required.**

- Category 2B providing that supporting validation information is presented and confirmed to justify its placement in this Category (in other words, numbers of *L. monocytogenes* increase by less than 0.5 log CFU/g throughout the stated shelf-life of the RTE product). **Validation required.**

- Category 2A when the RTE product has a stated refrigerated shelf-life that is ≤ 5 days. **Validation NOT required.**

- Category 2A providing that the following conditions are met:
    - It is known to occasionally contain low levels of *L. monocytogenes* **and** it does not have a kill step for *L. monocytogenes* that achieves a ≥ 5-log reduction in numbers of *L. monocytogenes* (in other words, achieves a < 5-log reduction in numbers of *L. monocytogenes* from non-thermal process(es) or no kill step altogether – see Figure 1)

  and
    - Supporting validation information is provided and confirmed to justify its classification as Category 2A. **Validation required.**

- Category 1 when the RTE product supports the growth of *L. monocytogenes.*

- Category 1 when the RTE product has a stated shelf-life longer than 5 days **and**:
    - insufficient, inadequate or no validation information is provided to consider it as either Category 2A or Category 2B RTE product

  or
    - its categorization, as either Category 2A or 2B, has not been confirmed by the relevant regulatory authority.

Note: Second generation RTE food products made from initially frozen RTE products (in other words, thawed for sale or used as an ingredient in other RTE products) also need to be categorized according to their final use. It is the responsibility of the secondary producer to know what type of RTE products will be made in order to comply to particular compliance criteria since these second generation of RTE products could fall into either of the categories described above.

If validation is required, processors and importers, as applicable, should submit their documentation to the relevant regulatory authority (for example, the Canadian Food Inspection

Agency, provincial or territorial government). The validation documentation will be evaluated based on the state of scientific evidence (Health Canada, 2012a; Health Canada, 2012b). In addition, the final classification of the RTE product, into either Category 2A or 2B, will be confirmed, if appropriate, by the relevant regulatory authority, in the context of Health Canada's *Policy on Listeria monocytogenes in Ready-to-Eat Foods* (Health Canada, 2011). If insufficient, inadequate or no information exists regarding the categorization of the RTE food product or if the categorization of the RTE food product has not been confirmed by the relevant regulatory authority, it will by default be considered as a RTE food in which growth of *L. monocytogenes* can occur (in other words, Category 1). If questions arise, it is the responsibility of the processor/importer to demonstrate to which category the RTE food belongs.

For additional information with regards to validation, please consult the Codex Alimentarius Commission document entitled *Guidelines for the Validation of Food Safety Control Measures* (Codex Alimentarius Commission, 2008).

2. Validation definition and procedure

The Codex Alimentarius Commission (2008) defines validation as follows:

> *Obtaining evidence that a control measure or combination of control measures, if properly implemented, is capable of controlling the hazard to a specified outcome.*

To that end, the following tasks must be conducted whenever validation is required, for changing the classification of a Category 1 RTE food into a Category 2A or 2B RTE foods:

- *Pre-validation tasks*:
  a) Hazard identification
  b) Identification of the food safety outcome
  c) Identification of the measure(s) that need to be validated

- *Validation*:
  The first task (a) is recommended, the second and third tasks (b, c) are essential, while the fourth task (d) is optional:

Recommended:	a) Literature review
Mandatory:	b) Challenge studies c) Identification of key process parameters, and provision of evidence of their control
Optional:	d) Modeling (useful when various RTE products are grouped for the purpose of challenge studies)

3. Validation procedure for Category 2A RTE foods

This category of RTE food includes:

- RTE foods which are known to occasionally contain low levels of *L. monocytogenes*[4] **and** do not have a kill step that achieves a ≥ 5-log reduction in numbers of *L. monocytogenes*, in other words, the processing of initial ingredients (in which *L. monocytogenes* may be present) for these specific RTE foods does not involve a heat treatment and achieves a < 5-log reduction in numbers of *L. monocytogenes* – see Figure 1 - ***Validation required*** to provide scientific evidence that limited growth of *L. monocytogenes* to levels not greater than 100 CFU/g can occur in these RTE foods throughout their stated shelf-life

and/or

- RTE refrigerated foods with a shelf-life of ≤ 5 days **- *Validation NOT required***.

