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JOINT FAO/WHO FOOD STANDARDS PROGRAMME

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PRACTICAL EXAMPLES ON THE APPLICATION AND ESTABLISHMENT OF MICROBIOLOGICAL CRITERIA

Examples prepared by drafting teams

Introduction

1. Seven examples on the application of microbiological criteria have been developed to help illustrate the various contexts in which microbiological criteria may be developed and applied in the context of food safety management. The primary purpose of these examples was to facilitate the revision of the principles and guidelines for the application of microbiological criteria.

2. The examples were considered by the physical Working Group (pWG) on the revision of the Application and Establishment of Microbiological Criteria (*see* CX/FH 12/44/6).

3. The pWG concluded that all examples, prepared by the drafting teams had been useful to revise the main document, which was now ready to be considered by the 44th Session of the CCFH for finalisation and to be forwarded to the Commission for adoption.

4. The pWG agreed to request the drafting teams to undertake some additional work on the practical examples to ensure that they have a harmonized structure; use consistent language and, where possible, are consistent with the main document. The pWG also asked the drafting teams to include in each example: (i) a brief introduction; (ii) a detailed indication of the type of food covered by the example; and (iii) a sentence to indicate that the examples were not peer-reviewed.

5. The pWG agreed that the compilation of examples would be circulated as a separate document for the 44th CCFH, and that the CCFH discuss how the examples should be made available. In this regard, the pWG requested the Codex Secretariat, in collaboration with FAO/WHO, to include in the document a list of potential options for making the examples available (*see* Appendix I) and to renumber the examples from 1 to 7 (*see* Appendix II).

6. The pWG recommended the 44th CCFH discuss how to use and where to locate the examples developed by the drafting teams.

Appendix I

RECOMMENDATIONS

Option 1: The examples are posted on the website of FAO and/or WHO Food Safety Programme

1. The benefits of this option are as follows:
   a. The examples are freely available to all interested parties.
   b. The examples would undergo a quality control process in the form of a peer review.
   c. All authors would be directly acknowledged and recognized.
   d. The examples could be made available in the context of other resource material related to sampling and food safety management and with a direct link to the revised Codex text, e.g., the examples related to poultry could be made available together with other documents and tools relating to the management of pathogens in poultry.
   e. There would be an opportunity to update the examples and/or add more examples in future.
2. The negatives for this option are:
   a. Additional work would be required by the drafting groups who prepared the examples after the peer review.
   b. The examples would not be on the Codex website.

Option 2: Codex archives

3. The benefits of this option are as follows:
   a. The examples would be available on the Codex website as part of the meeting archive.
   b. No further work would be required by the drafting groups.
   c. All authors would be directly acknowledged and recognized.
4. The negatives for this option are:
   a. The examples would not undergo any further quality control.
   b. There would be no opportunity for future updating of the examples.
   c. Apart from the related Codex text the examples would not be linked to any other resource which could provide additional context.

Option 3: Include as annexes to the Codex Principles and Guidelines for the Establishment and Application of Microbiological Criteria for Foods

5. For this option, the examples would be available together with the Codex text.
6. The negatives for this option are:
   a. The details of the examples would have be to be reviewed and agreed by the Committee.
   b. Further review and simplification of the texts would be required by the drafting groups.
   c. Inclusion in a Codex document could be understood by some countries that these examples have status in the context of the SPS Agreement.
   d. The development of the examples was to support the development of the text and were designed for and have already served that purpose.
   e. There is a bias towards certain pathogen commodity combinations and there is a risk of this being misinterpreted as particular importance of certain pathogen commodity combinations.

Appendix II

PRACTICAL EXAMPLES OF MC AND APPLICATIONS

This Appendix contains examples of several approaches MC and its applications. All of the examples described below are for purposes of illustration only, do not represent actual MC application scenarios in a global sense and should not be replicated as presented.

EXAMPLE 1: A GHP-BASED MICROBIOLOGICAL CRITERION

Introduction

A good hygiene practice (GHP)-based microbiological criteria is a criterion set for a specific stage of the production process, which can be used by the food business operators and competent authorities to monitor and verify that good hygiene practices and HACCP-procedures are being followed as expected.

Purpose (what is intended to be achieved);

1. The purpose of a GHP-based microbiological criterion is to assess the acceptability of a production or manufacturing process. A GHP-based microbiological criterion is used to verify that the production process of the establishment is functioning as expected and that good hygiene practices are correctly implemented and have been followed.

Who should establish and who should apply;

2. The GHP-based criterion could be established by the competent authority through legislation, guidance documents or other national or international standards (e.g. Codex). The competent authority could consult the sector when developing the criteria.

3. A GHP-based microbiological criterion should be implemented by the food businesses operator when developing their own food safety management system (e.g. GHP and HACCP-plan).

4. The criterion should apply to industry (e.g. abattoirs, cutting plants), but could also be adapted to other size enterprises (e.g. medium size butchers, craftsmen, etc).

5. The GHB-based microbiological criterion could be used by the competent authority when assessing food businesses operators.

Food or food process; Point in food chain where the MC is applied;

6. The GHP-based criterion of this example applies to the manufacturing or production process of meat preparations (fresh meat which has had foodstuffs, seasonings or additives added to it). This GHP-based MC applies only during the production process (preferably at the end of the production of the meat preparation), but does not apply to meat preparations already placed on the market or further transformed in other meat products that have undergone a heat treatment.

Organisms of concern;

7. When developing GHP-based MC, hygiene indicators should preferably be chosen. In this example *E. coli* is chosen as an indicator of faecal contamination during the manufacturing process.

8. Other hygiene indicator micro-organisms (e.g.: aerobic colony count, Enterobacteriaceae, coliforms, coagulase-positive staphylococci, etc.) could also be chosen when describing a GHP-based MC.

Sampling plan (# of samples, sample size/units, sampling approach);

9. The sampling plan (nr of samples, sample size/units, frequency of sampling) and the sampling limits of the GHP-based criterion should be chosen according to the expected conditions in which the foodstuff will be handled and consumed.

10. In this example of GHP-based MC a 3-class attribute sampling plan is used. This plan is defined by the values n, c, m and M. The quality of the product or the process can be divided into 3 attribute classes:

- Unacceptable quality;

- Acceptable quality;

- Satisfactory quality.

11. The sampling plan, number of samples, the sample size/units and the microbiological limits of the GHP-based MC should be chosen according to the production process, historical data, and the expected conditions in which the food will be handled and consumed. In cases where there is no previous experience in the microbiological criterion, microbiological data and scientific information could be used when choosing these parameters. The sampling plan and the microbiological limits could be evaluated after a period and in the light of the obtained results and the effectiveness of well-functioning HACCP-based procedures, could be reviewed and adapted accordingly.

In this example of GHP-based microbiological criterion for meat preparations, the following sampling plan, size and limits has been chosen as a starting point. Other microbiological limits or sampling size can be considered based e.g. on the kind of meat preparation, the microbiological data gathered at establishments, etc.

Sampling plan

n = number of units comprising the sample to be taken (n = 5)

c = maximum number of sample units giving values between m and M (c = 2)

Sample size = g (10 g)

Microbiological limits

M = upper microbiological limit (M = 5000 cfu/g)

m = lower microbiological limit (m = 500 cfu /g)

12. Sampling approach: The frequency of sampling could be stated by legislation or could be chosen by the food businesses operator depending on the volume of production, etc. Sampling frequency should be increased or decreased according to performance or changes in the manufacturing process. For example, it could be started with a sampling frequency of once a week. This frequency could be reduced after satisfactory results during a long period of time. When the production process change, a new provider is introduced, etc. the sampling frequency should be increased to the initial level.

Method(s) of analysis;

13. The analytical reference method of the GHP-based MC should preferably be an internationally recognised method (e.g. ISO, AOAC, etc.). Alternative analytical methods may be used when validated against the reference method.

14. In this example, ISO 16649-1 or 2 is chosen as the analytical reference method for *E. coli*.

Interpretation of results;

15. The level of *E. coli* gives an indication of the faecal contamination. The results of the analysis demonstrate the microbiological quality of the product or process tested. The quality can be divided into 3 attribute classes depending upon the level of contamination or the concentration of micro-organisms within the samples.

- Unacceptable quality: the result of one or more of the 5 units comprising the sample exceeded the value M (1 or more of the 5 unit samples exceed 5000 cfu *E. coli* /g)
- Acceptable quality: maximal c samples did exceed the level of m, but less than M. Maximal 2 of the 5 units comprising the samples have a result between 500 and 5000 cfu/g. The other 3 units of the sample give a result below 500 cfu *E. coli*/g.
- Satisfactory quality: the concentration of all units comprising the sample did not exceed the value m. The level of all the sample units did not exceed the level of m (all 5 unit samples are under 500 cfu *E. coli*/g).

(Nature of) actions in case of non-compliance.

16. Corrective measures should be taken by the food businesses operators if certain levels of contamination or trends indicating unsatisfactory results are detected. The corrective actions should be

focused on the improvement of the production hygiene and/or the selection of raw materials. The corrective actions should be proportionate to the risk involved and should be described in the food safety plan by the FBO.

17. Examples of corrective actions in case of non-compliance with the GHP-based criterion for meat preparations would focus on the review and improvement of: cleaning and disinfection plan, quality of the raw materials, transport system and materials, personal hygiene, production process, HACCP.

EXAMPLE 2: MICROBIOLOGICAL CRITERION TO ASSESS THE ACCEPTABILITY OF A FOOD LOT

(This example has not been peer reviewed)

Introduction

1. The primary objective of this example is to provide information to governments and industry on establishing and applying a microbiological criterion (MC) for food to assess the acceptability of a food lot. For this example, we have selected milk powder intended for direct consumption (i.e., milk powder that will be consumed without further treatment to inactivate microorganisms). This would include milk powder used to manufacture another product for which there is no microbial inactivation step in the production of that product (e.g., a whipped topping, a seasoning blend).

2. Competent authorities could use the criteria for testing milk powder for import/export or as part of domestic food control procedures. The criteria can be applied by a food business operator as a verification procedure for milk powder manufactured by that food business. Food business operators could also use the criteria for accepting from a supplier milk powder that will be used for manufacturing other products that will not receive a treatment that would inactivate pathogens prior to consumption. When food business operators are purchasing milk powder from a supplier, the testing could be performed for acceptance of each lot or as periodic verification of the supplier's controls, depending on the confidence in the supplier's control procedures.

3. A manufacturer may conduct periodic verification testing in accordance with the criteria below, but if such testing indicates a problem, the manufacturer may determine that other criteria may be appropriate (such as n=20 for *Salmonella*). Other criteria may also be appropriate when there is an unusual event such as construction or the need for wet cleaning in a dry milk facility.

Purpose

4. The purpose of these microbiological criteria is to assess the acceptability of a milk powder lot intended for direct consumption. (Milk powder will be consumed without further treatment to inactivate microorganisms.)

Who should establish and who should apply

5. Established by: Competent authorities or food business operators

6. Applied by: Competent authorities or food business operators

Food or food process (point in food chain where the MC is applied)

7. Milk powder at the manufacturing facility or in commerce for verification of lot acceptability by competent authorities as part of domestic food control procedures

8. Milk powder received for import/export inspection by competent authorities

9. Milk powder for lot acceptance by food business operators purchasing from a supplier

10. Milk powder at point of manufacture as a verification of process control

Organisms of concern

11. Hygiene Criteria:

- Mesophilic Aerobic Microorganisms
- Enterobacteriaceae

12. Food Safety Criterion

- *Salmonella* spp.

13. Epidemiological data suggest that the only significant hazard to be controlled during manufacturing of dried products is *Salmonella*. Other hazards such as *Staphylococcus aureus* or *Bacillus cereus* are only present sporadically at very low levels or occur as a result of major breakdowns of GHP. The presence of preformed staphylococcal enterotoxins or *B. cereus* emetic toxin would only be present if a major GHP breakdown resulted in growth to high levels. Low levels of these two bacteria ($<10^2$ CFU /g) do not

represent a risk to human health as long as the products are not mishandled after reconstitution and before consumption.

14. Mesophilic Aerobic Microorganisms are used as an index of utility, as indicators of general contamination, shelf life or spoilage, and are not usually related to a health hazard).

15. Enterobacteriaceae are tested as indicators of the history of the hygiene of the food production process. Significant numbers of them frequently indicate inadequacy of general hygiene.

Sampling plans (# of samples, sample size/units, sampling approach)

16. The size of the sample from which the analytical unit will be obtained should be approximately 100g. In order to maintain the integrity of the sample until arrival at the laboratory, milk powder should be transported at room temperature with a maximum of 30 ° C.

Organism	**Size of Analytical Unit**	**Sampling Plan**		**Limits**		**Class Plan**
		n	**c**	**m**	**M**	
Mesophilic Aerobic Colony Count	10 g	5	2	1 x 10^4 CFU/g	1 x 10^5 CFU/g	3
Enterobacteriaceae	10 g	5	2	<3 MPN/g (none detected)	9.4 MPN/g	3
Salmonella spp.	25 g	10	0	Not detected in 25 g		2

17. Where:

n = number of analytical units to be analyzed

c = the maximum allowable number of non-conforming analytical units in a 2-class plan or marginally acceptable analytical units (i.e., between m and M) in a 3-class plan;

m = a microbiological limit which, in a 2-class plan, separates conforming analytical units from non-conforming analytical units or, in a 3-class plan, separates conforming analytical units from marginally acceptable analytical units; and

M = a microbiological limit which, in a 3-class plan, separates marginally acceptable analytical units from non-conforming analytical units.

(Note: In a 3-class plan conforming is ≤ m and marginally acceptable is > m but ≤ M. In the case of a 2-class plan based on counts, a lot will be rejected if the analytical unit is greater than or equal to m (when c=0).)

Performance Characteristics of the sampling plans:

18. Mesophilic Aerobic Colony Count

n	c	m	M	Probability of lot rejection	stdev* = 0.25 (log_{10} cfu/g)	stdev = 0.50 (log_{10} cfu/g)	stdev = 0.80 (log_{10} cfu/g)	stdev = 1.2 (log_{10} cfu/g)
					Geometric mean concentration (log_{10} cfu/g)			
5	2	10^4 cfu/g ($4 log_{10}$ cfu/g)	10^5 cfu/g ($5 log_{10}$ cfu/g)	0.95	4.22	4.40	4.52	4.59

*stdev = standard deviation

19. Interpretation of the gray cell above: Assuming a log normal distribution, this sampling plan will provide 95% probability that a lot of food containing a geometric mean concentration of 4.22 log cfu/g and a standard deviation of 0.25 log cfu/g will be rejected. The arithmetic mean concentration of such a lot is 4.29 log cfu/g.

20. Enterobacteriaceae

n	c	m	M	Probability of lot rejection	stdev* = 0.25 (log_{10} cfu/g)	stdev = 0.50 (log_{10} cfu/g)	stdev = 0.80 (log_{10} cfu/g)	stdev = 1.2 (log_{10} cfu/g)
					Geometric mean concentration (log_{10} cfu/g)			
5	2	<3MPN/g (<0.48 log_{10} cfu/g)	9.4 MPN/g (0.97 log_{10} cfu/g)	0.95	0.68	0.76	0.78	0.77

*stdev = standard deviation

21. Interpretation of the gray cell above: Assuming a log normal distribution, this sampling plan will provide 95% probability that a lot of food containing a geometric mean concentration of 4.79 cfu/g (0.68 log cfu/g) and a standard deviation of 0.25 log cfu/g will be rejected. The arithmetic mean concentration of such a lot is 5.65 cfu/g.

22. *Salmonella* spp.

n	c	m	Probability of lot rejection	stdev* = 0.25 (log_{10} cfu/g)	stdev = 0.50 (log_{10} cfu/g)	stdev = 0.80 (log_{10} cfu/g)	stdev = 1.2 (log_{10} cfu/g)
				Geometric mean concentration (log_{10} cfu/g)			
10	0	absence in 25 g	0.95	-1.97	-2.08	-2.25	-2.49

*stdev = standard deviation

23. Interpretation of the gray cell above: Assuming a Poisson-log normal distribution, this sampling plan will provide 95% probability that a lot of food containing a geometric mean concentration of 0.011 cfu/g (-1.97 log cfu/g) and a standard deviation of 0.25 log cfu/g will be detected and rejected if any of the 10 analytical units are positive for *Salmonella*. The arithmetic mean concentration of such a lot is 0.013 cfu/g. Note that such a lot may consist of 74% of the 25 g analytical units being negative and up to 26% of the analytical units being positive for *Salmonella*; 0.38% of this lot could contain concentrations above 0.05 cfu/g.

Method(s) of analysis

24. Mesophilic aerobic colony count: ISO 4833

25. Enterobacteriaceae: ISO 21528-1 (MPN technique)

26. *Salmonella* spp.: ISO 6785 (Milk and milk products -- Detection of *Salmonella* spp.) or ISO 6579 (Horizontal method for the detection of *Salmonella* spp.)

27. NOTE: The most recent edition of the standards should be used. Other methods that provide equivalent sensitivity, reproducibility, and reliability can be employed if they have been appropriately validated.

Interpretation of results

28. *Salmonella* spp.

- satisfactory if all values observed indicate the absence of the bacterium
- unsatisfactory if the presence of the bacterium is detected in any of the sample units

29. Mesophilic aerobic colony count and/or Enterobacteriaceae

- satisfactory, if all the values observed are $\leq$ m
- acceptable, if a maximum of c units have values that are between m and M and the rest of the values observed are $\leq$ m
- unsatisfactory if one or more of the values observed are > M or more than c units are between m and M

Actions in case of non-conformance

Food business operators:

30. Non-conformance with *Salmonella* spp. criterion:

- Food business operators purchasing from a supplier: (1) Notify the supplier; (2) do not use the milk powder, or if the milk powder has been used do not ship the product; (3) if product has been shipped, recall the product; and (4) determine appropriate steps with respect to the supplier.
- Food business operators manufacturing the milk powder: (1) prevent the affected lot from being released for human consumption; (2) recall the product if it has been released for human consumption; and (3) determine and correct the root cause of the failure.

