

The Limitations of Microbiological Sampling

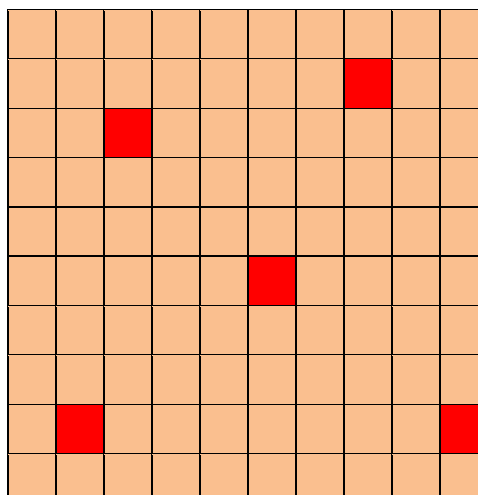
Microbiological sampling can be used to verify effectiveness of food safety management systems such as the adapted HACCP-based system outlined in the *Guide to Good Hygiene Practices in the production of artisan cheese and dairy products*.

It is important to stress however that there are limitations to the effectiveness of basing food safety management solely on testing and this was the reason that HACCP was first developed in the 1960's to ensure that foods developed for the space programme would be safe for astronauts to eat.

The certainty of finding a contaminant during microbiological sampling can be calculated using a statistical function called "*hypergeometric distribution*". Without showing the complicated equations to calculate it, we can look at certainty in the example shown below.

Finding a contaminant with a single sample.

This grid below is made up from 100 squares. 95 of them are green and 5 of them are red. We can say that the red squares have a **prevalence** of 5%. These represent unsatisfactory samples which show a non-conformity (such as contamination with a low level pathogen). The green squares represent satisfactory (non-contaminated) samples.



We know this batch of squares contains a number of non-conformities, but let us imagine a situation in which this is not yet known...

A person is blind-folded and asked to point to a single square from the 100, the certainty that they will point to a red square and therefore identify that the batch contains a non-conformity) is 5%.

There is a greater likelihood that they will not detect the non-conformity with one sample

Finding a contaminant with five samples.

Now let us imagine that the person was blind-folded and asked to point to five different squares.

Why five? While a food business operator can use a lower sample number for routine testing, Regulation (EC) 2073/2005 sets out the minimum sample number where the purpose of sampling is to **assess the acceptability of a batch or process**. For the food safety criteria, such as *Salmonella*, *Listeria monocytogenes* or Staphylococcal enterotoxin, the minimum sample number is $n=5$.

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The certainty that at least one red square will be chosen in a selection of five squares is 23%.

There is a greater likelihood that they will not detect the non-conformity with five samples.

In order to identify contaminants with greater certainty of detection, **13 samples** would be needed in order to have a **51% certainty of detection** while **95% certainty** would require **45 samples!**



Conclusion

- Sampling is an ineffective way of detecting low level contamination unless the number of samples taken is very large.
- Sampling every batch with single or multiple samples is unlikely to ensure consumer safety.
- 'Positive release' of products is not an effective way to manage food safety.
- **Food safety management systems should be based on hygienic milk production and control measures during the manufacturing process.**
- The producer should focus on maintaining and, where appropriate, improving milk production hygiene and good manufacturing practices in line with the recommendations given in *Guide to Good Hygiene Practices in the production of artisan cheese and dairy products*.

Training Exercise: “Microbiological Bingo”

Notes for Trainers

Two possible classroom-based exercises are outlined below in order for the trainer to demonstrate the limitations of microbiological sampling.

The exercises take the form of a game called “Microbiological Bingo”. It can be linked to training on HACCP (especially verification methods), self-monitoring and non-conformity management.

It is important for the trainer to convey the message that effective HACCP-based plans are a more reliable way of ensuring consumer safety than through increased sampling alone.

Exercise 1

The trainer should ask their trainees to pick one number between 1 and 200 and write it on a piece of paper.

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The trainer should pick ten numbers between 1 and 200 and write them on a piece of paper without disclosing them to the trainees.

The trainer should tell the trainees that the numbers represent the number of 25g samples in a batch of cheese made by a small producer. The batch consists of 10 x 500g cheeses; 5kg in total. The trainer should state that, unknown to the cheesemaker, the batch is contaminated with *Salmonella* with a prevalence of 5% (i.e. 5% of the samples will show the contaminant).

The trainer should begin reading out the numbers and the trainees shout call “bingo” if their number is called.



The trainer should ask the students to consider the proportion of the students who successfully identified the contaminant and the proportion who missed it.

Exercise 2

The trainer should ask their trainees to pick five numbers between 1 and 400 and write them on a piece of paper.

The trainer should pick twenty numbers between 1 and 400 and write them on a piece of paper without disclosing them to the trainees.

The trainer should tell the trainees that the numbers represent the number of 25g samples in a batch of cheese made by a small producer. The batch consists of 5 x 2kg cheeses; 10kg in total. The trainer should state that, unknown to the cheesemaker, the batch is contaminated with *Listeria monocytogenes* with a prevalence of 5% (i.e. 5% of the samples will show the contaminant).

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The trainer should begin reading out the numbers and the trainees shout call “bingo” if any of their numbers is called. The game continues until all twenty numbers have been read out.

The trainer should ask the students to consider the proportion of the students who successfully identified the contaminant and the proportion who missed it. The trainer should then ask to consider how many students identified the contaminant in:

- i. Two or more samples.
- ii. All five of their samples.

Suggestions for further discussion:

The trainer may wish to initiate group discussion on any of the following topics:



- The 'clustering' of microorganisms within a food.
- Sharing experiences of microbiological non-conformity.
- Explaining the microbiological criteria applicable to dairy products and the meaning of sample number (n), "big-M" and "small-m" as outlined in Regulation (EC) 2073/2005.
- Reduction of sample number based on historical results and the requirement to test with $n=5$ in the event that contamination is suspected or a new process is developed.

Suggested questions for discussion:

- *Listeria monocytogenes* is identified in a batch of ripened blue cheese. The physicochemical parameters of cheese suggests that it is able to support the growth of *Listeria*. After holding the cheese for a period of two weeks, a single sample is analysed for presence of *Listeria monocytogenes*. *Listeria* is not detected in the 25g sample. Can the cheese be placed on the market?
- Coagulase-positive Staphylococci are identified in a hard cheese at 48 hours after production. The count exceeds 100,000cfu/g. A 25g sample is sent for toxin testing but staphylococcal enterotoxin is not detected. Can the product be placed on the market?