Processors and importers, as applicable, who intend to submit validation documentation for a RTE product that they want to be classified as a Category 2A food must first provide the rationale used to determine that this product meets the criteria of a Category 2A product.

Non-thermal processing step(s) that achieve a < 5-log reduction in numbers of *L. monocytogenes*, can impact the RTE product in the following three ways:

1) The background microflora of the RTE product could compete with *L. monocytogenes*, should it be present.

2) It triggers the need to use an inoculum of 10-30 CFU/g, when conducting challenge studies with the goal of confirming that the levels of *L. monocytogenes* do not exceed 100 CFU/g throughout the stated shelf-life of the RTE product.

3) As such, testing of the RTE product at the beginning of its shelf-life becomes a key process parameter to confirm that the RTE product falls under the criteria that were used in the challenge study. This is to ensure that the concentration of *L. monocytogenes*, at the beginning of the shelf-life of the RTE product (time = 0), never exceeds the level that was used as an inoculum in the challenge study.

3.1. Pre-validation tasks for Category 2A RTE foods

a) Hazard identification:

The presence of unacceptable levels of *L. monocytogenes* in RTE foods as per the requirements of Health Canada's *Policy on Listeria monocytogenes in Ready-to-Eat Foods* (Health Canada,

[4] *L. monocytogenes* contamination of Category 2A RTE foods may be sporadic; initial contamination levels could be lower than the assay limit of detection, but over time may grow to detectable levels.

2011). In other words, the presence of more than 100 CFU/g of *L. monocytogenes* at any time throughout the stated shelf-life of the RTE product.

b) Identification of the food safety outcome:

The level of *L. monocytogenes*, when present, should be limited to a maximum of 100 CFU/g throughout the stated shelf-life of the RTE product.

c) Identification of the measure(s) that need to be validated:

Challenge studies need to demonstrate that when RTE foods are processed according to the parameters specified, growth of *L. monocytogenes* will be limited to a maximum of 100 CFU/g throughout the stated shelf-life of the RTE product. In order to demonstrate this:

i. The production process must be clearly defined, monitored and documented to ensure consistency. The RTE products produced using this process must consistently meet the criteria of Category 2A (in other words, RTE products known to occasionally contain low levels of *L. monocytogenes* **and** not having a kill step that achieves a ≥ 5-log reduction in numbers of *L. monocytogenes* – see Figure 1). For RTE products which have a shelf-life greater than 5 days, challenge studies must demonstrate that only limited growth of *L. monocytogenes* to levels not greater than 100 CFU/g will occur throughout the stated shelf-life.

ii. Processors must use an inoculum level of 10-30 CFU/g of *L. monocytogenes* in the challenge studies.

3.2. Validation process for Category 2A RTE foods

a) Literature review:

- It is recommended that a literature review be completed with regards to the possible level of *L. monocytogenes* contamination and growth profile of this microorganism in RTE product(s) under consideration, as well as similar RTE products.

- Pertinent publications should be included.

b) Challenge studies:

- Challenge studies should be conducted as per Health Canada's recommendations found in Health Canada's document *Listeria monocytogenes Challenge Testing of Refrigerated Ready-to-Eat Foods* which suggests experimental designs that consider the following parameters (Health Canada, 2012c):
    - *Listeria monocytogenes* strains

- Preparation and enumeration of cells (maintenance of cultures and inoculum preparation as well as inoculum level)
- Sampling design (note: a minimum of three lots of products must be tested for *L. monocytogenes* in triplicate at each sampling time (i.e., minimum of 5 time points throughout the stated shelf-life of the product, including time zero and at end of shelf-life))
- Preparation of food products
- Inoculation of food products
- Special product packaging conditions
- Incubation of inoculated food products (note: validation studies for refrigerated RTE foods must be performed at a temperature of 7^{o}C or above)
- Enumeration and enrichment methods
- Documentation of results

- The choices made when performing the challenge studies must be detailed and a rationale should be provided, for example, the selection of strains that will be used for inoculation, the number of tests performed, etc. (Health Canada, 2012b).