31. Non-conformance with mesophilic aerobic microorganisms and/or Enterobacteriaceae:

- Food business operators purchasing from a supplier: (1) Notify the supplier; (2) determine appropriate disposition of the non-conforming lot (e.g., refuse lot or accept marginal quality lot, depending on business contractual arrangements)
- Food business operators manufacturing the milk powder: (1) Check on the efficacy of heat treatment and controls for prevention of recontamination, (2) determine and correct the root cause of the failure: (3) consider the need for pathogen verification testing; and, (3) as appropriate, review and revise monitoring procedures, environmental surveillance and prerequisite programs. Determine disposition of lot (which may include an alternative use for the milk powder).

Competent authorities:

32. Non-conformance with *Salmonella* spp. criterion:

(1) prevent the affected lot from being released for human consumption;

(2) ensure recall of product if it has been released for human consumption;

(3) reject lot at port of entry

34. Non-conformance with mesophilic aerobic microorganisms and/or Enterobacteriaceae

(1) determine whether to reject lot at port of entry (e.g., destroy at port of entry or return to country of origin), allow reconditioning of lot, or use of lot for other purposes

(2) notify the competent authority in the country of origin so the manufacturing facility can take corrective actions with respect to hygiene practices and verify the efficacy of heat treatment and procedures to prevent recontamination

(3) reject lot as part of domestic food control procedures and determine disposition of the lot, e.g., require destruction, allow reconditioning of lot, or use of lot for other purposes

EXAMPLE 3A: MICROBIOLOGICAL CRITERIA (MC) FOR VERIFYING THE PERFORMANCE OF A HAZARD ANALYSIS CRITICAL CONTROL POINT (HACCP) SYSTEM BY THE FOOD BUSINESS OPERATOR (FBO)

Introduction

1. A HACCP system is built on prerequisite programs (PRPs) in place and on management awareness and commitment of the FBO. Consequently, the implementation, application and performance of a HACCP system depend also on the effectiveness of chosen prerequisite programs. An MC applied for the verification of the performance of a HACCP system can be used to verify that the prerequisites for that plan are functioning as presumed.

2. In a food business, microbiological testing and the related MCs are applied at various locations covered by the scope of the food safety system, e.g. the raw materials, the ingredients and/or processing aids, at appropriate steps along the process, and/or for food contact or other surfaces of equipment and/or premises, and/or for food at the point where it is put onto the market.

3. However, sampling locations along the food processing line is correlated to the parts of the system that are to be verified. Sampling of end products can be used to verify the performance of the entire food safety system, whereas sampling earlier in the process flow are typically used to verify the performance of specific control measure(s) or steps.

For example:

- An MC for *B. cereus* spores in purchased dried spice can be applied to verify that the supplier meets communicated or expected specifications
- An MC for *Enterobacteriaceae* on 100 cm2 surface can be applied to verify that the cleaning procedure of an open system is effective
- An MC for *S. aureus* in fresh cheese can be applied to verify that the combination of all the hygiene procedures applied up to that particular process step, keep *S. aureus* under control.
- An MC for thermoduric bacteria in packaged processed cheese can be applied to verify that the entire food safety system is in control.

2. General application

2.1 Purpose of the MC (what is intended to be achieved)

4. The purpose of the MC (or multiple MC) for this example is to verify the intended performance over time of a plant & product specific food safety control system managed by a HACCP system, through the use of the moving window approach as outlined in section 4.9 of the main document.

2.2 Who should establish and who should apply

5. MCs used to verify the performance of a HACCP system are normally established by the FBO, i.e. the HACCP team. The hazard analysis and subsequent design of the HACCP plan provide the rationale for selecting microbiological testing as the means of verification.

6. It is the FBO that applies the MC to its process. However, competent authorities may apply the same or different MC to support an audit.

2.3 Food or process to which the MC applies

7. This type of MC is applied for verification of the HACCP system and is based on analysis of food samples.

2.4 Specified point in the food chain where the MC applies

8. End products prior to release to the market.

2.5 Microorganism(s) and the reason for their selection

9. Microorganisms of concern for the purpose of the MC are (i) the pathogen(s) that is(are) intended to be controlled by the HACCP system and/or indicators for the presence of the pathogen(s), and (ii) indicators used to document specific or general hygiene control.

10. The hazard analysis and subsequent design of the HACCP plan can identify whether microbiological testing is the appropriate means of verification, and if so, which microorganism(s) that would be give meaningful results, if tested.

11. Where the probability of detecting a significant hazard (pathogen) is very low, alternative means of verification is more meaningful, such as relying on process criteria (requires validation against the pathogen) or use of indicator organisms (e.g. total plate count, *Enterobacteriaceae*).

***Note**: MC for spoilage organisms is usually established by the FBO to verify adherence to quality control specifications. Control of spoilage organisms is not within the scope of HACCP, although the concept can be applied to their control.*

2.6 Microbiological limits (m, M)

12. Acceptable levels expressed by MCs established by regulation (competent authorities) should be used, where applicable, and when microbiological testing is chosen as the means of verification of a HACCP system.

2.7 Sampling plan (sampling approach, frequency and adequacy)

13. The approach of the moving window is based on taking a defined number of samples at a specified frequency over a specified or defined time period or over a defined volume of production or processing lines.

Adequate sampling frequency depends on:

A) The number of lines subjected to the verification.

Two or more processing lines can be pooled provided that they do not differ in terms of level of pathogen control and that they process similar products.

B) Sufficient production frequency.

The moving window approach is intended for frequent (e.g. daily) production; it is not adequate if production frequency is too low (e.g. once a month).

C) Distribution of the microorganism in the food.

Food products with relatively higher standard deviation should be sampled more frequently (e.g. portioned solid products).

D) Probability of detection and analytical method.

The probability of detecting the target organism in the food and the detection level of the analytical method must be accounted for when establishing the frequency of sampling and the length of the moving window.

2.8 Method(s) of analysis

14. The appropriate analytical method used, including any confirmatory tests applied, will depend on the chosen type of limit, (absence/presence, MPN or direct colony counting) and the organism. The choice of analytical method must correspond with the nature of the verification procedure and of the planned action.

2.9 Interpretation of results

15. A criterion for conformity or non-conformity with the planned performance of the food safety control system is typically established as a specified maximum frequency of exceeding a level during a specified period, but never above an absolute maximum level (e.g. in the case of a three-class MC, the number "c" samples out of "n" samples taken during a month that may exceed the limit "m", but not the limit "M").

16. This is illustrated hypothetically below. The number of sampling times equals the number "n" of the MC. The "window" is the picture obtained when "boxing" the most recent "n" sampling times. When a new sample is taken, the "window" moves one step to the right, always "boxing" the last "n" samples. The figure shows the approach for a 3 class MC of n=5; c=2. Two "windows" showing non-conformity are highlighted: The left one is non-conforming because "m" is exceeded more than "c= 2" times and the right is non-conforming because M is exceeded.

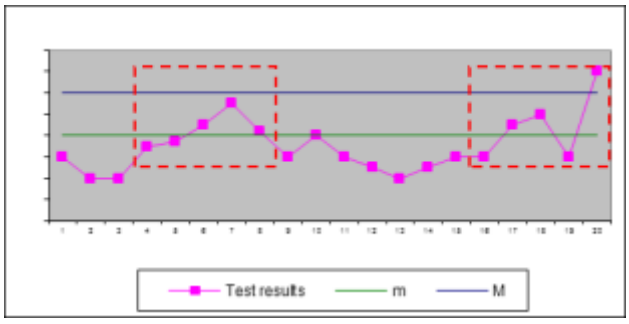


Figure 3-1. Approach for a 3 class MC of n=5; c=2.

2.10 Actions* to be taken when the MC is not met:

17. The MC typically includes action(s) to be applied in case of non-conforming individual results as well as distinctive action(s) to be applied in case of non-conforming results obtained over time.

> ***Note**: These "actions" applicable to verification results should not be confused with "corrective actions" applied to monitoring, i.e. to exceeding critical limits of CCPs.

Action on individual results is triggered by exceeding the limit "M" of a 3 class MC or the limit "m" of a 2 class MC. In case of pathogens, the action will include handling the affected lot as a potentially unsafe product, which can lead to a withdrawal/recall. In case of hygiene indicators, the action will include further assessment of the affected lots, such as reviewing the monitoring records of that lot, carrying out lot-by-lot testing for pathogens that relate to the indicator, etc. Exceeding "M" for a hygiene indicator does not trigger a withdrawal/recall on its own. The action is targeted on the lot exceeding the limit, not previous lots in the same "window".

Action on results over time (the full window) is used for 3 class MC and is triggered if an unacceptable number of samples exceeding "m". Such non-conformity is most likely caused by a non-functioning system. The action is therefore not to be targeted at any specific lots, but at the HACCP system. The action can include a review of and consequential changes to:

- the monitoring system (it may be inadequate to detect loss of control),
- the effectiveness of prerequisite programs (e.g. cleaning procedures), and/or
- the effect of control measures at CCPs and the corresponding critical limits (revalidation may be needed).

18. For pooled samples, corrective actions should be taken on those processing lines, spots or product types, which are represented by the pooled sample. A first action could be to analyze non-pooled samples to track the source.

19. A higher frequency of verification should be considered for a short period to verify the effect of the action.

3. Hypothetical illustration of the approach

20. It is to be emphasized that no specific MC for the purpose of verification of HACCP for a certain food is universally needed and if needed, the same limits and sampling plans may not be appropriate (it will all depend on the design of the HACCP/GHP system, in particular the degree of uncertainty in the control of the chosen organism). Therefore, the example below should not lead to misinterpretation as it being internationally harmonized nor recommended. Other target organism, sampling frequencies and limits may be more appropriate according to the system in place.

21. This example has not been peer-reviewed.

22. A FBO produces fresh cheese using one milk processing line and 3 cheese manufacturing lines. Source assessment and estimated levels of identified significant pathogens has concluded that microbiological testing is an appropriate means of verifying that *Staphylococcus aureus* is in control.

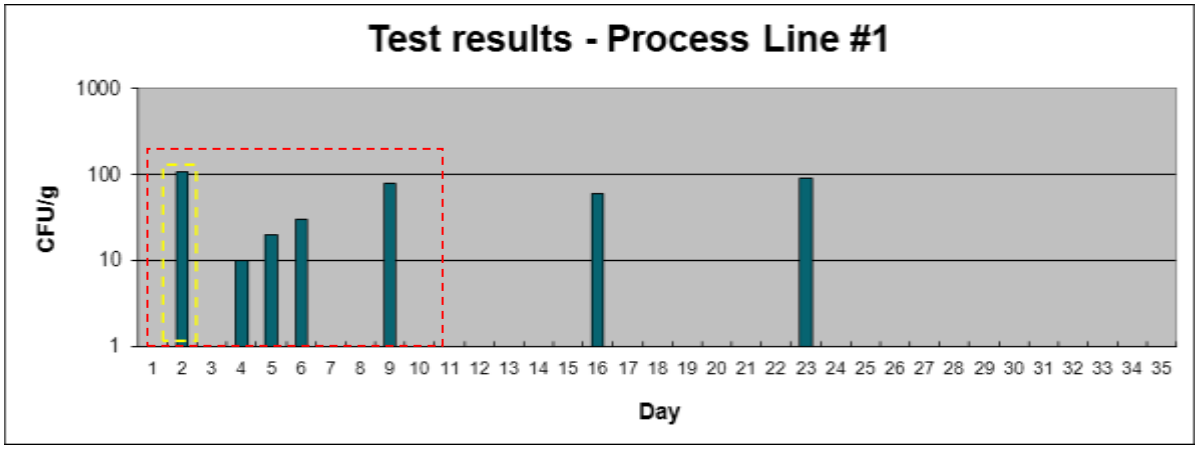
23. The cheese is to be kept refrigerated (labeling instruction). *S. aureus* may grow in the end product, if not stored correctly during shelf life.

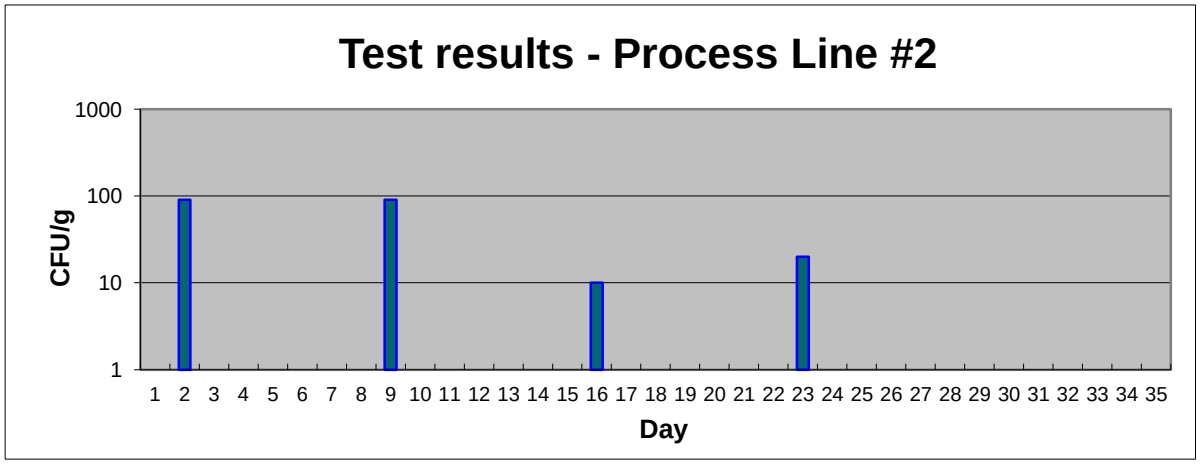
24. The control measures in place are designed to ensure that *S. aureus* concentration is low in the end product so that growth does not reach high levels during shelf life in the case that the recommended storage instructions are violated by the consumer.

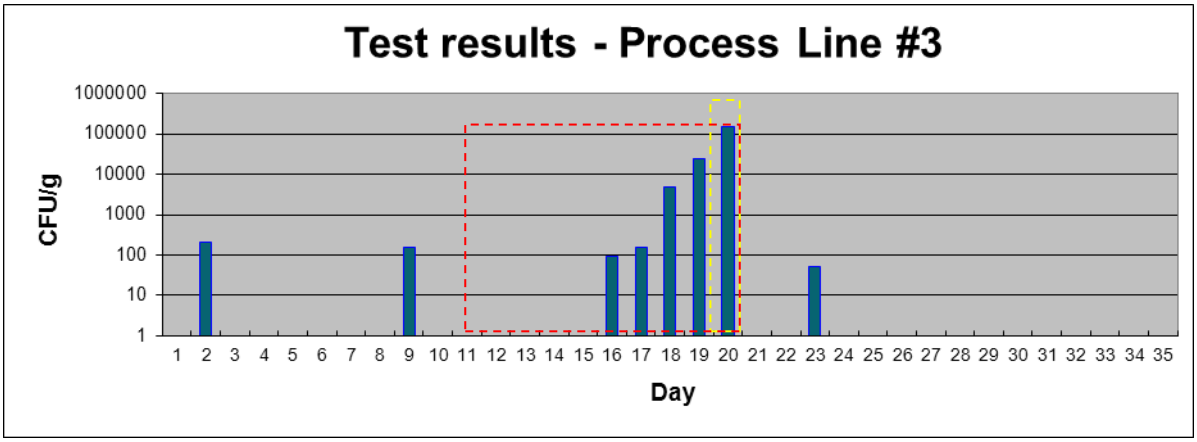
25. The MC to verify this has been established by the FBO as n=10; c=3; m=0 cfu/g; M=100 cfu/g

26. The attributes sampling plan here consists of one sample daily from each of the three manufacturing lines. This sampling approach will permit identifying loss of control in the three manufacturing lines as well as in the preceding milk processing line.

27. Results obtained during 5 weeks are shown in the figures below (analytical results showing absence not shown):







28. From the results obtained, the following was observed:

	Interpretation of results	Cause analysis	Corrective action
Line #1	The MC was not met in the period "n" consisting of days 1-10 (the "window" highlighted in red spotted frame). The limit "M" was exceeded on day 2 (110 cfu/g)	As the non-conformity occurred only in one of the three process lines, the cause is contamination during cheese manufacture and/ or packaging. Since low levels were found, it is likely that the cause might be contamination from biofilm and not for example, loss of temperature or time control.	Increase of cleaning frequency to avoid biofilm buildup. Subsequent testing showed that the issue was resolved.
Line #2	The MC was met throughout the period	Despite conformity with the MC, trend analysis carried out on day 24 showed an apparent systematic pattern in occurrence (every Monday). In fact, the pattern is the same in all three lines, which assisted in locating the problem to the steps prior to cheese manufacture that took place during weekends. Potential causes could be as follows: - biofilm in the regenerator of the pasteurizer - different staff operating procedures - different storage or collection procedures	Reinforcing staff hygiene instructions and operating procedures carried out during weekends. Reinforcing instructions on correct storage conditions (time/temperature) at farms during weekends. Subsequent testing showed that the issue apparently was resolved
Line #3	The MC was not met in the period "n" consisting of days 11-20 (the "window" highlighted in red spotted frame) The limit "M" was exceeded on day 17, 18, 19 and 20 The level of 100.000, above which enterotoxin production is possible, was exceeded on day 20 (see the yellow spotted frame)	As the non-conformity only occurred in one of the three process lines, the cause is contamination during cheese manufacture and/or packaging. The development indicates a source of severe contamination, such as a defective valve or gasket.	Check and replacement as needed, of all joints and gaskets. Testing of all batches manufactured by Line 3 on day 20 for incidence of staphylococcal enterotoxins. If detected, effect the withdrawal/recall of affected batches that have left the control of the plant. Subsequent testing showed that the issue was resolved. As enterotoxins were not detected in the batches of day 20, it was assumed that the strains of *S. aureus* present do not produce toxins in this cheese. Consequently, the product was considered safe to sell.

EXAMPLE 3B OF A MICROBIOLOGICAL CRITERION (MC) FOR VERIFYING THE PERFORMANCE OF A FOOD SAFETY CONTROL SYSTEM: *CAMPYLOBACTER* PERFORMANCE TARGET AT END OF PROCESSING OF BROILER CHICKENS

Drafting team: New Zealand (lead), Costa Rica, Kenya, Kiribati, Samoa

Introduction

1. An unacceptably-high rate of foodborne campylobacteriosis was seen in New Zealand in 2006. Attribution studies estimated that more than 50% of human cases were attributed to the consumption of poultry meat. This led to the implementation of a risk management strategy for *Campylobacter* in broiler chicken meat. Control measures were applied by the poultry industry from primary production to consumption. The *Campylobacter* performance target (CPT) was introduced by the competent authority to verify the effectiveness of these control measures in reducing levels of *Campylobacter* contamination during the slaughter and dressing of broiler chickens.

