Microbiological Testing Basics

Robert L. Buchanan University of Maryland

- Testing foods against a specified "criterion"
 - Pathogens
 - Indicator microorganisms



- Set of technologically-based, statistically-based tools
 - Safety of "batches"
 - Process control verification
 - Process control validation
 - Investigational sampling
 - Surveillance sampling
- Need right tool for the right job



- Microbiological testing is inherently "probabilistic" in nature
 - Must consider the likelihood that the target microorganism is in the food sample being examined
 - Cannot prove absence, only disprove it
 - Negative findings are used to determine
 - The likelihood of contamination is below some preset value
 - What level of confidence one can have in negative results

- Generally works on the assumption that
 - Samples are taken randomly and independently
- Concept of "defect rates" critical to understanding microbial testing of foods

Types of Microbiological Testing

- Two general classes of sampling plans are used to evaluate the microbiological safety or quality of a food batch
 - Attribute:
 - 2-class: "presence/absence"
 - 2-class: "numerical" (e.g. ≤100 vs. >100)
 - 3-class: Numerical with inclusion of marginally acceptable (e.g., ≤10 vs. >10 - ≤100 vs. >100)
 - Variables:
 - Direct use of quantitative data
- These basic forms include both cultural and "omics" based testing

Microbiological Testing Stringency

- Utility, effectiveness and cost of testing is dependent on:
 - Inherent cost of analysis
 - Defect rate
 - Distribution (variance) of contamination
 - Number and size of samples
 - Frequency of sampling
 - Method's lower limit of detection (i.e., when is zero zero?)
 - The degree of confidence required (i.e., 90%, 95%, 99%?)

Sampling Site

- Microbiological contamination typically flows with a process
- Microbiological sample integrates all the preceding processing steps
- Conceptually nothing special about end product testing
 - This is the point where regulatory controls start



Requirements for Detection

- Two primary factors determine the lower limit of detection
 - Number of microorganisms needed to distinguish "signal" from "noise"
 - Impact of the particulate nature of microorganisms
 - Bacteria are not chemicals
- Same principles are true for traditional and rapid methods



Requirements for Detection

- The size of the analytical unit is a key determinant
- To be 99% confident that at least one cell will be present in the analytical unit, the Mean Log Concentration must be:
 - 1 μl: MLC = 4.3 Log(CFU/ml)
 - 10 μl: MLC = 3.3 Log(CFU/ml)
 - 100 μl: MLC = 2.3 Log(CFU/ml)

Requirements for Detection

- To get around this key limitation, one can:
 - Increase analytical unit size
 - Increase the number of analytical units examined
 - Concentrate the target microorganism from the sample
 - Enrich the sample to allow target bacterium to increase in numbers





Conclusions

For more information, see <u>www.icmsf.org</u> or consult ICMSF Books 7 and 8.



• Microbiological Testing:

- Is a set of technologicallybased, statistically-based tools
- The sensitivity and effectiveness of these tools are dependent on several factors including the microbial levels in the sample
- Understanding the principles that influence microbiological testing is key to its effective use