- The inoculation level used for the challenge study must be 10-30 CFU/g.

- The results of the challenge studies must indicate that the food safety outcome is met, in other words, that the concentration of *L. monocytogenes* will be ≤ 100 CFU/g throughout the stated shelf-life of the RTE product.

c) Identification of key process parameters, and provision of evidence of their control:

- Key process parameters need to be clearly identified (for example, the concentration of *L. monocytogenes* at the beginning of the shelf-life of the RTE product (time = 0), pH, a_w, washing conditions, cold smoking conditions, etc.).

- Once identified, adequate controls of these process parameters must be implemented and documented to provide evidence that the food safety outcome is not compromised.

  Note: The same control measures must apply to the process parameters that were used when the challenge studies were performed. For example:

  - The production process parameters are the same; controls are monitored and documented.

  - The RTE product is tested regularly for *L. monocytogenes* at the beginning of the shelf-life of the RTE product (time = 0) to demonstrate that the levels do not exceed the initial inoculum which was used to perform the challenge study, in other words, the inoculum concentration of 10-30 CFU/g. This verifies the acceptability of the challenge study, thus confirming the validation status. If the

results at the beginning of the shelf-life of the RTE product indicate that the levels of *L. monocytogenes* (in other words, time = 0) exceed the inoculation level which was used to perform the challenge study, the categorization of the RTE food may be compromised. An opinion from the Bureau of Microbial Hazards (BMH), Food Directorate, Health Products and Food Branch, Health Canada may be requested. A subsequent challenge study, inoculated at this higher contamination level, may need to be performed by the processor/importer, as applicable.

d) Modeling:

It might not be practical to conduct challenge studies for each RTE product being manufactured, and therefore it could be acceptable to group similar RTE products (for example, same product type with distinct spice combinations for different clients) and conduct challenge studies using the RTE product in the group which represents the highest risk for *L. monocytogenes*. A rationale should be provided to justify the grouping of RTE products, as well as the selection of the highest risk RTE product within the group. The output data of the selected mathematical model[5], using the physico-chemical parameters of the highest risk RTE product for *L. monocytogenes* in each specific grouping, should be compared to the output data of the challenge testing performed on the same RTE product, at the same experimental storage temperature. It is expected that the challenge testing data would indicate more limited growth of *L. monocytogenes*, throughout the stated shelf-life, than the predicted mathematical modeling data, especially if the modeling was done in broth under ideal growing conditions. The output data obtained from the selected mathematical model for all RTE products, under each respective grouping, must also be presented and compared to the data (mathematical modeling and challenge testing) obtained for the highest risk RTE product. The rationale and the modeling documentation will be evaluated and the classification of the RTE products, which have been tentatively grouped into Category 2A, will be confirmed by the relevant regulatory authority.

4. Validation procedure for Category 2B RTE foods

The following RTE products are considered to be classified as Category 2B, since they do not support the growth of *L. monocytogenes* (Codex Alimentarius Commisssion, 2009):

- RTE products with the following pH and a_w values:
  - pH < 4.4, regardless of a_w;
  - a_w < 0.92, regardless of pH;
  - combination of pH < 5.0 and a_w < 0.94.

- Frozen (until consumption) RTE foods.

[5] Health Canada does not endorse specific mathematical models. Any model that is published and has been peer-reviewed could be considered acceptable.

Validation is **NOT** required for RTE products that consistently meet the above mentioned physico-chemical parameters/storage conditions throughout their stated shelf-life.

Validation information is required for all other RTE products being considered for classification into Category 2B.