2. The CPT was developed using quantitative *Campylobacter* data collected nationally for broiler chicken carcass rinsates. The baseline distribution for the rinsates had a mean log10 Campylobacter count of 4.16 log10 cfu/carcass.

3. The competent authority and the poultry industry agreed to aim for a hazard reduction of one $\log_{10}$ decrease in the mean of the baseline distribution. Statistical advice was sought to determine how a target could be set up to deliver this. As a result, the target count (the CPT) was established to reflect the 90^{th} percentile of the target distribution curve; a value of 3.78 $\log_{10}$ cfu/carcass (or 6,000 cfu/carcass) rinsate. This also allowed pass/fail levels to be set.

4. The CPT was verified against actual data and by computer simulation prior to implementation.

5. The CPT is a hazard-based microbiological criterion aimed at assisting the competent authority in meeting a reduction in human foodborne campylobacteriosis cases.[1]. Further to the CPT being in place, trend analysis of the broiler chicken carcass rinsate results and human cases, has occurred. There is now a strong association demonstrable between the introduction of the microbiological criterion and the reduction in human foodborne campylobacteriosis in New Zealand, achieved since regulating the CPT.

6. This example shows how the MC can be used to improve food safety and assist in attaining a public health goal in regard to a particular foodborne disease.

7. New Zealand was not able to set a Food Safety Objective (FSO)[2] (as the levels of *Campylobacter* present at the point of human consumption were unknown and unregulated) or an Appropriate Level of Protection (ALOP)[3], as a risk assessment model that could be used to predict the level of protection, was not available. This also precluded the establishment of a performance objective (PO)[2] at an appropriate step in the food chain.

8. Instead New Zealand set provisional food chain targets for both the poultry industry and the competent authority and monitored progress in reducing illness using a high quality surveillance system. This established a quantitative relationship between the industry target and the level of human illness achieved, thereby validating the target as risk-based. Establishing and validating this relationship facilitates continuous improvement in the reduction of a) the levels of *Campylobacter* present on broiler carcasses and b) the human cases of foodborne campylobacteriosis.

Food or food process; Point in the food chain where the MC is applied

9. The MC is applied to raw broiler chicken carcasses after slaughter, dressing and initial chilling (which is usually achieved using immersion chillers) to check the status at the end of primary processing.

Purpose (what is intended to be achieved)

10. The MC is used to verify whether a food safety control system applied to the slaughter and dressing of broiler chickens achieves a regulated performance target for *Campylobacter*.

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[1] Competent authority goal of 50% reduction in human foodborne cases over five years

[2] As defined in CAC/GL 63 – 2007: Principles and Guidelines for the Conduct of Microbiological Risk Management (MRM)

[3] As defined in the WTO SPS Agreement

Who should establish and who should apply

11. The MC should be established by the competent authority in consultation with the food business operators (broiler chicken processors) to ensure that it is practical and achievable.

12. The MC should be applied by the food business operators. Each operator decides which good hygienic practice-based and hazard-based measures to apply in order to meet the target. The Codex *Guidelines for the Control of Campylobacter and Salmonella in Chicken Meat*[4] provide valuable information to assist with these decisions.

Sampling plan (number of samples, sample size/units, sampling approach)

13. Two levels of processing by the food business operator have been recognized:

a) Processing operations which slaughter greater than 1 million broilers per year, must sample 3 whole broiler carcasses every processing day.

b) Processing operations which slaughter less than 1 million broilers per year, must sample 3 whole broiler carcasses on one randomly selected processing day per processing period (processing period is 5 days).

14. Each sample is taken by trained samplers at separate randomly selected times.

15. Each sample is a whole carcass selected from the processing line after initial chilling. Each carcass is handled and drained in a manner reflecting usual practices at the premises.

16. Carcasses are bagged and sent to the laboratory at refrigeration temperatures for rinsing, or rinsed at the processing premises and the rinsate is sent to the laboratory at refrigeration temperatures. In the latter case, (where the rinsate has been sent), the carcass can be returned to the processing line at the start of the chilling process.

17. The whole carcass is rinsed using 400ml of single strength buffered peptone water (ssBPW) in accordance with standard procedures which are defined in a technical schedule[5].

Organisms of concern

18. All 3 broiler chicken carcasses are analysed and enumerated for thermotolerant *Campylobacter* spp.

Method of analysis

19. Direct plate enumeration method using plating media (mCCDA) for each carcass rinsed with 400ml of single strength buffered peptone water confirmed by Oxidase/Latex. Standard methods are given in a technical schedule[5].

Interpretation of results

20. Results are evaluated by the competent authority for each food business operator, and at a national level (using aggregated results from all operators).

21. Each food business operator's results determines whether the operation is complying with the target. The national level analysis provides information back to each food business operator on how they are performing compared to the other operators.

22. In order to ensure that compliance is fairly assessed, the individual food business operator's results are interpreted by analysis of a "moving window". This takes into account the consistency of performance over time and does not penalize operators for one-off high results which may not give a true picture of performance. The moving window considers the results of the latest 3 processing periods. Each processing period is five processing days. The addition of the samples of the latest processing period displaces the results from the oldest processing period in the previous window.

23. Processing operations which slaughter greater than 1 million broilers per year are required to take 3 samples per processing day, resulting in 15 samples per processing period and 45 samples per moving window (which is 3 processing periods).

[4] CAC/GL 78-2011

[5] Contact the Ministry for Primary Industries at www.mpi.govt.nz for the technical schedule

24. A "moving window limit" failure occurs when the log count for 7 or more out of 45 samples from the moving window (three successive processing periods) are greater than the target of 3.78 $\log_{10}$ cfu/carcass. Figure 1 demonstrates how the moving window works.

25. Processing operations which slaughter less than 1 million broilers per year are required to take 3 samples from one day each processing period resulting in 9 samples per moving window. In this case, a "moving window limit" failure occurs when the log count for 2 or more out of 9 samples from the moving window (three successive processing periods) are greater than the target of 3.78 $\log_{10}$ cfu/carcass.

Figure 1: Example of a moving window for a processing operation

Sampling date	Campylobacter (log CFU/carcass)			Samples >3.78	No. > 3.78	Total Score (add 1 if No >3.78 is 7 or more)	Response Level	Processing period.day	Moving window
29-Mar-12	2.00	2.00	2.00	0				1.1	
30-Mar-12	3.34	3.70	3.30	0				1.2	
2-Apr-12	2.00	2.00	2.00	0				1.3	
3-Apr-12	3.15	2.78	2.00	0				1.4	
4-Apr-12	3.00	3.20	4.55	1				1.5	
5-Apr-12	2.60	4.15	4.68	2				2.1	
10-Apr-12	3.26	3.15	2.78	0				2.2	
11-Apr-12	3.20	2.00	2.00	0				2.3	
12-Apr-12	3.53	3.83	3.58	1				2.4	
13-Apr-12	2.60	3.64	2.90	0				2.5	
16-Apr-12	3.30	3.15	2.90	0				3.1	
17-Apr-12	2.00	2.00	2.00	0				3.2	
18-Apr-12	2.30	2.60	2.00	0				3.3	
19-Apr-12	2.00	2.30	2.00	0				3.4	
20-Apr-12	2.00	2.00	2.00	0	4	0	0	3.5	
23-Apr-12	2.78	3.66	4.44	1			☺	4.1	
24-Apr-12	2.30	2.30	2.00	0				4.2	
26-Apr-12	2.00	2.30	4.25	1				4.3	
27-Apr-12	2.00	2.00	2.00	0				4.4	
30-Apr-12	2.90	2.30	3.85	1	6	0	0	4.5	
1-May-12	2.30	2.78	2.78	0			☺	5.1	
2-May-12	3.26	4.09	3.86	2				5.2	
3-May-12	3.87	2.60	2.90	1				5.3	
4-May-12	2.00	2.30	3.41	0				5.4	
7-May-12	3.94	3.08	4.98	2	8	1	1	5.5	
8-May-12	3.70	3.48	4.08	1				6.1	
9-May-12	3.08	3.76	3.38	0			Review HACCP & GHP	6.2	
10-May-12	2.00	3.08	2.60	0				6.3	
11-May-12	2.60	2.60	2.60	0				6.4	
14-May-12	2.00	3.00	4.14	1	10	2	2	6.5	
15-May-12	6.03	3.86	3.98	3				7.1	
16-May-12	2.00	3.15	4.40	1			Review Processing Equipment	7.2	
17-May-12	2.00	2.00	2.00	0				7.3	
18-May-12	3.20	2.00	2.30	0				7.4	
21-May-12	2.00	2.00	2.00	0	11	3	2	7.5	
22-May-12	2.00	2.00	2.00	0				8.1	
23-May-12	2.00	2.00	2.00	0			Review Farm Biosecurity	8.2	
24-May-12	2.00	2.00	2.00	0				8.3	
25-May-12	4.72	4.10	2.00	2				8.4	
28-May-12	3.15	4.21	3.88	2	10	4	3	8.5	
29-May-12	2.00	3.15	2.00	0				9.1	
30-May-12	2.00	2.00	2.00	0			Further Sampling & Testing	9.2	
31-May-12	2.00	3.76	2.30	0				9.3	
1-Jun-12	2.00	2.00	2.00	0				9.4	
5-Jun-12	4.32	4.42	4.21	3	11	5	3	9.5	
6-Jun-12	2.00	2.00	2.00	0				10.1	
7-Jun-12	2.00	2.00	2.00	0			Implement Extra Interventions	10.2	
8-Jun-12	2.30	2.00	3.88	1				10.3	
11-Jun-12	2.00	2.00	2.00	0				10.4	
12-Jun-12	2.00	2.00	2.00	0	8	6	4	10.5	
13-Jun-12	2.00	2.00	2.00	0				11.1	
14-Jun-12	3.53	2.00	2.00	0			Competent Authority Visit	11.2	
15-Jun-12	2.00	2.00	2.00	0				11.3	
18-Jun-12	2.00	2.00	2.00	0				11.4	
19-Jun-12	3.79	3.45	2.00	1	5	0	0	11.5	
							☺		

(Nature of) actions in case of non-compliance

26. An escalating response to non-compliance is mandated with corrective action initially expected by the food business operator with a requirement to notify the competent authority's verifier of the action taken. Responses include:

- Review of HACCP and GHP
- Review processing equipment
- Review farm biosecurity
- Further sampling and testing
- Implementation of extra interventions

27. When responses by the food business operator have been unsuccessful (resulting in response level 4 (see Figure 1 for an example), the competent authority will lead a team of experts to check the food business operators actions taken to-date and to determine whether further corrective actions, additional interventions, and/or mandatory sanctions are necessary.

28. Competent authority sanctions may include a requirement to apply additional control measures such as freezing raw product, restrictions on processing or use of products, extra verification visits (at the food business operators cost) or ultimately the closure of the broiler chicken processing operation if the process is not brought back into compliance with the CPT within an agreed timeframe.

29. A food business operator must demonstrate that they can achieve consistent compliance after corrective actions have been implemented. When a processing operation once again meets the target for a full moving window, then the response level is reset to zero.

30. Further details on the corrective action required in the case of non-compliance are given in a technical schedule[5].

This example has not been peer-reviewed.

EXAMPLE 4: A PRACTICAL EXAMPLE OF THE ESTABLISHMENT OF A MICROBIOLOGICAL CRITERION FOR LOT-WISE VERIFICATION, BASED ON A QUANTITATIVE RISK ASSESSMENT

[This text is to be regarded only as an example and individual governments and industries should decide what use they wish to make of the approach described here]

Introduction:

1. The principle of the procedure is that a simplified, standardized quantitative risk assessment is performed for each lot that is subject to sampling and testing. The acceptable limit of the microbiological criterion is the level of consumer risk deemed acceptable.

Initially, the current risk of a certain food product/pathogen combination in a country is established by performing a Quantitative Microbiological Risk Assessment (QMRA). The outcome of this QMRA establishes the "baseline risk", as an expression of the expected risk (or mean risk) of a specific food product consumed in this country. Next, the risk of a new lot (or food lot) is calculated based on a number of samples from that lot, by applying the same QMRA model. The risk of this lot may then be calculated as a relative risk, i.e. the risk of the lot divided by the "baseline risk" estimate. Once the QMRA model has been established, the relative risk can be estimated by applying a simple mathematical equation (see below, "interpretation of results", for an example).*

A prerequisite for calculating the relative risk, and indeed to perform the pre-requisite QMRA, is that data regarding the presence of the actual organism is generated in (semi-)quantitative terms.

The outcome of the procedure is an estimation of the relative risk of a certain lot. In practice this means, that if the relative risk is e.g. 5, the lot tested is estimated to be 5 times more risky than the average, meaning that 5 times more people would be diseased by eating this lot as compared to eating a "baseline" lot.

As more and more lots are tested the new results may be implemented in the calculation of the "baseline risk" making the initial QMRA even more reliable and flexible reflecting the actual risk of a certain food/pathogen combination.

How high an increase in relative risk is acceptable? This is clearly a risk management decision. The risk managers could be national governments. It could in fact also be the food business manager, wanting to fulfill the requirements of a costumer, or to live up to an internal benchmark. According to the SPS agreement (Agreement on the Application of Sanitary and Phytosanitary Measures, WTO 2010) governments are permitted to maintain appropriate sanitary protection, and they are encouraged to do so in consistence with international standards. In reality this means that a government may establish control measures that will maintain the current level of health protection in its territory. The approach presented here is especially suited for that purpose: it compares the anticipated public health impact of a specific lot of foods to that of the average of the current lots (the baseline), enabling science-based decisions regarding acceptance or rejection of lots that are estimated to have a large negative impact on the level of protection. The assessment of the impact on the level of protection is based on international agreed principles for performing risk assessment.

Purpose:

2. The purpose of the use of the microbiological criteria *[in this example]* is to relate acceptance/rejection decisions to an Appropriate Level of Protection with specific reference to the occurrence of *Campylobacter* in broilers

To perform a QMRA it is necessary to have a substantial amount of quantitative data. The reliability of the risk estimate will increase with increasing amounts of data. Testing is expensive and demanding in labor. Therefore in order to get a reasonable amount of data with a reasonable amount of testing it makes sense to choose for this example a pathogen which is present in the selected food with a relatively high prevalence. For this reason Campylobacter in broilers are used for this example. However, when data are available, the approach may be used on other pathogen/food combinations.

Who could implement the criterion?

3. This microbiological criterion would likely be set by the competent authorities, but could in theory also be implemented by a food business operator.

As mentioned above a pre-established QMRA must be available in order to use this approach. Once established, this model can be made available, open-source, for anyone that chooses to use the approach. This would likely be competent authorities, but could actually also be a food business operator. All that would be needed would be an Excel spreadsheet with the model prepared by the risk assessors, in which the risk managers could fill in the results of the analysis, and read the calculated estimated relative risk of the lot tested.

Product :

4. Broiler carcasses, either chilled or frozen.

Organism:

5. *Campylobacter* spp.

Sampling plan:

6. 20 single samples of neck-skin or surface meat are sampled. At least 10 grams of neck-skin should be collected.

The number of samples to be collected ("20" is just an example) is a decision balanced between the degree of uncertainty and accuracy needed to fulfil the purpose of the risk estimate weighed with the resources available.

Method:

7. Each sample is homogenized and 10-fold dilutions are prepared and tested quantitatively for *Campylobacter* spp. according to ISO 10272-2: 2006 (Microbiology of food and animal feeding stuffs -- Horizontal method for detection and enumeration of Campylobacter spp. -- Part 2: Colony-count technique)

Interpretation of results:

8. The results of the analysis are grouped in:

$N_{<100}$: no of samples <100 cfu/g,

$N_{100\text{-}1000}$: no of samples >100 and <1000 cfu/g, and

$N_{>1000}$: no of samples >1000 cfu/g.

Note that with sample size n=20, $N_{<100} + N_{100\text{-}1000} + N_{>1000} = 20$

The relative risk (RR) of the lot is calculated by: $\mathbf{RR = a*N_{100\text{-}1000} + b*N_{>1000}}$

For an explanation of the technical background for the equation, please refer to the attached "Technical annex".

In the equation the parameters "a" and "b" relate to the relative risk of chicken meat that contained between 100 and 1000 cfu/g, and above 1000 cfu/g at the point of sampling. These must be determined by performing a quantitative risk assessment that combines an exposure assessment model from processed carcasses to consumption and a dose response model. A QMRA performed in Denmark (ref.: Rosenqvist et al., 2003) has estimated a" and "b" to be 0.271 and 0.988 respectively on the basis of a data from a retail meat survey in 2005 (the "baseline data") for which RR=1.

(As the sensitivity of the method is 100 cfu/g, results of <100 cfu/g are for calculation purposes set at "0". Based on the risk assessment these are considered not to contribute to increased risk and therefore they do not appear in the formula for calculation).

Limit of acceptance

9. Relative risk = 10

The precise value of the limit of acceptance is a risk management decision. The level could be any value, according to the situation. This has to be decided by the national competent authorities.

Action in case of non-compliance:

The actions proposed here only serves as examples. This is strictly a risk management decision.

10. Examples of corrective action: Option a) the lot is rejected for human consumption, and withdrawn.

Option b) the lot is subjected to heat treatment. Option c) The lot is subjected to decontamination. The acceptable effect of decontamination (the performance criterion) may be defined, i.e. a 2 log reduction. Option d) the producer of the lot is fined, without withdrawing the lot.

11. Example of a possible scenario:

From a lot of broilers 20 samples of neck skin is sampled, and analysed quantitatively for *Campylobacter* spp.

The resultof the analysis is that 8 of the samples show levels between 100 and 1000 cfu/g, and 5 samples show levels above 1000 cfu/g.

Based on the equation and using the estimates (for technical background see the attached "Technical annex").

Relative Risk = 8 x 0.271 + 5 x 0.988 = 7.108

The result show that the lot tested is estimated to be 7.1 times more risky than avarage lots. It is up to the risk managers to decide if this is acceptable. In case that the threshould of the estimated relative risk has been set at 10, this lot would be acceptable.

If the result of the analyses had been that 8 samples had levels between 100 and 1000 cfu/g, and another 8 samples had levels above 1000 cfu/g the resulting estimation of the relative risk would have been:

Relative Risk = 8 x 0.271 + 8 x 0.988 = 10.072

In this case the lot would not have met the microbiological criterion, and would have been subjected to corrective action.

12. Specific Requirements for application of this method

The establishment of a Microbiological Criterion by this approach has the advantage that it directly links food sample data from a lot of food to a relative risk estimate by a simple mathematical equation. The specific requirements to apply this approach in a country for a specific food – pathogen combination (as compared are to other MCs) are (1) the need of an appropriate QMRA model covering exposure assessment from the point of assessment to exposure combined with a dose response model, to estimate the risk attending the sampled product and (2) quantitative "baseline data" representative for the current contamination levels of the food product at the point of sampling.

Ad. (1): This implies that the QMRA need not cover the whole farm-to-fork food chain, but only the last part before exposure. For Campylobacter in broiler meat several of such QMRA models have been compared by Nauta and Christensen (2011). Although the QMRA models may need to be made country specific, it is a feasible option to use models that are currently available. The error that is made by using a generic QMRA model is smaller for the assessment of relative risks than for absolute risks.

Ad (2): The establishment of a "baseline risk" demands a representative data set, the "baseline data". Whether a data set is considered "representative" is again a risk manager's decision, depending on the situation. It may be relevant that the baseline includes samples from all seasons, from different parts of the country, imports and domestic production, frozen and fresh products, etc. It may also be possible to use a limited baseline or the baseline of another country, and assess the relative risk compared to a "surrogate baseline". . In reality when applying microbiological criterion the proportion of lots that are rejected has to be in a manageble order. The objective is to remove the most risky lots from the market, not to remove risk enterely. Therefore the approach could be used for this purpose, by setting the acceptable relative risk at a level where only a reasonable (practical and manageble) proportion of lots will be removed from the market. Removing the most risky lots from the market would provide impetus for improvement in industries involved, and would tend to reward producers with low contamination levels. In principle this could result in decrease of the prevalence over time, enabling continuation of improvement in relation to public health.

13. Definition:

**Relative risk (abbreviated definition according to the National Cancer Institute, USA): measure of the risk of a certain event happening in one group compared to the risk of the same event happening in another group.*

14. References

Nauta,M, Christensen,B.B, 2011. The impact of consumer phase models in microbial risk analysis. Risk Analysis 31, 255-265.

Rosenquist,H, Nielsen,N.L, Sommer,H.M, Nørrung,B, Christensen,B.B, 2003. Quantitative risk assessment of human campylobacteriosis associated with thermophilic Campylobacter species in chickens. International Journal of Food Microbiology 83, 87-103.

EXAMPLE 4, Technical annex:

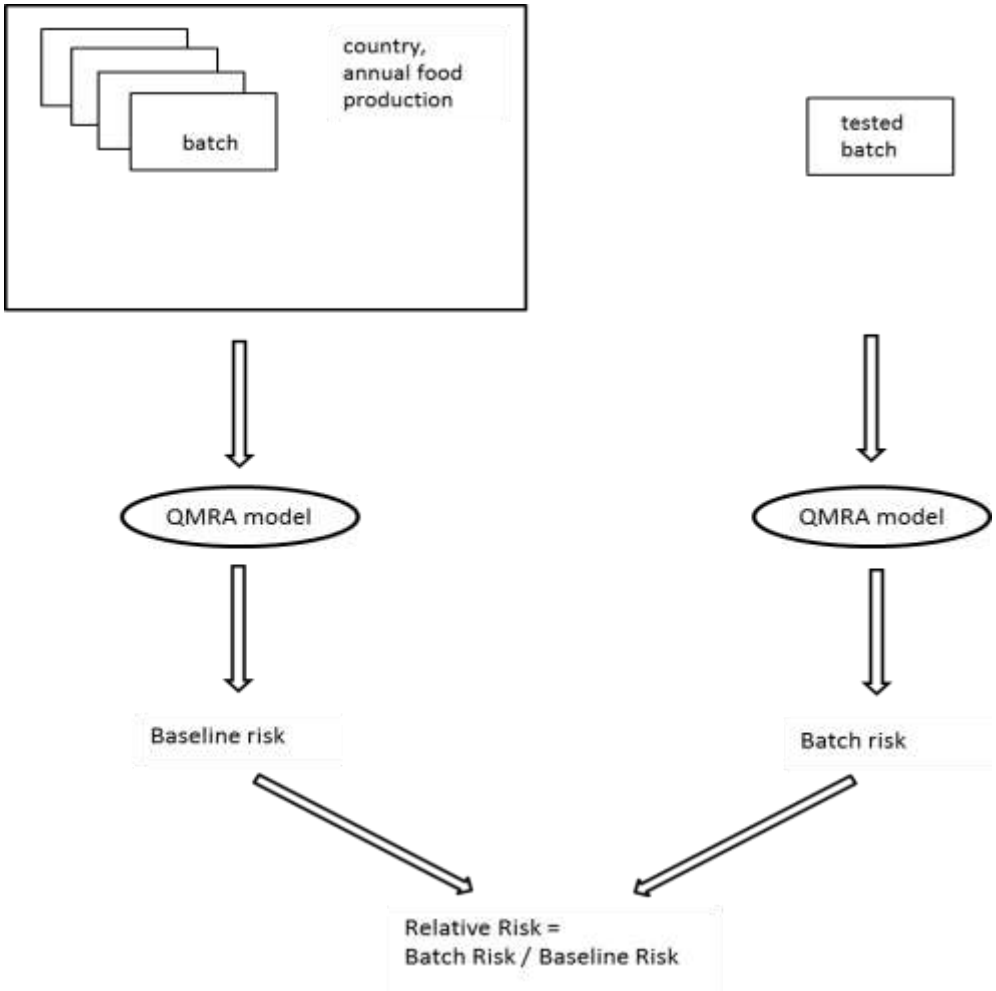
A PRACTICAL EXAMPLE OF THE ESTABLISHMENT OF A MICROBIOLOGICAL CRITERION FOR BATCH-WISE VERIFICATION, BASED ON A QUANTITATIVE RISK ASSESSMENT ;

General approach

1) A QMRA (quantitative microbiological risk assessment) model, that can assess the public health risk attending a sampled food product, is used to assess the baseline risk, which is the expected current risk (or mean risk) of a specific food product consumed in a country. This baseline risk can for example be based on monitoring data.

2) The same model is used to assess the risk of a new tested batch. A relative risk is estimated by dividing the batch risk by the baseline risk.

3) If risk managers decide that this relative risk is too high, corrective action can be taken.



The QMRA model used in the example is the last part of the QMRA model of Rosenquist et al (2003), that covers the food chain from the broiler meat produced at the end of industrial processing to consumption, and the dose response model. This model is described in the technical annex.

Once a Batch risk is calculated with the QMRA model, a simple formula can be derived that facilitates the calculation of the Relative risk for a tested batch that is of which *n* samples are taken.

$$\mathbf{RR = a*N_{100\text{-}1000} + b*N_{>1000}}$$

This allows to quickly assess the relative risk as soon as the sampling results are available.

It is explained below how the parameters a and b are obtained in an example that that resembles the approach applied for *Campylobacter* in broiler meat in Denmark

Mathematical description

The model used for the risk assessment considers the probability of contamination and the level of contamination. The ingested dose is described as a function of the concentration per gram of fresh broiler meat, $C_{broiler}$, sampled at retail. The model considers two routes of transfer from the fresh meat to the meals: one where the meat is re-contaminated, after cooking, via the environment (contact with hands, cutting board or knife) and another where a side dish (like a salad) is contaminated via this environment. The probability of transfer to consumed meat per cfu on the fresh meat equals: $p_{BB} = t_{BE} \times t_{EB,}$ where t_{BE} is the transfer rate from broiler meat to environment and t_{EB} the transfer rate back from environment to the broiler meat dish. Similarly, the probability of transfer per cfu from fresh broiler meat to salad (side dish) equals: $p_{BS} = t_{BE} \times t_{ES}$, where t_{BE} is the transfer rate from fresh broiler meat to environment and t_{ES} the transfer rate from environment to salad.

It is assumed that the frequencies of cross contamination via each of these routes are equal, so these frequencies cancel out when calculating the final output; the relative risk. Based on the results of Zhao et al. (1998), all three transfer rates *t* were sampled from a BetaPert distribution with minimum 1, mode 2 and maximum 6, such that $-\log(t) \sim BP(1,2,6)$.

The number of *Campylobacter* bacteria ingested via the meat N_{meat} was

(1) $N_{meat} \sim Poisson[C_{broiler} \times \varphi \times w_{meat} \times p_{BB}]$

with w_{meat} the portion size of the meat (in g) sampled from the distribution: LogNormal(189, 126.9).

Similarly, the number of *Campylobacter* bacteria ingested via the salad N_{salad} was then

(2) $N_{salad} \sim Poisson[C_{broiler} \times \varphi \times w_{salad} \times p_{BS}]$

with w_{salad} the portion size of the salad (in g) sampled from the distribution: LogNormal(91.4,65.6).

The portion weights included in eq. 1 and 2 are based on consumption data in Denmark for men in the age class 30-65 (Andersen et al., 1996; Christensen et al., 2001), and as the retail data are obtained per gram of meat surface and the consumption data provide grams of consumed meat, a transition factor was included as $\varphi = 0.2$. This value is based on the observation that the skin of the broiler constitutes about one tenth of the weight of the whole carcass. To convert from cfu *Campylobacter* per gram carcass to cfu per gram eatable chicken, this value was further multiplied by 2 because only half the carcass mass is considered edible.

The ingested dose then equals

(3) $d = N_{meat} + N_{salad}$

As dose response model, we used the same model as applied in the Danish *Campylobacter* risk assessment (Rosenquist et al., 2003), which is based on the data obtained from Black et al. (1988) (Teunis and Havelaar, 2000), and used in many *Campylobacter* risk assessments (Nauta et al., 2009).

The probability of illness after exposure to (discrete) dose *d* equals

(4) $$P_{ill}(d) = 0.33 \times \left(1 - \frac{\Gamma(\alpha+\beta)\Gamma(\beta+d)}{\Gamma(\beta)\Gamma(\alpha+\beta+d)}\right)$$

where $\alpha = 0.145$ and $\beta = 7.59$ and $\Gamma()$ represent the Gamma function.

With this risk assessment model, the mean probability of illness from fresh broiler meat with any concentration $C_{broiler}$ can be assessed by Monte Carlo simulation. Similarly, the probability of illness can be estimated for each of the 20 samples from a batch and their mean is interpreted as the mean risk of the batch.

The risk model to assess the relative risk for a batch of meat, based on twenty samples taken from that batch, has been simplified to an equation that allowed a quick risk estimation:

(5) $RR = P_{ill}\ (sample) / P_{ill}\ (baseline) = \Sigma\ \beta_c\ N_c$

where c is the "class", the interval of enumeration results (<1, 1-10, 10-100, 100-1000 and > 1000 cfu/g); N_c is the number of samples that is found in that class; and β_c is a vector of coefficients. The values for these β_c are derived by using the baseline data, for which by definition RR=1. The parameter values are given in table 1.

The risk of fresh broiler meat with a concentration $C_{broiler}$ within class c (r_c) was assessed by Monte Carlo simulation. Log($C_{broiler}$) values were sampled from Uniform distributions with concentration class boundary values as the lower and upper limit, and using the risk assessment model outlined above. For the >1000 cfu/g class the upper limit of the Uniform distribution was set at 10,000 cfu/g.

If x_c is the relative frequency of finding a sample in class *c* among the baseline data, $E(N_c) = 20\ x_c$.

The values for the parameters β_c for which the sum over all classes $\Sigma_c\ E(N_c)\beta_c = 1$, or $\Sigma_c\ x_c\beta_c = 1/20$, can be obtained by defining

$S = \Sigma_c\ x_c r_{c,} = 0.00523$,

so for each class *c* it follows

$\beta_c = r_c / (20\ S)$.

With the obtained values and realizing that RR>>1 for a batch to be deemed unsafe, it shows that the first three classes can be neglected for the calculation of the relative risk of a batch. Therefore the relative risk calculation simplifies to

(6) $RR = 0.271\ N_{100\text{-}1000} + 0.988\ N_{>1000}$.

with $N_{100\text{-}1000}$ being the number of samples with a concentration between 100 and 1000 cfu/g and $N_{>1000}$ the number of samples with more than 1000 cfu/g, where the β_c originate from table 1. This simplification allowed the detection limit in the microbiological analysis to be 100 cfu/g.

Table 1. Parameter values used to derive the equation for RR (6). The relative frequency of concentrations in the baseline, x_C, is used to estimate the expected number of samples in each of the concentration classes c, $E(N_c)$ The mean risk per class, r_c, is obtained by Monte Carlo simulation of the risk assessment model after 500.000 iterations. Coefficient β_c is the estimated single sample attribution to the relative risk of the batch, for each class c.

Log cfu/g (interval)	class *c*	<0	0..1	1..2	2..3	>3
Rel.frequency baseline	x_c	73.20%	9.44%	9.14%	5.00%	3.22%
Expected frequency with n=20 samples	$E(N_c)$	14.64	1.89	1.83	1.00	0.64
Mean risk	r_c	0	0.00052	0.0047	0.0284	0.1033
Coefficient	β_c	0.000	0.005	0.045	0.271	0.988

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EXAMPLE 5A: BASE DOCUMENT: OPERATIONALISING A PERFORMANCE OBJECTIVE WITH A MICROBIOLOGICAL CRITERION FOR A RISK-BASED APPROACH

Disclaimer: Example 5A has been compiled by a working group formed by Canada, France, India, Brazil and ICMSF and has not been peer reviewed

Purpose:

1. A performance objective (PO) is a risk-based metric that allows government risk managers and food business operators to specify quantitatively the required stringency of a food safety management system at a particular point in a food supply chain. This target should be achieved by the responsible food business operator at that particular point, taking into consideration the other control measures used in the food safety management system. To establish whether the target is met, establishment of a microbiological criterion (MC) is one way to "operationalise" the PO.

Who should establish a PO/MC and who should apply it?

2. A PO can be derived from a health target (e.g., ALOP) or a food safety objective (FSO) developed by a competent authority, or it can be established on the basis of a quantitative risk assessment developed for the relevant pathogen in a particular food for/by a competent authority. Food business operators can establish a PO on the basis of either an FSO set by a competent authority, or an evaluation (usually quantitative) of a hazard in the food supply chain for which they are responsible.

3. When a competent authority establishes a PO at a particular point in the food supply chain for the purpose of providing regulatory guidance to the relevant food industry, the authority may choose to establish an MC that corresponds to that specific PO. Where a competent authority establishes an MC, based on a PO, as a regulatory standard, the MC can be used to assess whether the food business operator is meeting the standard.

4. Likewise, if a PO is established by a food business operator or by multiple operators as part of their management of a food supply chain, an MC can be established by the food business operator(s) to verify that the PO is being met consistently.

5. In both cases, it is the industry or food business operator that takes action to achieve the MC, i.e., designs a food safety management system that will consistently meet the MC.

Point in the food chain where the MC is applied:

6. Since a PO can be established at any point in a food supply chain (other than at the point of consumption), such as raw materials, ingredients, partial and final products within primary production, manufacture, distribution, products on the market and in foodservice operations, an MC established based on a PO relates to the corresponding point in the food supply chain.

Establishment and implementation of an MC in relation to a FSO/PO

7. **Assumptions/decisions to be made for the establishment of an MC**:

   i. Firstly, an assumption must be made regarding the distribution of the pathogens in the lot of food. Knowing the actual distribution within a lot can be very beneficial in establishing a suitable MC and should be used where available. In the absence of available data, a log-normal distribution is often assumed and a default value for the standard deviation applied. In such cases, generally, variability in concentration levels within a lot can be described as having a standard deviation of 0.2 $\log_{10}$ cfu g^{-1} for foods with a "homogenous" distribution of microbes (e.g., liquids with a degree of mixing), 0.4 $\log_{10}$ cfu g^{-1} for foods with "intermediate homogeneity" (e.g., ground semi-solids) and 0.8 $\log_{10}$ cfu g^{-1} for foods that are not homogenous (e.g., solid foods). It could be that in certain cases even greater non-homogeneity could occur, e.g., if clumping occurs or if the contamination is restricted to surface contamination of a food.

   ii. The second requirement is to define the ''maximum frequency and/or concentration" of the hazard that will be used to specify the FSO/PO, including what proportion (e.g., 95%, 99%, 99.9%, etc.) of the distribution of possible concentrations must satisfy the test limit, so that the FSO/PO is met. Alternatively, the assumption may be made whether it is acceptable for a part of the frequency distribution represented in an MC to be exceeded and by how much, e.g., whether, for example, it is acceptable that 1 or 5% of the sample units exceed the regulatory limit.

iii. In establishing an MC for regulatory purposes, it is up to the competent authorities to decide the acceptable proportion of the distribution that should either meet or exceed the regulatory limit, based on the public health outcome to be achieved.

iv. The third decision is to specify the level of confidence needed that a non- conforming lot is detected and rejected (e.g., with 95% or 99% confidence). Alternatively, the probability of rejecting a conforming lot may be considered.

v. The fourth decision is the analytical methodology that should be used.

Sampling plan:

8. The sampling plan appropriate to assess an MC depends on the specific situation for which the PO is established. Notably, a PO is a maximum frequency and/or concentration of a hazard. Therefore, a PO can be set as:

1. A frequency (prevalence) limit (independent of concentration of the hazard),
2. A concentration limit (independent of frequency), or
3. A limit for concentration and frequency combined.

9. In reality, however, the prevalence and concentration are not independent. The higher the concentration of the contaminant in a batch, the higher will be the expected prevalence in a sample drawn from that lot. At very low concentrations few sub-samples would be expected to contain a contaminant, i.e., the observed prevalence would also be low.

10. At higher concentrations, such that the concentration per sample is much greater than "1 cell per sample", the prevalence will become 100% or '1' in terms of probability. However, in practice, the concentration of contaminants will not be completely homogenous throughout the batch being tested but will vary. In this case, the chances of detecting a contaminant will depend on the average concentration, the sample size and the variability in the contamination level.