4.1 Pre-validation tasks for Category 2B RTE foods

a) Hazard identification:

The presence of unacceptable levels of *L. monocytogenes* in RTE foods as per the requirements of Health Canada's *Policy on Listeria monocytogenes in Ready-to-Eat Foods* (Health Canada, 2011). In other words, the presence of more than 100 CFU/g of *L. monocytogenes* at any time throughout the stated shelf-life of the RTE product.

b) Identification of the food safety outcome:

The growth of *L. monocytogenes* will be less than a 0.5 log CFU/g increase throughout the stated shelf-life of the RTE product.

c) Identification of the measure(s) that need to be validated:

Challenge studies need to demonstrate that when processed, according to the parameters specified, *L. monocytogenes* cannot grow (in other words, the number of *L. monocytogenes* increase by less than 0.5 log CFU/g) throughout the stated shelf-life of the RTE product.

Examples of control measures that need to be validated:

- Use of permitted antibacterial agent(s) (Health Canada, 2012d) and/or water activity depressants (for example, sugar, salt, etc.) and/or pH reducing agents (for example, vinegar, concentrated lemon juice, etc.) in the formulation of the RTE product, so that the growth of *L. monocytogenes* is no longer supported throughout its stated shelf-life (in other words, numbers of *L. monocytogenes* increase by less than 0.5 log CFU/g). Each ingredient must be identified, along with the effective concentration (within permitted level, if applicable) at which it will be used.
    - Once validated, the process control for the addition of ingredient(s) (for example, permitted antibacterial agent(s) (Health Canada, 2012d)) must be implemented, monitored and documented. For processors using a HACCP system, this step is expected to become a Critical Control Point (CCP) in the HACCP plan.
- Use of a specific multi-hurdle approach. Each hurdle, for example, pH, a_w, presence of antibacterial agent(s), needs to be detailed and explained. The combined use of hurdles must be designed so that it results in the production of a RTE food that no longer

supports the growth of *L. monocytogenes* throughout its stated shelf-life (in other words, numbers of *L. monocytogenes* increase by less than 0.5 log CFU/g).

- Once validated, the process parameters of each hurdle must be implemented, monitored and documented. They are expected to be covered under process control(s). For processors using a HACCP system, the(se) process control(s) is/are expected to become (a) CCP(s) in the HACCP plan.

4.2 Validation process for Category 2B RTE foods

a) Literature review:

- It is recommended that a literature review be completed with regards to the growth profile of *L. monocytogenes* in RTE product(s) under consideration, as well as similar RTE products.

- Pertinent publications should be included.

b) Challenge studies:

- Challenge studies should be conducted as per Health Canada's recommendations found in Health Canada's document *Listeria monocytogenes Challenge Testing of Refrigerated Ready-to-Eat Foods* which suggests experimental designs that consider the following parameters (Health Canada, 2012c):
  - *Listeria monocytogenes* strains
  - Preparation and enumeration of cells (maintenance of cultures and inoculum preparation as well as inoculum level)
  - Sampling design (note: a minimum of three lots of products must be tested for *L. monocytogenes* in triplicate at each sampling time (i.e., minimum of 5 time points throughout the stated shelf-life of the product, including time zero and at end of shelf-life))
  - Preparation of food products
  - Inoculation of food products
  - Special product packaging conditions
  - Incubation of inoculated food products (note: validation studies for refrigerated RTE foods must be performed at a temperature of 7^{o}C or above)
  - Enumeration and enrichment methods
  - Documentation of results

- The choices made when performing the challenge studies must be detailed and rationale should be provided, for example, the selection of strains that will be used for inoculation, the initial level of inoculation, the number of tests performed, etc. (Health Canada, 2012b).

- The result of the challenge studies must indicate that the food safety outcome is met, in other words, that the growth of *L. monocytogenes* is less than 0.5 log CFU/g increase throughout the stated shelf-life of the RTE product.

c) Identification of key process parameters, and provision of evidence of their control:

- Key process parameters need to be clearly identified (for example, pH, a_w, concentration of antibacterial agent(s) used, etc.).