11. Because of the generally heterogeneous nature of the distribution of contamination levels, even if the average concentration is below the PO, some samples will test "positive", i.e., no matter what the criterion or sampling plan is, there is always a definable probability that some units in the sampled batch will exceed the concentration specified. When the average concentration is closer to the acceptable limit, or when the variability in the contaminant levels is higher, more samples in the batch will test "positive", even if the average level is below the MC.

12. An MC is a practical tool to verify whether a lot can be accepted or rejected based on microbial analysis. The MC tests, with a certain level of confidence, whether or not a lot meets the criterion for acceptability. To define these criteria, one has to understand the likely distribution of the target pathogen in the lot and have a metric to define acceptable and non-acceptable lots (which in this case would be the PO).

13. In any case, the lower the proportion of units that can be tolerated to exceed the limit, the more stringent the sampling plan needs to be.

14. The stringency of a sampling plan is defined by the parameters *n*, *c*, *m* and *M*. For more stringent sampling plans, more samples (*n*) can be taken with fewer being allowed to test 'positive' (*c*), and/or the limits *m* and *M* must be lower (if quantitative) or the sample unit larger (if using presence/absence tests). When the distribution of target organisms in the lot is more variable (at equal amount of total organisms, so more clustered), a more stringent plan is usually required to have confidence that the food business operator is consistently complying with the MC, i.e., the confidence one has that a food business operator is complying with the MC, depends on the sampling plan, the confidence limit set, and the variability of the distribution of the hazard in the food lot.

15. The size of the analytical unit (portion in g or ml of the sample analyzed), the standard deviation of the distribution of the hazard within the food, the probability of acceptance and the statistical definition of the PO, are other factors that determine the practicality of application of an MC for confirmation purposes.

Organism(s) of concern:

16. Within an overall food safety management system, a PO can be established to control any pathogen at an identified point in the food chain. Generally, the MC should be established for the pathogen for which the

PO is established. Within the scope of the present document, establishing an MC for indicator organisms or for indicators of general hygiene of the food or food environment should only be considered if a clear correlation with pathogens can be effectively established.

Method(s) of analysis:

17. The appropriate analytical method (absence/presence, MPN or viable/colony counting) used to assess compliance with the MC will depend on the type of limit specified and the organism. For many foodborne pathogens, particularly those causing illness by infection, 'presence/absence' test methods are often specified, because they generally have a lower limit of detection than direct plating methods and thus may increase confidence that even if a pathogen is present at low levels, it will be detected.

Interpretation of results:

18. An MC used to assess the conformity with the required performance of the food safety management system (i.e., one that consistently satisfies the PO) is typically defined by:

- a specified maximum proportion of units in the lot exceeding a specified level, e.g., "*c*" the number samples out of "*n*" samples taken that may exceed the microbiological limit "*m*" (for 2-class plans) and, in the case of "3-class" plans, a microbiological limit "*M*" that no analytical units may exceed.

19. When an MC specifies a 3-class plan, a specific maximum frequency of exceeding "*m*" alerts the food business operator to a possible loss of control of the performance of their food safety management system, and that action needs to be taken to re-establish and maintain ongoing control.

Actions in case of non-compliance:

20. The typical actions that are taken when there is a failure to meet an MC include: i) preventing the affected lot from being released for human consumption; ii) recalling the product if it has been released for human consumption, and/or iii) determining and correcting the root cause of the failure.

Other practical aspects:

21. When the practicality of testing and the interpretations of the results are considered, the situation regarding fresh or raw foods may be different from foods processed for safety.

22. In the case of such processed foods pathogens should, in principle, not be present or their presence (due to survival or unavoidable recontamination and growth) should be at levels that present a negligible risk to public health. Unacceptable levels of recontamination should not occur, or should be detected, and such incriminated batches should not be released for sale. Microbiological testing should be used to detect such lots when no other means are available. However, while the distribution of the pathogens in the lot is not known, it is most likely that they are not homogeneously distributed throughout a consignment. Moreover, random sampling is often not possible for reasons of accessibility of units in consignments on trucks, ships, etc. Consequently, in these cases, the calculations and interpretations of pathogen testing data have only limited validity: in simple terms it can be argued that a positive finding (i.e., presence of a pathogen) means something, while a negative one means very little. Even when the necessary data are available to allow statistical interpretation of the test results, the number of samples needed to obtain a meaningful result may be too large to be practical.

23. The situation may be different for foods that are not processed for safety, that are raw or that may originate from polluted environments. In these situations, testing may be useful because contamination levels and/or frequencies would be expected to be higher.

Illustrative practical examples:

24. The three case scenarios below follow the general approach described in the basic document to illustrate operationalisation of a PO with an MC in different situations.

Responsibilities for setting metrics:

25. Table 5A-1 provides an overview of the various metrics and notes responsibilities and conditions for setting individual metrics.

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EXAMPLE 5A, SCENARIO 1: Deriving an MC from a PO That is set as a Numerical Limit to the Concentration of a Pathogen

1. While the PO is a risk-based metric related to the acceptability of batches/lots in the context of established FSO and/or ALOP values, the MC is a practical tool to verify whether individual batches/lots meet the PO by taking samples from such batches/lots for microbiological analysis. Key parameters such as the microbiological limit (*m*) and the number of samples found to be meeting this microbiological limit, need to be established in the process of deriving an MC from a PO. For the purpose of this scenario, it is assumed that a Competent Authority has established a PO for the concentration of a specific pathogen in a certain commodity. The role of the PO for lot acceptability and the *m* for sample acceptability in such a case are illustrated in Figure 5A-1.1.

2. The following PO for the concentration of a specific pathogen (for example *Listeria monocytogenes*) in a certain commodity (for example a ready-to-eat food product not supporting growth) has been set:

- PO such that P(logC>4 log cfu/g) ≤ 0.5% (equivalent to PO such that logC is ≤4 log cfu/g for 99.5% of the product units).
- This means that less than or equal to 0.5% of the products in the batch/lot is allowed to have a level of microorganisms greater than 4 log cfu/g.
- In other words, the PO is set to give a very high probability, namely 99.5%, that the pathogen level of each product in the batch/lot is below 4 log cfu/g.

3. The competent authority also defines a suitable Microbiological Criterion that can be used to verify that an individual batch/lot complies with this PO. The competent authority responsible for deriving a suitable MC from the PO will have to make several (risk management) decisions regarding the sampling plan for the MC, such as choosing the values for *m*, *n*, and the confidence limit.

4. Firstly, the competent authority establishes or decides on the microbiological characteristics of a typical batch/lot that just complies with the PO (step 1 + 2; see Figure 5A 1.2). Data/knowledge available to competent authorities for determining the microbiological characteristics of typical batches can come from sources such as surveillance studies, the scientific literature or industry consultation.

5. Secondly, a suitable sampling plan for the MC is designed. This scenario focuses on establishing values for the microbiological limit (*m*) (step 3 in Figure 5A 1.2) and the number of samples (*n*) that would be needed to detect/reject non-compliant batches/lots with a certain confidence (steps 4-6 in Figure 5A 1.2), using microbiological sampling and analysis.

6. The following step-wise process details the individual steps in a worked scenario. Notably, suitable values for the sampling plan parameters and the confidence limit other than the ones developed here, may be equally or more suitable for a given situation.

Step-wise process:

1. Establish/Decide on the concentration distribution in the batch/lot and the standard deviation of the pathogen concentration

7. The distribution of pathogen concentrations in typical batches/lots of a certain commodity and the associated standard deviation ideally are derived from observation and this data/knowledge is accumulated over time by food business operators and governmental organizations such as public health institutes and food inspection services. Where such specific data/knowledge does not exist, it is often a good choice to assume a lognormal distribution of concentrations, with a standard deviation of 0.8 log cfu/g as a default.

8. In this scenario, it is indeed assumed that the distribution of concentrations of the pathogen over all units in a lot/batch is lognormal, and that the standard deviation of concentrations is 0.8 log cfu/g. Other distributions and standard deviations may apply based on specific data/knowledge.

2. Calculate a mean log concentration so that the distribution with this mean log concentration and standard deviation complies with the PO

9. Based on a lognormal distribution of concentrations (this means that the logarithm of the concentrations are normally distributed) and a standard deviation of 0.8 log cfu/g, the competent authority then determines the mean log concentration of the relevant pathogen in a batch/lot that is just complying with the *PO* value ($\log C \geq 4$ log cfu/g)$\leq 0.5\%$. A mean log concentration of 1.94 log cfu/g (1.93933655) would comply, as calculated using the following formula:

$$P_{\text{normal,cumulative}}(\text{PO}; \mu, s)=0.995$$

if $P_{\text{normal,cumulative}}(4; \mu, 0.8)=0.995$

then $\mu=1.94$

$$P_{\text{normal,cumulative}}(4;1.94,0.8)=0.995$$

[This can also be verified in Excel by typing in the formula =NORMDIST(A1;A2;A3;TRUE) with values 4;1.9393365;0.8 typed respectively in the cells A1, A2 and A3. The probability will be displayed as 0.995, meaning that a lognormal distribution with a mean log concentration of 1.94 log cfu/g and a standard deviation of 0.8 log cfu/g has 0.995=99.5% of the units below a level of 4 logs, and thus 0.5% of the units above a level of 4 logs. One can use the 'Solver" function by changing values for 'mean' (cell A2), when starting with an unknown value for 'mean' for a known probability (target cell value equal to 0.995)]

3. Decide on the microbiological limit* m *for the sampling plan of the MC

10. The competent authority then chooses a microbiological limit (*m*) for microbiological analysis of samples such that a practical and feasible sampling plan is established. The choice of *m* is made taking into consideration the microbiological characteristics of the product batch/lot concerned, the mean-log concentration of just compliant batches/lots, the analytical method used to detect and quantify the target micro-organism, etc.

11. In this scenario, the value for *m* is chosen to be 2 log cfu/g (*i.e.*, 100 cfu/g), considering that lower or higher values would be either not practical because of constraints regarding microbiological analysis (such as the need to analyze a very large number of samples), or otherwise not suitable to verify the PO (such as the sensitivity and accuracy of the analytical methods available). Note the paragraph with comments on the choice of the value for *m* below.

12. Please note that the value of *m* (2 log cfu/g) chosen in this scenario is only by chance close to the value established for the mean log concentration of the pathogen for a just compliant batch/lot.

***4. Calculate what the probability is for 'n' samples, to be able to detect a just compliant lot, given its mean log concentration and standard deviation, and the value chosen for* m**

13. Given the microbiological characteristics of a batch/lot that just complies with the PO (a log normal distribution of concentrations and a standard deviation of 0.8 log cfu/g as well as a mean concentration of 1.94 log cfu/g) and m = 2 log cfu/g, the probability of 'n' samples being negative can be established as follows:

14. When using a sampling plan with one sample (n=1), the probability of accepting (P_{accept}) such a batch/lot can be calculated as follows:

$P_{accept} = P_{normal,cumulative}(m; \mu, s)$

$P_{accept} = P_{normal,\ cumulative}(2;1.94,0.8) = 0.53$ (or 53%)

15. Which is the probability of a sample being below m=2 log cfu/g.

16. The corresponding probability of rejection ($\underline{P_{reject}}$) can be obtained by subtracting the value obtained for the probability of acceptance from the value 1 (100% probability).

17. For n=1 thus P_{accept} =0.53, $\underline{P_{reject}}$ = 0.47 or 47% probability ((1-0.53)*100)

[The scenario can also be calculated as =NORMDIST(2;1.9393365;0.8;TRUE)=0.530, meaning that one sample has only a 53% probability of being below 100 cfu/g. Therefore, there is a 47% probability that it is above 100 cfu/g, and thus represents a defective sample.]

17 bis. The values for P_{accept} with n samples can be calculated by taking 0.53 to the power n (since all of the n samples have to be below 100 cfu/g) and converting to % probability. The corresponding $\underline{P_{reject}}$ values can be obtained from the P_{accept} values. The following values are obtained for n = 2 to 8:

n	*P*accept	*P*reject
1	53%	47%
2	28%	72%
3	15%	85%
4	7.9%	92.1%
5	4.2%	95.8%
6	2.2%	97.8%
7	1.2%	98.8%
8	0.62%	99.38%

The overall formula to calculate these values is:

$P_{accept} = [P_{normal}(2,1.94,0.8)]^n$

$P_{reject} = 1-[P_{normal}(2,1.94,0.8)]^n$

5. Decide on the probability with which a non-compliant lot should be rejected

18. The competent authority then needs to choose a level of confidence with which non-compliant batches/lots will be rejected using the MC. Different confidence levels can be chosen depending on the severity of the pathogen, insight into the distribution of concentration and the standard deviation of the target microorganism across batches/lots, the business impact of a possible high rejection rate of compliant batches/lots, etc.

19. In this scenario, it is assumed that a sampling plan that detects/rejects such a non-compliant lot with greater than 95% probability is deemed appropriate.

***6. From this follows how many samples would need to be taken to achieve the selected probability of rejection, given the mean log concentration and standard deviation of the lot/batch and* m**

20. Considering the values for the probability of rejection (P_{reject}) for n = 2 to 8 shown above, for P_{reject} to be over 95%, at least 5 samples would need to be taken (P_{reject} for n=5 is 95.8%, thus >95%).

Result of the step-wise process to establish an MC from the PO.

21. To verify compliance of a batch/lot having the microbiological characteristics assumed here (i.e., log normal distribution; standard deviation of 0.8 log cfu/g) with the established PO (99.5% below 4 log cfu/g) with more than 95% confidence, a suitable MC would have an m value of 2 log cfu/g (i.e., 100 cfu/g) and an n value of 5 as part of the sampling plan.

22. Please note that several other aspects of an MC and the underlying sampling plan need to be additionally defined, as explained in the main text of the Codex guideline on establishing MCs.

Practical example for reference:

23. For example, the Codex sampling plan for *Listeria monocytogenes* for a product not supporting growth is n=5, c=0 and m=2 log cfu/g (CAC/GL 61 – 2007).

Analysis	Standard/Guideline				Assessment	
	n	*C*	*m*	*M*	**Satisfactory**	**Unsatisfactory**
L. monocytogenes	5	0	100	-	<*m*/g	>m/g in any of the *n* sub sample units tested

[Note 1: If one, for example, wants to detect such a lot with more than 99% probability, 8 samples would need to be taken (P_{reject}=99.4%>99%).

Note 2: The *n* for P_{reject} = 0.95 (that is P_{accept} = 0.05) in this case can also be determined as follows:

For *n*=1, P_{accept} = 0.53 as calculated above.

The probability of acceptance with a sampling plan with *n* samples, can be calculated by taking 0.53 to the power *n*, since all *n* samples have to be below 100 cfu/g.

Therefore, $P_{accept} = 0.53^n$

Substituting P_{accept} = 0.05 in the above equation: $0.05 = 0.53^n$

Therefore, log 0.05 = *n* log 0.53

That is, *n*= log 0.05/log 0.53 = 4.7 (rounded value for *n* = 5)]

Comments on the choice of *m*:

24. Regarding the choice of *m*, the following *m* and *n* values would give alternative designs of the sampling plan that can detect/reject non-compliant lots with the same confidence:

m		*n*
(cfu/g)	(log cfu/g)	
4	0.60	1
19	1.28	2
100	2	5
500	2.7	16
1000	3	31
1738	3.24	57

The *m* values of 0.60 or 1.28 log cfu/g would be constrained by the method for microbiological enumeration (e.g., by sensitivity, accuracy, standard deviation), while *m* values of, e.g., 2.7 log cfu/g and higher, would require a very large number of samples to be analyzed.

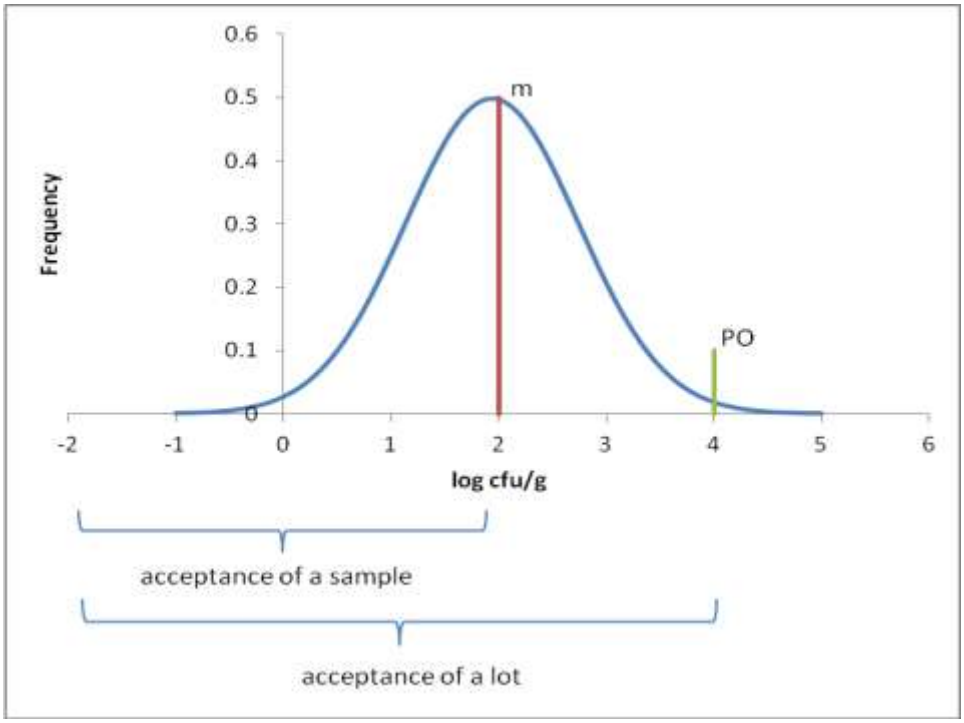


Figure 5A-1.1 The role of the PO for lot acceptability and the *m* for sample acceptability. In this graph, it can be seen that the probability that the concentration in a sample unit is below the *m* value is 53%, while the probability that the concentration is below the PO value is 99.5%, so 99.5% complies with the limit of 4 log cfu/g as set in the PO.

Example 1: Deriving an MC from a PO that is set as an actual limit to the number of a microorganism
(Figure 5A-1.2)

PO established at a specific point in the value chain
(i.e., pathogen level of ≤ 4 log cfu/g for 99.5% of the products in the batch)

1 **Establish/decide on the concentration distribution in batch/lot and the standard deviation of the pathogen concentration**
(*i.e.*, lognormal; s.d. 0.8 log cfu/g)

2 **Calculate a mean log concentration of the pathogen such that this batch/lot just complies with the PO**
(*i.e.*, 1.94 log cfu/g)

3 **Decide on the microbiological limit *m* for the sampling plan of the MC**
(i.e., *m* = 2 log cfu/g)

4 **Calculate what the probability is for '*n*' samples, to be able to detect a just compliant lot**
(*i.e.*, *n* = 1, 53% up to *n*= 8, 0.6%)

5 **Decide on the probability with which a non-compliant batch/lot should be rejected**
(*i.e.*, > 95% confidence)

6 **From this follows how many samples would be needed to achieve the selected probability of rejection**
(*i.e.*, *n* = 5)

MC: suitable sampling plan parameters *m* and *n*
(i.e., *m* = 2 log cfu/g and *n* = 5)

EXAMPLE 5A SCENARIO 2: Deriving an MC from a PO that is set as the limit to the prevalence or proportion of a microorganism

25. In this scenario, it is assumed that a PO has been established for a specific pathogen in a certain commodity at a certain point in the value chain that uses bacterial prevalence (i.e., presence/absence of the target microorganism in a certain quantity of batches/lots).

26. As an illustrative scenario, contaminated carcasses are the target commodity and the PO has been based on the prevalence of the pathogen, with the microbiological analysis being enrichment and testing 10-g samples after chilling.

27. The PO is defined such that ≤10% of the carcasses in a batch/lot or in a moving window approach are tolerated to be positive with the test. In other words, the pathogen may be present in ≤10% of the 10-g samples, but in the remainder the pathogen should not be present.

28. The following step-wise process details the individual steps in a worked example. The process is depicted in Figure 5A-2.1.

Step-wise process:

1. Calculate what the probability is for 'n' samples to be negative given the PO

29. The probability of having a negative sample in a batch/lot with exactly a 10% contamination rate (i.e., target fraction contaminated = 0.1) is the value 1 minus the target prevalence: 0.9.

30. For batches/lots with exactly a 10% contamination rate, the probability for '*n*' samples to be negative (*P*negative) would be 0.9 to the power *n*. The resulting probabilities are shown below for selected values of *n*.

n	*P*negative= *P*accept	*P*reject
1	0.90	0.10
5	0.59	0.41
10	0.35	0.65
15	0.21	0.79
20	0.12	0.88
25	0.072	0.928
26	0.065	0.935
27	0.058	0.942
28	0.052	0.948
29	0.047	0.953
30	0.042	0.958

2. Decide on the probability that a lot with this prevalence should be rejected

31. A decision has to be made on the level of confidence with which non-complaint batches/lots will be rejected using the MC. Different confidence levels can be chosen considering the severity of the pathogen, typical concentrations occurring in the target commodity, constraints of the microbiological test chosen, the business impact of a possible high rejection rate of compliant batches/lots, etc.

32. In this scenario, it is assumed that a sampling plan that detects/rejects batches/lots of carcasses with a contamination rate >10% with greater than 95% probability is deemed appropriate.

3. From this follows the number of samples that would need to be taken to achieve the selected probability of rejection

33. From the calculation above, it can be seen that all 29 samples need to be negative (29=0.9^29=0.047) to achieve the chosen probability (>95%) that the sampling plan will detect/reject a lot with a contamination rate of >10%.

34. It should be noted that with this sampling scheme, a batch having, for scenario, a 9% contamination rate (so complying with the PO), would only have a 6.5% probability of being accepted. This illustrates that the prevalence sampling schemes do not have a very high discriminatory power.

Result of the step-wise process to establish an MC from the PO.

35. Given the PO, there is a less than 5% probability that batches/lots with a 10% contamination rate or higher would not be detected/rejected by a sampling plan with 29 samples ($n = 29$).

36. For practical purposes, this is a rather large *n*. It should be realized that this large number of samples is only able to detect a contamination prevalence as high as 10% with >95% probability. To have more stringent targets (i.e., lower acceptable prevalence or higher confidence), a much larger number of samples would need to be tested. The table below (Table 5A-2) illustrates this point for some lower values of acceptable prevalence.

37. Please note that several other aspects of an MC and the underlying sampling plan need to be additionally defined, as explained in the main text of the Codex guideline on establishing MCs.

Table 5A-2 (Adapted from van Schothorst et al., 2009; Food Control 20:967-979).

Sampling plans derived for *Salmonella* in carcasses intended to test compliance with different POs.

Proportion of contaminated carcasses tolerated (PO) (%)	Number of samples (*n*) required to reject defective lots with 95% probability ($c = 0$) *	Proportion of contaminated carcasses accepted with 95% probability (%) **
15	19	0.27
10	29	0.18
5	59	0.09
1	298	0.02

This could also be calculated with the negative binomial distribution:

NEGBINOMIAL(0;19;1-0.15) = 0.05; NEGBINOMIAL(0;19;1 - 0.0027) = 0.95.

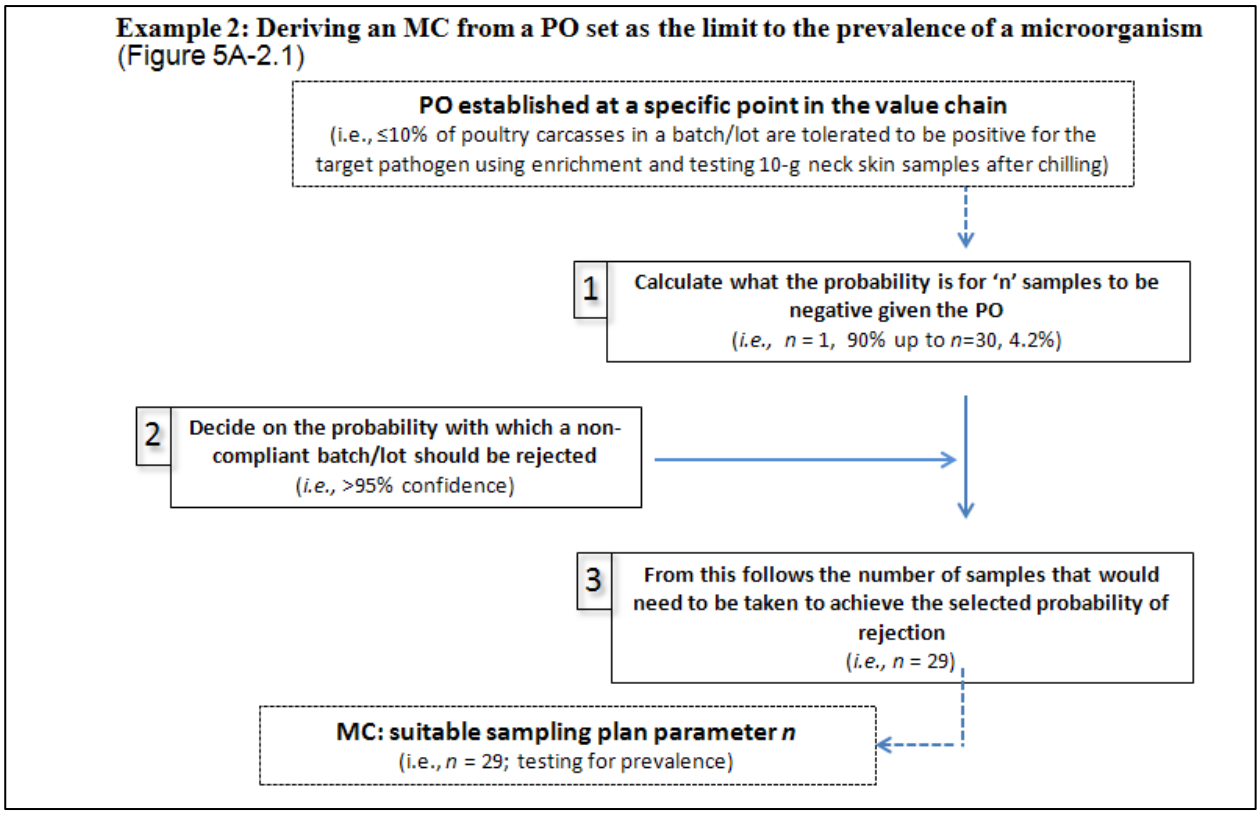
* $(1 - P)^n = 0.05$, $n \log (1 - P) = \log (0.05)$, $n = \log (0.05)/\log (1 - P)$.

** $(1 - P)^n = 0.95$, $\log (1 - P) = \log (0.95)/n$, $1 - P = 0.95^{1/n}$, $P = 1 - 0.95^{1/n}$.

Practical example for reference:

Analysis	**Standard/Guideline**				**Assessment**	
	n	*c*	*m**	*M*	**Satisfactory**	**Unsatisfactory**
Salmonella spp.	29	0	0	-	Not Detected	Present

* absence in a 10 g sample.



EXAMPLE 5A SCENARIO 3: Deriving an MC from an FSO for a product supporting growth of the target pathogen between PO and FSO

38. In this scenario, it is assumed that at the point of consumption, an FSO is set as a numerical limit to the acceptable concentration of a specific pathogen in a commodity that supports growth of a pathogen, after the point in the value chain where a PO/MC is to be set.

39. In this worked scenario, packed lettuce is the target commodity. Firstly, a suitable PO is derived, considering possible increases and decreases in pathogen concentrations between the point of the PO and the FSO. Secondly, a practically feasible MC is derived for the verification of batches/lots meeting the PO. Specific literature sources are referenced for relevant details.

40. For the purpose of this scenario, it is assumed that an FSO has been set for packaged lettuce (Zwietering *et al.*, 2010) and that this FSO is established as ≤ 0.2% of products having a pathogen concentration of > 100 cfu/g. In other words, the FSO requires that equal to or less than 0.2% of the products have a concentration of the target pathogen of > 100 cfu/g (see Table 2 in Zwietering et al., 2010).

41. The following step-wise process details the individual steps taken to develop this scenario. Steps 1-3 show how a suitable PO can be derived. Steps 4-7 describe deriving an MC for this PO. The overall process is depicted in Figure 5A-3.1.

Step-wise process:

1. Establish/Decide on the concentration distribution in the batch/lot and the standard deviation of the pathogen concentration at the point of the FSO

42. For typical batches/lots of packed lettuce at the point where the PO is to be set, it is assumed that pathogen concentrations are lognormally distributed and it is estimated that the standard deviation of pathogen concentrations is 1.112 log cfu/g (Zwietering *et al.*, 2010).

2. Calculate mean log concentration so that the distribution with this mean log concentration and standard deviation complies with the FSO

43. Batches/lots with a mean concentration of -1.2 log cfu/g and a standard deviation of 1.112 would exactly comply with the level set for the FSO (see Table 2 in Zwietering *et al.*, 2010).

$$P_{\text{normal,cumulative}}(2;-1.2,1.112)=0.998$$

(Please refer to Step 2 of scenario 1 for the method of calculation using PO=2; s=1.112 and $P_{normal, cumulative}$ = .998 to solve for μ)

44. With such a low mean concentration, quantitative testing based on enumeration of the pathogen in samples is not practical, since only 0.2% of the samples would be above a limit of 100 cfu/g. A sampling plan with a limit of 100 cfu/g that would be able to detect/reject non-compliant lots with >95% probability, would need at least 1497 samples, which is unrealistic.

[This can be calculated as 0.998^1497=0.05]

45. Therefore, in such a case, enrichment and presence/absence testing could be performed, moving from an enumeration approach to one using presence/absence. For example, using a sampling quantity of 25g and with the given distribution of concentrations, one sample would have a probability to detect/reject non-compliance of 63.7% (calculated by the ICMSF sampling tool, or by using the Poisson-lognormal distribution).

46. The following P_{reject} values can be calculated for '*n*' samples:

n	P_{reject}
1	63.7%
2	86.8%
3	95.2%
4	98.3%
5	99.4%

47. Thus, in order to detect a non-complying lot with a >95% probability, at least 3 samples of 25g should be taken.

48. However, verifying compliance at the point of consumption is generally not realistic and a suitable PO/MC should be established at an earlier point in the value chain.

49. The mean log concentration and standard deviation of the distribution of a batch/lot that complies with the FSO can be converted to a mean log concentration and standard deviation at the point of the PO, where an MC is then derived for verifying compliance. Insight into the likely/possible increases or decreases in log numbers of the pathogen between the FSO and PO (including the variability in these) would be important in making this conversion.

3. Derive a suitable mean log concentration and standard deviation of the distribution at the PO from the mean log concentration and standard deviation at consumption complying to the FSO

50. If a PO is defined earlier in the food chain, for example before storage and washing of the lettuce, the pathogen concentration can, for instance, decrease due to the washing process and it may increase during storage. Considering the decreases during washing and increases during storage [including the relevant level of reduction, i.e., 1.4 log cfu/g ; s.d.=0.5 log cfu/g, and the level of increase, i.e., 2.7 log cfu/g; s.d.= 0.59 log cfu/g], it has been estimated that the lognormal distribution of pathogen concentrations in the batch/lot at the point of the PO should have a mean log concentration of -2.5 log cfu/g with a standard deviation of 0.8 log cfu/g (see also Table 2 in Zwietering *et al.*, 2010), in order to account for changes in the pathogen levels between the PO and FSO.

51. [FSO=-1.2 with s.d. 1.112; reduction (R) is 1.4 logs with s.d. 0.50 and increase (I) is 2.7 logs with s.d. 0.59 (data from Zwietering *et al.*, 2010, based on experimental data of Szabo *et al.*, 2003);

$FSO=Ho-R+I$

$Ho=FSO+R-I=-1.2+1.4-2.7=-2.5$

$s^2(FSO)=s^2(Ho)+s^2(R)+s^2(I)$

$s^2(Ho)=s^2(FSO)-s^2(R)-s^2(I)=1.112^2-0.5^2-0.59^2=0.638$

$s(Ho)=0.80$

52. Notably, a batch/lot with a mean log concentration of -2.5 log cfu/g and a standard deviation of 0.8, has a 99.9% probability of having a concentration below 1 cfu/g (0 log cfu/g). This could be articulated as the PO level at the particular point in the value chain. Given the levels of reduction and increase (including the standard deviations for these) with their standard deviation, this articulated PO would be well aligned

with the established FSO.

4. Decide on the microbiological limit* m *for the sampling plan of the MC

53. A suitable microbiological limit needs to be chosen for microbiological analysis of samples, such that a practically feasible sampling plan is established. The methodology chosen is presence/absence testing of the target pathogen.

54. The microbiological limit (*m*) chosen in this scenario is the absence of the pathogen (non detectable) in 25g of product using an appropriate method of analysis: $m = 0$, in 25g samples.

5. Calculate what the probability is for 'n' samples, to detect a just compliant batch/lot, given its mean log concentration and standard deviation, and the value chosen for m

55. The probability of one sample from a batch/lot with a mean log concentration of -2.5 log cfu/g and standard deviation of 0.8 log cfu/g resulting in a positive detection is 18.5% (calculated by the ICMSF sampling tool, 18.531258% or by the Poisson-lognormal distribution). Thus, if taking 1 sample, the chance of rejecting a non-compliant batch (P_{reject}) would be 18.5%.

56. P_{reject} values for '*n*' samples taken can be calculated from $1-(1-0.18531258)^n$ and are shown below for selected values of *n*.

n	P_{reject}
1	18.5%
2	33.6%
3	45.9%
4	55.9%
5	64.1%
6	70.8%
7	76.2%
8	80.6%
9	84.2%
10	87.1%
11	89.5%
12	91.5%
13	93.0%
14	94.3%
15	95.4%

So with *n* going up you have a better chance of detecting a batch just complying with the PO

6. Decide on the probability that a lot with this prevalence should be rejected

57. A decision has to be made on the level of confidence with which non-complaint batches/lots will be rejected using the MC. Different confidence levels can be chosen considering the severity of the pathogen, typical concentrations occurring in the target commodity, constraints of the microbiological test chosen, the business impact of a possible high rejection rate of compliant batches/lots, etc.

58. In this scenario, it is assumed that a sampling plan that detects/rejects non-compliant batches/lots with greater than 95% probability is deemed appropriate.

7. From this follows the number of samples that would need to be taken to achieve the selected probability of rejection

59. From the calculation above, it can be seen that at least 15 samples of 25g would need to be tested and be found negative to conclude with > 95% confidence that the mean log concentration of the target pathogen in the batch/lot is less than -2.5 log cfu/g (with a standard deviation of 0.8).

60. Given the decreases and increases (with their variability) of the pathogen level after the point of the PO, these lots would comply with the FSO set as ≤ 0.2% of the units being above 2 log cfu/g at the time of consumption.

Literature references:

Szabo, E. A., L. Simons, M.J. Coventry, M.B. Cole. (2003). Assessment of control measures to achieve a food safety objective of less than 100 CFU of *Listeria monocytogenes* per gram at the point of consumption for fresh precut iceberg lettuce. Journal of Food Protection, 66, 256-264.

Zwietering, M.H., C.M. Stewart, R.C. Whiting, International Commission on Microbiological

Specifications for Foods (ICMSF). (2010). Validation of control measures in a food chain using the FSO concept. Food Control, 21, 1716-1722.