- Once identified, adequate controls of these process parameters must be implemented and documented to provide evidence that the food safety outcome is not compromised.

- These key process parameters are specific to the control measure being validated. For instance:

    - When validating the use of ingredients (for example, permitted antibacterial agent(s) (Health Canada, 2012d) which act to limit the growth of *L. monocytogenes* on/in the RTE product to the specified outcome (in other words, less than 0.5 log CFU/g increase of *L. monocytogenes* throughout the stated shelf-life of the RTE product), the addition of the such ingredient(s) is/are expected to become a process control (for example, a CCP when a HACCP plan is in place). At the process step(s) where these ingredient(s) is/are used (for example, an antibacterial agent during formulation (Health Canada, 2012d)), processors, must ensure that the appropriate and permitted ingredient(s) is/are used at the prescribed effective concentration(s).

    - When using a specific multi-hurdle approach, each hurdle parameter is expected to be covered under a process control, as well as any interactions between the different hurdles, if applicable, for example in a time sequence (for example, a CCP when a HACCP plan is in place).

d) Modeling:

It might not be practical to conduct challenge studies for each RTE product being manufactured and therefore it could be acceptable to group similar RTE products (for example, same product type with distinct spice combinations for different clients) and conduct challenge studies using the RTE product in the group which represents the highest risk for *L. monocytogenes*. Rationale should be provided to justify the grouping of RTE products, as well as the selection of the highest risk RTE product within the group. The output data of the selected mathematical model[6], using the physico-chemical parameters of the highest risk RTE product for *L. monocytogenes* in

[6] Health Canada does not endorse specific mathematical models. Any model that is published and has been peer-reviewed could be considered acceptable.

each specific grouping, should be compared to the output data of the challenge testing performed on the same RTE product, at the same experimental storage temperature. It is expected that the challenge testing data would indicate more limited growth of *L. monocytogenes*, throughout the stated shelf-life, than the predicted mathematical modeling data, especially if the modeling was done in broth under ideal growing conditions. The output data obtained from the selected mathematical model for all RTE products, under each respective grouping, must also be presented and compared to the data (mathematical modeling and challenge testing) obtained for the highest risk RTE product. The rationale and the modeling documentation will be evaluated and the classification of the RTE products, which have been tentatively grouped, into Category 2B, will be confirmed by the relevant regulatory authority.

5. Conclusion

Before starting a challenge study, it may be a good idea to consult with the Bureau of Microbial Hazards for guidance on the acceptability of the proposed approach (for example, protocol, repeatability, etc.) (Health Canada, 2012b; Health Canada, 2012c). Thereafter, processors and importers, as applicable, should submit their validation documentation to the relevant regulatory authority (for example, the Canadian Food Inspection Agency, provincial or territorial government). The validation documentation will be evaluated and the final classification of the RTE product will be confirmed by the relevant regulatory authority (Health Canada, 2012a; Health Canada, 2012b). Please note that the confirmed classification of RTE products must be available at all times to the regulatory authorities.

After validation, if a change is intended or is made to a control measure or in a product- or process-related factor that is likely to affect the effectiveness of the control measure (for example, process control, process parameters, etc.) the appropriateness of the validation studies supporting the categorization of the RTE food product under consideration may need to be confirmed. If the change is determined by the relevant regulatory authority to be substantial, the need for re-validation/re-categorization may be triggered (CAC, 2008). On-going monitoring and verification procedures are key in providing reassurance that the food safety outcome is maintained.

Other approaches to validation may be considered by the regulatory authorities. Proposals to use different approaches would need to be evaluated by the relevant regulatory authority to determine whether they are appropriate, prior to commencing the scientific research. As stated previously, provinces and territories may also be involved in the validation process.

6. References

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Health Canada (2012c). *Listeria monocytogenes* Challenge Testing of Refrigerated Ready-to-Eat Foods.

Health Canada (2012d). Food Additives that May Currently be Used as Class 2 Preservatives to Potentially Control the Growth of *Listeria monocytogenes* in Ready-to-Eat Foods Sold in Canada.