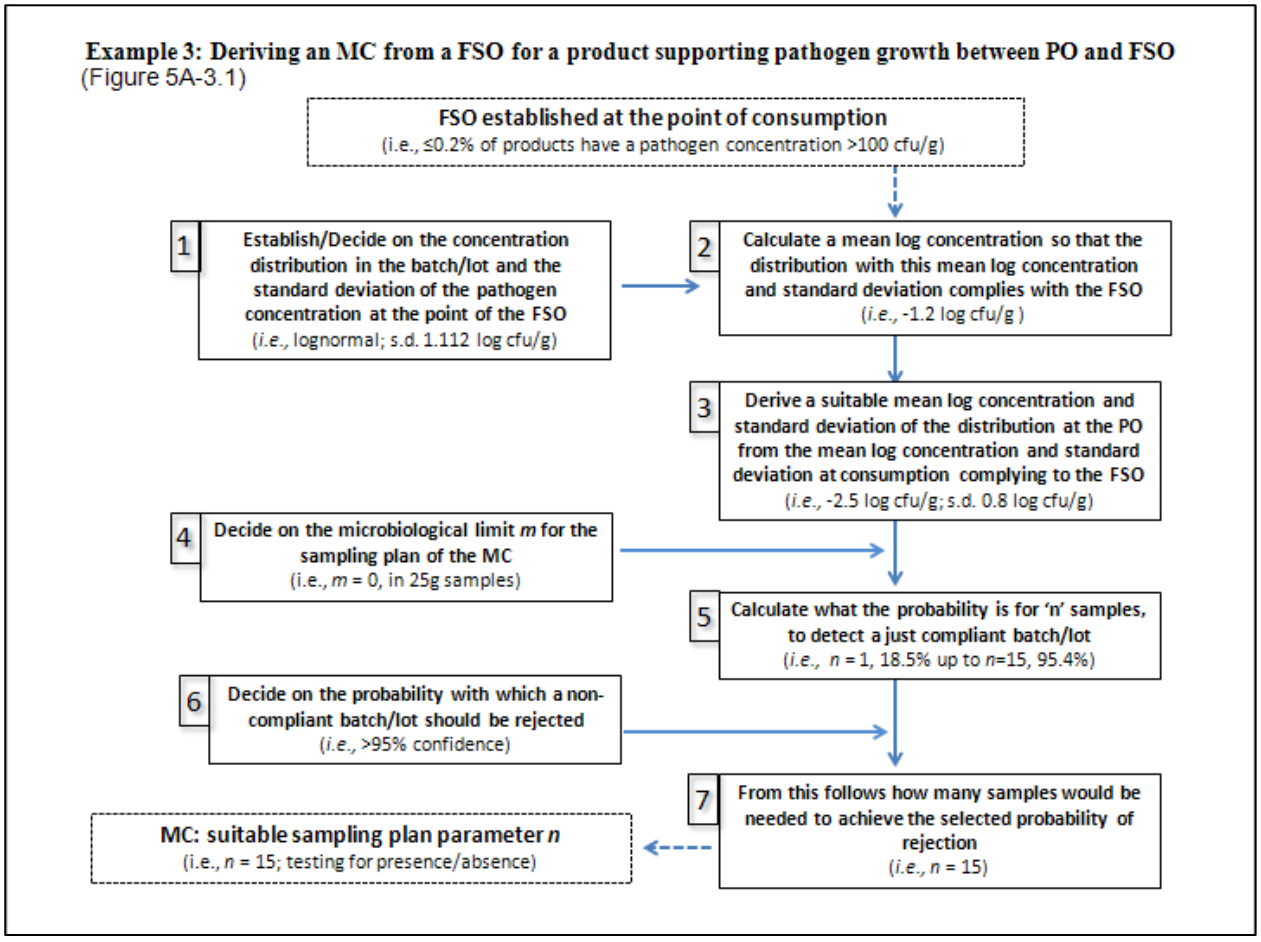


Figure 5A-3.1. The step-wise process for deriving an MC from a FSO for a product supporting growth of the target pathogen between the PO and the FSO

Table 5A-1. Responsibilities for developing metrics

Metric	Developed by	Comments	Example(s)
Appropriate level of protection (ALOP)	Governments/ member countries	An ALOP is level of public health impact based on what is currently achievable in a country (as opposed to a *public health goal* which looks forward to what one wants to achieve in the future)	Less than x cases/year of foodborne disease y in the country
Food Safety Objective (FSO) "The maximum frequency and/or concentration of a hazard in a food at the time of consumption that provides or contributes to the ALOP" (Codex definition)	Governments/ member countries	- FSO is a number, a frequency or a combination of both of a hazard in a food at the time of consumption - not necessary to establish for all foods - should only be developed where it assists in making a public health impact - From an FSO, one can derive an MC	x% of product is less than 100 cfu/g of *Listeria monocytogenes* in a smoked salmon product at the time of consumption

Metric	Developed by	Comments	Example(s)
Performance Objective (PO)	Industry Governments/ member countries	- can be established at any point in the food chain - A PO can be derived from an ALOP, FSO or another PO - From a PO, one can derive an MC - PO's may be stricter or more lenient than FSOs to account for any increases or decreases in the levels of a pathogen - industry can set POs to ensure that FSOs are met	No more than 10% of raw chicken carcasses after cooling can contain *Salmonella* spp.
Microbiological criterion (MC)	Governments/ member countries Industry	- an MC should only be established when there is a definite need and where its application is practical - for governments, the need is public health protection related, while for industry the need is meeting government or industry targets with regards to controlling hazards in foods - can be established from an FSO, PO or an ALOP - includes information such as the food product, the sampling plan, the method and the microbiological limit(s) to be met	*Cronobacter* spp. in powdered infant formula; n=30; c=0; m = 0/10g; 2-class plan; ISO method

EXAMPLE 5B: OPERATIONALISING A PERFORMANCE OBJECTIVE WITH A MICROBIOLOGICAL CRITERION FOR A RISK-BASED APPROACH

Introduction

1. In updating and revising the *Principles for the Establishment and Application of Microbiological Criteria for Foods* (CAC/GL 21; Codex Alimentarius, 1997), the Codex Committee on Food Hygiene (CCFH) decided to include practical examples that would be understandable and thus helpful to the users of the document. Two examples will focus on Performance Objectives (POs), which are articulated by risk managers and are consistent with their risk management priorities and policies. The goal of the examples is to illustrate the connection between a particular objective and the final product, in other words, to identify the risk and its associated Appropriate Level of Protection (ALOP). This example (5b) involves *Salmonella* in poultry because this microbiological hazard is universal.

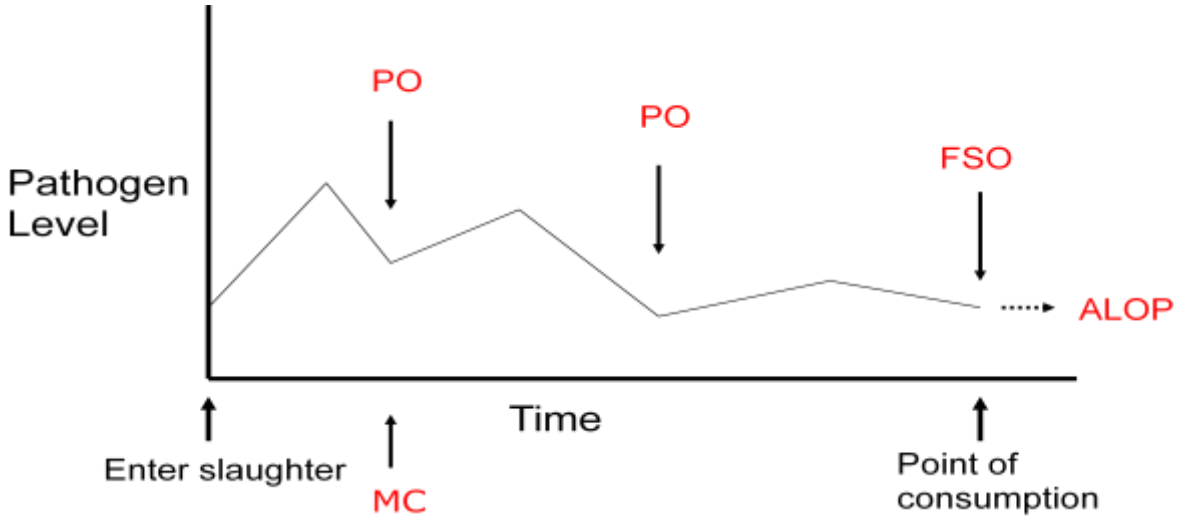
2. The Codex guideline *Principles and Guidelines for the Conduct of Microbiological Risk Management* (CAC/GL 63; Codex Alimentarius, 2007) explains the risk management metrics used here and the relationship between them. In terms of a practical approach, the user will identify a PO at a specific point in a food processing system (food chain) and determine if that PO is practical in terms of setting a microbiological criterion (MC) with its accompanying sampling plan (for more discussion, see e.g., Havelaar et al., 2004; van Schothorst et al., 2009; Crouch et al., 2009). Since a PO is conceptually linked to the ALOP, the impact of the steps in the food chain before and subsequent to the PO needs to be considered when articulating a PO. Therefore, a risk assessment that estimates the level of risk (or limit of a microbiological hazard) at each point of the food processing system is necessary to link the PO to the level of risk to the consumer at the end of the food chain.

3. This example is drafted by the working group for example 5b. It has not been peer reviewed at this time. The CCFH may decide that if this example, with other examples from other working groups, are to accompany the main "*Principles*" document, then this example would be subject to peer review, most likely via a mechanism through FAO/WHO.

Purpose (what is intended to be achieved)

4. The establishment and application of an MC serve different purposes for different organizations (either regulatory authorities or food business operator). The primary purpose of this example (5b) is to extend beyond the establishment and application of MC from GHP-based and HACCP-based to operationalize a PO with a microbiological criterion MC for a risk-based approach. The point in the food chain where the PO should be described also depends on the practicality and feasibility to apply the PO to any specific point in the food chain. Once countries have determined their own ALOP via appropriate risk assessment, related risk management metrics (such as POs and MCs) can been determined, e.g., an MC would be determined for the selected PO. A compatible sampling plan together with statistical performance characteristics can also be determined.

5. The graph below is a representation of a food processing system (line on the graph) from beginning of slaughter to the point of consumption. The appropriate risk assessment will help determine what points in the system where POs can be described – the risk assessment links the POs to the ALOP. The Food Safety Objective (FSO) is another risk management metric that is conceptually similar to the PO, but is determined at the point of consumption. A decision is then made for which PO is the most practical (here the first one is selected, but more than one PO may be selected if desired). Then an MC is determined for that PO. Ultimately, a sampling plan compatible with the MC and practical to implement is then determined (and its performance characteristics detailed).



6. It is not the intention of this example to set quantitative limits for *Salmonella* in broiler chickens for international trade. This example provides a procedure that can enable countries and industries to establish an MC appropriate to the situation in their own countries.

Who should establish and who should apply

7. Risk managers in national governments usually establish the risk management metrics for their countries. They oversee the entire food chain for their country and they most likely have the authority to set such standards. Also, national governments are usually the ones that perform the necessary risk assessment(s) that allow them to articulate the level of control of a hazard via the PO. Once the PO is articulated, the appropriate MC can be established. Without a national PO they must follow, food business operators may find it beneficial to establish their own POs to maintain a level of control in their business; these are not universally common and usually relate to their specific position in the food chain.

8. Once a PO has been established, both national governments and food business operators apply the appropriate MC to achieve that PO.

Risk assessment

9. To enable a risk-based approach for the articulation of a PO, a risk assessment is necessary to provide the link between the PO and the level of risk to the consumer at the end of the food chain. The World Health Organization and the Food and Agriculture Organization of the United Nations (WHO/FAO) have published an assessment of risk of *Salmonella* in eggs and broiler chickens (WHO/FAO, 2002a,b). It is referenced by many national governments and is a useful reference for this risk-based approach.

10. The nature of the link between formal risk assessment processes and the ALOP concept is still being conceptualized. Estimating the ALOP requires the construction of an abstract model food system that delivers the level of food safety that is the manifestation of the minimum set of official requirements of the domestic market (see Figure 1). Risk assessment addresses the best practices involved in deriving, in a way that maximizes objectivity and completeness, an estimate of the health risk that is expected from a system, as well as the level of certainty that may be associated with those estimates. This requires the inclusion of all forms of variability with the goal of adequately describing real-world conditions and to improve the quality of population health risk estimates. It further recommends the characterization and quantification of uncertainty, ideally, to generate a quantified measure of the level of confidence that may be associated with the risk estimates.

Figure 1: Representative risk assessment model

11. The outcome of the risk assessment is a quantitative statement of what must be achieved by food producers at a specific point in the food production system, in order to achieve a particular system-wide goal. The quantified outcome is not descriptive of a physical reality as is expected in descriptive application. Rather, it is a quantitative allocation of responsibility for performance in the food system.

12. As a food safety design application, the concept of a design load could be implemented in the form of a quantitative risk assessment model that explicitly includes a level of variability in downstream conditions that upstream producers would be reasonably expected to accommodate. The level of variability that is described in the model would constitute a blend of uncertainty and variability, to the extent that it is determined that it is reasonable to expect the upstream producer to accommodate it. If it is reasonable to expect the upstream producer to accommodate all of the downstream variability and the associated uncertainty in the extent of variability, then a wide distribution of variability could be included, resulting in more stringent performance requirements. If it is only reasonable to expect upstream producers to accommodate compliant downstream behavior, then the variability included in the model would be limited to that which would be considered the normal variability among compliant behaviors. The resulting risk assessment model (Figure 2) would allow for the derivation of performance objectives by determining the level of performance that would generate a tolerable level of risk in the model system.

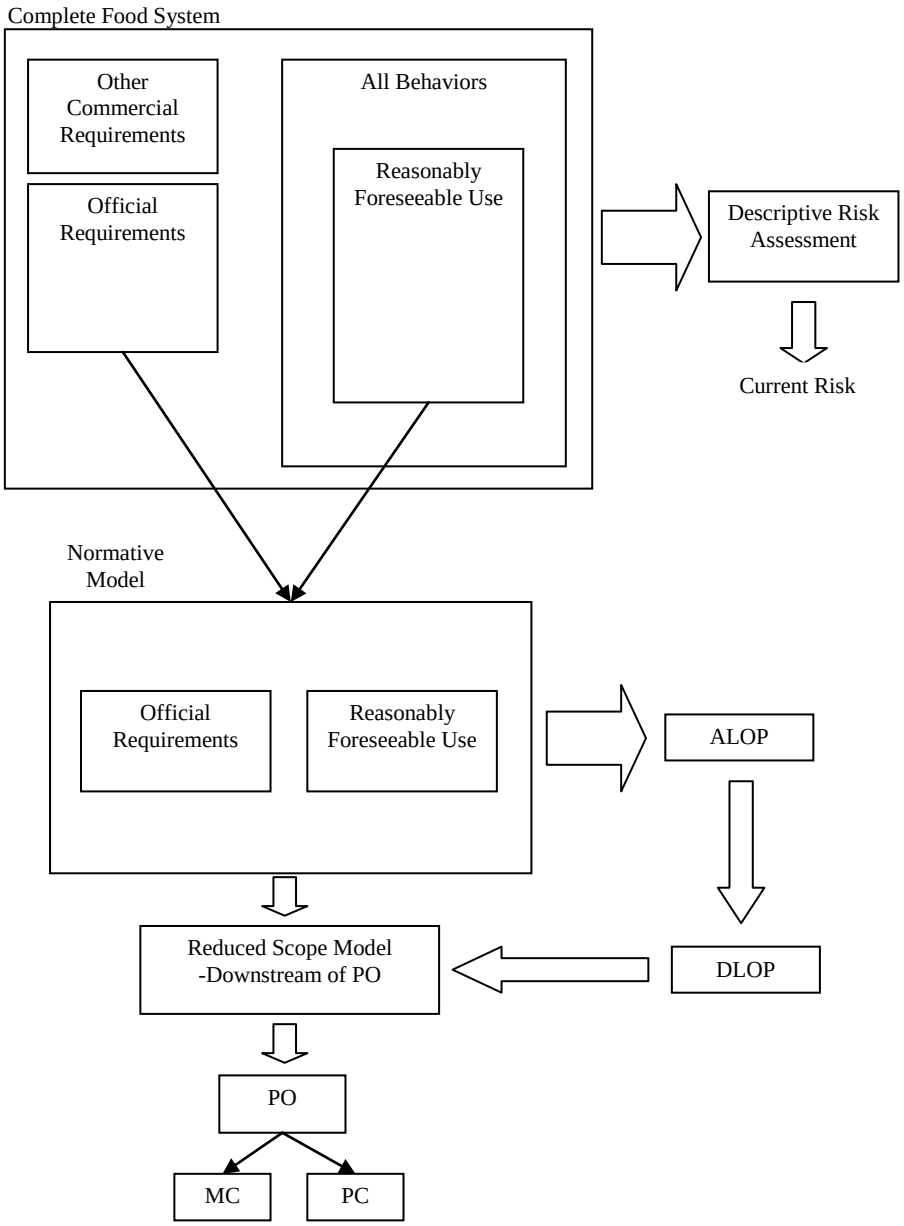


Figure 2: A schematic of the applications of risk assessment. The top of the diagram represents the standard risk assessment paradigm. The bottom of the diagram describes a normative application of risk assessment, which can be linked to the concept of the ALOP and the establishment of

performance objectives. The ALOP in this circumstance can also be thought of as a desired level of protection (DLOP) or public health goal based on the food processing system being modeled.

Food or food process; Point in food chain where the MC is applied

13. The food examined is broilers (whole fresh chicken) sampled during the slaughter process.

14. The point in the food chain that appears to be the most practical to apply an MC is at post-chill. This point is described in the *Guidelines for the Control of Campylobacter and Salmonella in Chicken Meat* (CAC/GL 78; Codex Alimentarius, 2011). Specifically, the MC would be applied at post-chill after Steps 19: Chill Carcass (air or immersion) and 20: Post-Chill Applications (immersion, spray, or dip) and before Step 21: Portion (i.e., at the end of the drip line after all interventions have taken place, but before the bird enters the cooler or proceeds to further processing).

Organism of concern

15. *Salmonella* spp. is the selected organism. This organism is a universal microbiological hazard in poultry and fits well for the purposes of this example.

16. The risk assessment model used in the MC determinations for this example includes the assumption that it examines all *Salmonella* spp. The result is that the MC calculations assume that the *Salmonella* spp. that are detected are capable of causing human illness. Therefore, there is an additional level of protection afforded by the MC to the extent that some proportion of the detected *Salmonella* are not pathogenic to humans. If the nature of the pathogenic versus non-pathogenic *Salmonella* is a key consideration for a country's analysis, it could be included in a parallel analysis, by adjusting the estimate of *Salmonella* prevalence for the proportion of the *Salmonella* serovars that are expected to be in the product, but are known to be non-pathogenic to humans.

Sampling plan (sampling approach, # of samples, sample size/units)

17. Since prevalence will be used for the interpretation of *Salmonella* presence, a two class plan is utilized.

18. Typically, a rinsate will be sampled. Once a whole fresh chicken carcass has been selected at post-chill, a rinsate of the whole carcass is taken. The carcass is typically placed into a sterile collecting bag and 400 ml of a rinsing solution is applied (rinsing solution: Buffered Peptone Water (BPW)). As much of the BPW rinsate is collected as possible (but at least 200 ml). National governments will likely have specific instructions for how the carcass is selected and handled.

19. The number of samples to be collected is determined by the MC. Carcasses for sampling should be selected at random, regardless of the origin of the carcass, and samples should be taken daily until the numbers of samples specified in the sampling plan are collected. The lot or farm where the carcass originated does not need to be considered because variability derived from the source of the product is already considered when establishing the criterion to manage risk. Alternatively, samples could be taken on a continuous basis and the information from the sampling can be applied to the established PO using a "moving window" of samples.

20. Once the rinsate is collected, it is sent to the laboratory for analysis. The rinsate is kept at 4^{o} C until analyzed. A 30 ± 0.6 ml portion of the rinsate is used as the analytical unit for the MC.

21. While rinsate at post-chill is the selected sample for this example, it is noted that there are other possible samples to utilize and points to sample in the processing system (with an associated PO). For instance, also at post-chill, 25 g of skin and muscle from the neck, wing, and cloaca can be taken. Other points to sample may include at re-hang (after the picker and prior to evisceration of the bird) or taking 25 g from final product. If samples other than rinsate are used, some degree of compositing samples could be explored (rinsate samples are not combined).

Method(s) of analysis

22. The analytical unit will be assayed for the absence or presence of *Salmonella* spp. National governments typically apply their method of analysis (e.g., see USDA/FSIS, 2011). Many national governments also rely on available methodologies such as from the International Organization for Standardization (ISO) which has a methodology, ISO 6579, for analyzing *Salmonella* (ISO, 2002).

23. Other methods that provide equivalent sensitivity, reproducibility, and reliability can be employed if they have been appropriately validated.

Derivation of POs and MCs based on Risk Assessment Model

24. The purpose of this procedure is to establish a set of candidate MCs for consideration by risk managers. These criteria are derived to provide a specified degree of confidence that poultry production systems that can achieve these criteria will have levels of prevalence and concentration of *Salmonella* spp. that generate a tolerable level of risk that is estimated in the determination of an ALOP. The approach is discussed in the following paragraphs, but a more general step by step discussion is provided in Appendix 1.

25. The analysis to determine a microbiological criterion for *Salmonella* spp. in poultry uses a quantitative risk assessment that considers the processing to illness continuum, ending with an estimate of the burden of disease, specifically Disability Adjusted Life Years (DALYs). The risk assessment model has been adapted to be a normative model, in that it considers only storage, and cooking practices that can be expected given reasonable use of fresh, whole chicken. Specifically, for the purposes of this example, extremes of temperature abuse and undercooking have been removed from the analysis. Running the normative risk assessment model leads to an estimate of the burden of disease from *Salmonella* of 0.8 DALYs per million servings that may be expected through reasonable storage and preparation of fresh, whole chicken. Setting this as the basis, the model was then used to determine the combinations of *Salmonella* concentration and prevalence that would at least meet this goal (0.8 DALYs per year) for a particular point in the processing chain. These combinations are the Performance Objective (PO), and for this analysis are set at post-chill. Each PO is defined in terms of the median concentration (log CFU per carcass), the standard deviation of concentration, and the prevalence of *Salmonella* (per carcass). The analysis results in the following set of combinations that meet the target:

Prevalence (per carcass)	**Concentration (Log CFU/carcass)**	**SD (Log CFU)**
0.2	2.9	0.5
0.075	3.5	0.5
0.02	3.5	1
0.04	3.9	0.5
0.2	1.5	1

26. National baseline studies that determine prevalence and concentrations of specific analytes (in this case *Salmonella*) are commonly used to determine the parameters that should be used to inform these calculations. Samples in the baseline study would ideally be collected in multiple establishments, over the course of a year (to account for seasonal variation), at the same point in the process where the PO is to be established, and using an analytic method equivalent to the method proposed in the sampling plan. Additionally, the number of samples taken should be large enough to ensure a high degree of statistical confidence that the results reflect the "true prevalence" of the analyte in the country.

27. For each of these possible POs there is a set of possible sampling plan options that will meet the level of control required. For this analysis, a sampling plan is developed that demonstrates that a process system is in compliance (i.e., process control) with 95% confidence given the associated prevalence and standard deviation. Selecting a PO from above, the set of acceptable sampling plans can be established. For this example, a PO of concentration = 1.5 log CFU/carcass, standard deviation = 1, and prevalence = 0.2 is used. For sampling plan analysis, a 400 ml rinse volume is assumed; therefore the concentration that is controlled for is -1.1 log CFU/ml. Using an analytical sample size of 30 ml, the following sampling plans provide control for -1.1 log CFU/ml with at least 95% confidence (this analysis assumes 100% recovery):

N	**C**
20	0
32	1
43	2

Microbiological criterion for Whole Chicken Carcasses for Salmonella

Point of application	Microorganism	n	c	m	Class Plan
Applied at post-chill	*Salmonella* spp.	X	Y	Absence in 30 ml rinsate	2

Where:

n = number of analytical units analyzed

c = the maximum allowable number of defective sample units in a 2-class plan

m = a microbiological limit (where, in a 2 class plan that measures presence and absence of *Salmonella* in the samples, m = 0)

X and Y = the n and c, respectively, chosen from the set of acceptable sampling plans from the above analysis (e.g., n = 43, c = 2 as one choice from the table above)

Example operating characteristic curves are shown in Appendix 1.

Interpretation of results and other considerations

28. Simply stated, if the process is found to be non-compliant with the MC, the processing system is suspect to not adequately control *Salmonella* present in the chicken processing system.

29. The criterion chosen for the individual country is to provide a specified degree of confidence that the process will control levels of *Salmonella* present in food to reduce the risk of illness to consumers.

30. Since a normative model approach was used in this example, extreme behaviors outside the "norm" were not taken into account in the risk assessment model. Countries can use their risk assessment to include such extreme variations in the process being modeled and would therefore generate a different set of n and c combinations than those found in this example.

31. The procedure described above would ideally generate a set of candidate criteria that provide the approximate level of protection desired. The final selection of an MC would be made by risk managers in consideration of other relevant factors, including cost, practicality and linkages to other MCs. As an example, if the same field samples are to be used for multiple purposes (e.g., also to test for *Campylobacter*), then the number and size of samples may be driven partly by this consideration. This can lead to an iterative interaction between the risk assessor and risk manager, to find a set of criteria that best suit the situation, while still having a known linkage to the level of protection that is provided by the MC that is ultimately selected.

32. Due to uncertainties in the risk assessment processes and scientific assumptions underlying the calculations, the specific quantities derived for an MC (e.g., sample size, number of samples to be taken) should not be understood as a precise and inflexible number. For example, one could calculate the performance of a sampling plan of 9 samples of 9.5 g. It would be a reasonable expectation that this would be rounded up to 10 samples of 10 g as the final determinations are made in consultation with the risk management function.

(Nature of) actions in case of non-compliance

33. Non-compliance is determined whenever the c value is exceeded, in which cases appropriate actions by food business operators and/or national competent authorities are necessary to regain process control and conformity with the MC. Competent authorities should decide on which actions suit the national plan being developed. CAC/GL 78 (Codex Alimentarius, 2011) recommendations should be used as a reference for the development of risk-based control measures.

34. In the present example, the first step would be to adopt strategies to prevent any products possibly hazardous to public health as a consequence of the deviation to enter commerce. Possible actions include detention or recall of products. If the prevalence of specific pathogenic *Salmonella* that may be involved are known, a specific microbiological criterion for such pathogenic *Salmonella* could be developed.

35. The second step would be to determine strategies to decrease the microbial load of any products possibly hazardous to public health as a result of the deviation. The competent authority usually recommends approved chemical and/or physical inactivation means so that the specific non-compliance

products would recover the conformity of the MC. Possible actions include the use of chemical decontaminants approved by the competent authority, as well as heat treating the products. The selected actions should be appropriately validated in order to show a reduction of contamination adequate to eliminate the hazard detected (e.g., the competent authority may have an expected log reduction of the *Salmonella* load).

36. The third step would be to adopt strategies to regain process control and to promote conformity with the MC. Recommendations of CAC/GL 78 (Codex Alimentarius, 2011) should be taken into consideration when developing hazard-based control measures. Actions that might be necessary can include: increase of hygiene procedures and monitoring, reassessment of the HACCP and pre-requisite plans, and/or lot by lot testing to determine product acceptance.

37. The fourth (and likely last step) would be the official verification on the adequacy and effectiveness of the actions taken. The procedures might include the suspension of official health certification on the absence of *Salmonella*, a "follow-up" sampling plan with increased stringency of the sampling plan, as well as the conduct of an official investigation on the root causes of the deviation and corrective actions taken by the food business operators, which could lead to additional enforcement actions.

38. Additionally, a trend analysis on the national plan results could provide important information on possible improvements necessary in the national food inspection system, as well as the adequacy and/or need for reassessment of the PO that has been set.

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Appendix 1: Example Process for Deriving POs and MCs from an ALOP using a Quantitative Risk Assessment Model

Overview of Derivation Process

1. This example illustrates one possible process for deriving performance objectives (POs)and associated microbiological criteria (MC) including the linkage to the concept of an ALOP. The derivation process is based on the application of a previously developed risk assessment model that estimates the risk by modelling the fate of pathogens from the post-chill step in a poultry slaughter operation through to domestic consumption and corresponding illness.

Step 1: Convert a descriptive risk assessment model into a normative model

2. The risk assessment previously derived was based on describing a broad range of downstream variability in the way poultry may be stored, handled and prepared. This variability is important to describe the level of risk that may exist in the population. However, for performance measurement, it may be desirable to explicitly derive a normative model that excludes some amount of downstream variability, leaving only that which might be deemed "reasonably foreseeable" downstream use. This model may be more appropriate for allocating responsibility in the food system (which is essentially what a PO imposes).

Step 2: Establish an ALOP based on the normative risk model

3. A target level of health risk can then be established (for example, a 10% reduction in risk from this level). For illustrative purposes, the target level of risk will be given the label ALOP to be consistent with current documentation on risk management metrics (though the terminology is not strictly appropriate given the legal meaning of the ALOP).

Step 3: Select a point within the production system for the PO

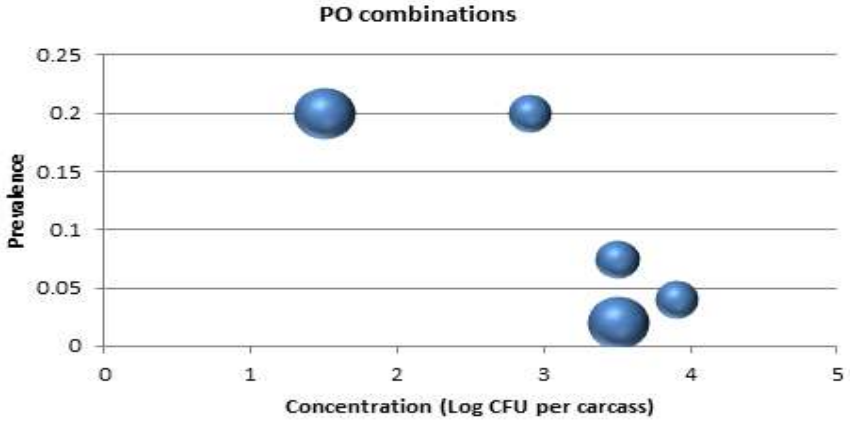
4. The risk manager, based on technical, practical and legal considerations, chooses points in the continuum for which POs may be derived. In this example, the post-chill point of poultry slaughter is selected as one such point.

Step 4: Conduct a Search for candidate POs

5. For the case of poultry production, both prevalence and concentration are thought to be important, since different management approaches can be applied to each of these dimensions of measurement of the level of contamination.

6. By repeatedly simulating the risk assessment model, it is possible to identify a set of combinations of prevalence (P), and the median ($\log_{10}\mu$) and standard deviation ($\log_{10}\sigma$) of concentration, among carcasses at the post-chill point in production, that lead to a level of risk that is less than or equal to the chosen ALOP. These combinations have a known relationship to risk and are referred to here as candidate POs.

7. The figure below shows a set of candidate POs with the associated prevalence (P) on the y-axis, the median concentration on positive carcasses on the x-axis, and the standard deviation illustrated by the size of the circle. Note that all three sub-measures of performance are inversely related; as any one increases, one or both of the other two must be reduced to achieve the same level of risk.



Step 5: Select one or more candidate POs for further consideration

8. Based on practicality and other considerations, select one or more candidate POs from which one or more MC may be derived. The ability of industry (or the specific part of the industry for whom the PO is being considered) to meet a particular combination may be the dominant selection criteria, presuming that the PO is most effective if it is seen to be achievable with currently available risk mitigation options (i.e., it can be achieved by at least part of the industry now, or is known to be achievable with particular mitigations in place).

Step 6: Calculate a MC that demonstrates performance in meeting a PO

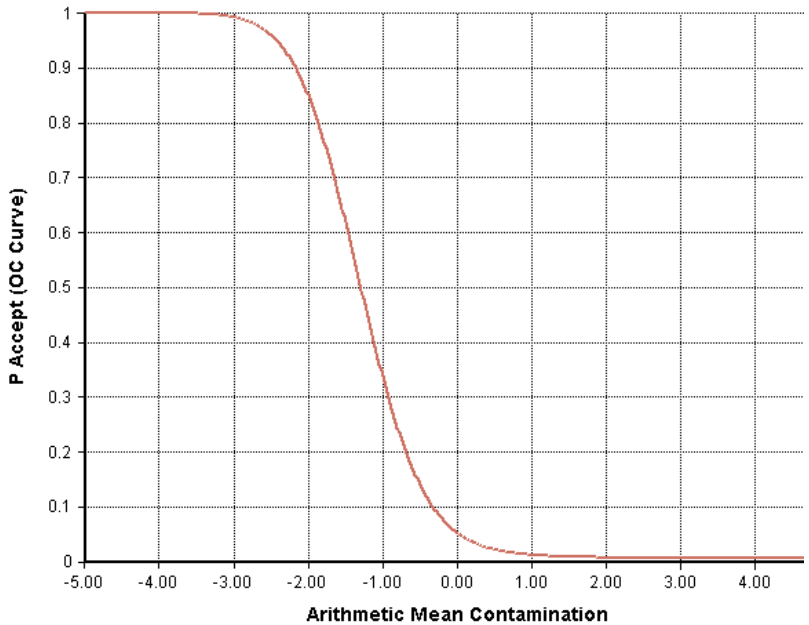
9. To establish a PO, the risk manager must then select a level of tolerance for accepting non-performing product (often rejection rates such as 95%, though there is no scientific or public health basis for the frequent selection of the value 95%). With this tolerance, it is possible to determine an MC, through standard statistical calculations, for which a production system that is performing at the level of the PO or worse will "fail" at the prescribed rate. The scope of potential MCs may need to be reduced for technical, practical or economic reasons. For example, in this case, it was determined to limit the scope of MCs to presence/absence based testing for all *Salmonella* spp. (as opposed to including concentration-based testing, or consideration of only specific serovars that are pathogenic to humans). This choice of using presence/absence sampling might be very different if the illustrative example had focused on *Campylobacter* spp.

Step 7: Derive additional MCs for one more POs and present to Risk Manager

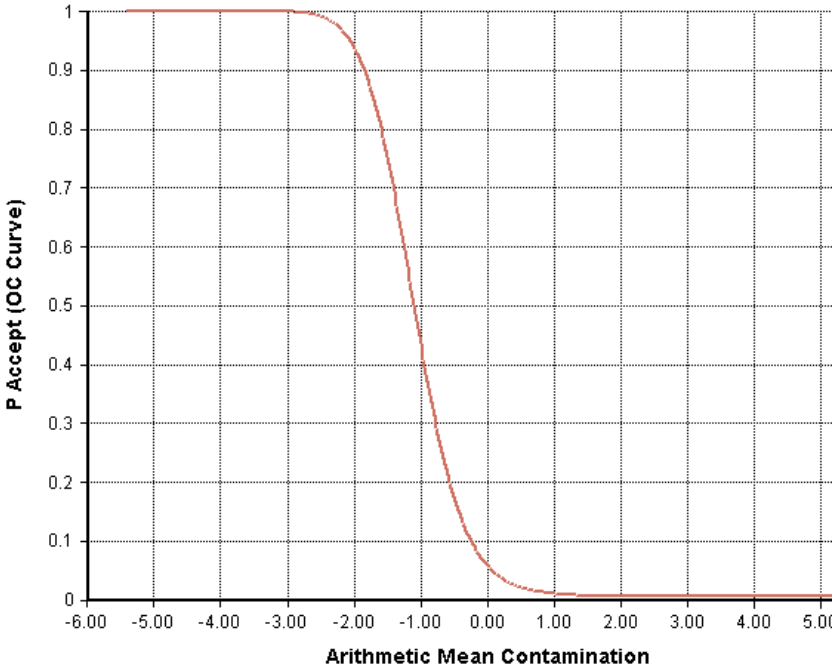
10. The selection of POs and MCs is not a purely analytical process and requires considerable input from a risk management perspective. Ideally, the analyst will provide an array of potential ALOP/PO/MC combinations, with associated explanations of the level of key assumptions (particularly those related to inherently value-laden components like the tolerable probability of rejection). The purpose of the array of ALOP/PO/MC combinations is to allow the risk manager to consider other important practical and related inputs including the importance of feasibility, the amount of product that might be rejected in the sampling process, the cost of the associated sampling regime, other test strategies for other hazards like *Campylobacter* spp. in the same product.

11. The figures below demonstrate the operating characteristic (OC) curves (showing the probability of accepting the product ("passing the test" defined by the MC) as the concentration increases. These can also be provided in tabular form. Note that this demonstration is limited to presence/absence based tests. It is important to understand that there are a very large number of possible MCs possible for any given PO, and a large number of potential candidate POs. The scope of the candidate POs and MCs to be considered needs to be limited in consultation with the risk manager based on practicality, cost and other considerations.

N=32, c=1



N=43, c=2


P Accept (OC Curve)
1
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
-6.00
-5.00
-4.00
-3.00
-2.00
-1.00
0.00
1.00
2.00
3.00
4.00
Arithmetic Mean Contamination