INTERPRETATION OF TEST RESULTS - MICROBIOLOGY

Presented by

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AIM

• Define the acceptability of a product.
• Apply to products placed on the market.
• Also apply to samples taken by Competent Authority at Food Business Operators.
• Set indicative contamination values above which corrective actions are required & also If exceeded, could result in withdrawal or recall.
• Apply to food imported into India.
SIGNIFICANCE

• Food may contain a variety of harmful bacteria which are categorized under Hygiene Indicators (e.g. TPC, Coliform etc.) & safety Indicators (E.coli, Salmonella etc.) based on food safety criteria.

• It is important to understand the nature of food and its safety-regulation criteria if any.

• Integrity of the test sample like sampling activity & transportation plays a major role in the interpretation of test results.


• **Sampling plan includes both 2-class & 3-class** plan for hygiene & safety indicators.

• **Laboratory is not involved in sampling activity.**
DEFINITION

• ‘Microbiological criterion’ means a criterion defining the acceptability of a product, a batch of foodstuffs or a process, based on the absence, presence or number of micro-organisms, and/or on the quantity of these micro-organisms, per unit(s) of mass, volume, area or batch.

A microbiological criterion contains the following components:

✓ The sampling plan (the number of samples of a food that should be taken for testing).
✓ The laboratory method (the method which should be used to test the food).
✓ The stages where the criterion applies.
✓ The corrective action to be taken.

• Two types - Process hygiene criteria criteria & Food safety criteria
There are two widely accepted types of sampling plans as defined by the International Commission for Microbiological Specifications for Food (ICMSF):

- **Two-class plans** are used essentially for pathogens and where a presence/absence test is to be performed (e.g. presence/absence of *Salmonella*). In the case of a presence /absence analysis, \( c = 0 \) and \( m = 0 \). The presence of the target microorganism is unacceptable.
Three-class plans are frequently used to examine for hygiene indicators where enumeration of microbes in a unit volume or mass is possible. Such plans can be used to separate the quality of a lot into acceptable, marginally acceptable and unacceptable.

<table>
<thead>
<tr>
<th>Acceptable</th>
<th>Marginally acceptable</th>
<th>Unacceptable</th>
</tr>
</thead>
<tbody>
<tr>
<td>m</td>
<td>M</td>
<td></td>
</tr>
</tbody>
</table>
KEY ELEMENTS

- **Food category** – Milk products
- **Micro-organisms** – TPC, Coliform count, E.coli, Salmonella/Shigella, *S.aureus* (coagulase+ve), Yeast & mold, *L.monocytogenes*, Spore count (aerobic & anaerobic), *Fecal streptococci*, *Enterobacter sakazakii*, *B.cereus*
- **Sampling plan**: n and c
- **Limits**: m and M
- **Analytical reference method** - are used to assess if food products are in agreement with the microbiological specifications. Further information on use of microbiological methods is available in Annex B of The Food safety and Standards Act, 2006 book.
- **Stages where the criterion applies** – Satisfactory; Unsatisfactory; Potentially hazardous
PERFORMANCE CHARACTERISTICS OF SAMPLING PLAN

- Mathematical/statistical basis for the performance characteristics of the sampling plans
- $n$: number of units which must be examined from the batch/lot of food products;
- $C$: the maximum allowable number of defective sample units giving values between $m$ and $M$
- $m$: is the minimum level accepted and values above $m$ are marginally acceptable.
- $M$: is the maximum level tolerated and values above $M$ are unacceptable.

The above terms are adopted from International Commission on Microbiological Specifications of Foods (ICMSF).
Microbiological limit(s)

☐ Acceptable (< m )
☐ Marginally acceptable (m<value<M)
☐ Unacceptable (>M)
SAMPLING PLAN – MILK POWDER

- **Food category**: Milk Powder
- **Micro-organisms**: TPC, Coliform count & Salmonella
- **Sampling plan**: $n=5$ and $c=0$ & $c=2$
- **Limits**: TPC & Coliform – $m=value$ & $M=value$; **Salmonella** – $m=M$
- **Analytical reference method**:
- **Microbiological criteria & interpretation**
# SAMPLING PLAN – MILK POWDER

<table>
<thead>
<tr>
<th>Microorganism</th>
<th>Analytical unit</th>
<th>n</th>
<th>c</th>
<th>m</th>
<th>M</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total Plate Count</td>
<td>10/50 g</td>
<td>5</td>
<td>2</td>
<td>40000/g</td>
<td>50000/g</td>
</tr>
<tr>
<td>Coliform count</td>
<td>10/50 g</td>
<td>5</td>
<td>2</td>
<td>10/g</td>
<td>50/g</td>
</tr>
<tr>
<td><em>Salmonella</em> spp.</td>
<td>25 g</td>
<td>5</td>
<td>0</td>
<td>Absent/25 g</td>
<td>--</td>
</tr>
</tbody>
</table>
INTERPRETATION OF RESULTS

• Satisfactory – if all the values are \( < m \) or if a maximum of \( c \) units(2) are between \( m \) & \( M \) and the rest of the values are \( < m \)……results are compliance/acceptable.

• Unsatisfactory – if one or more of the values observed are \( > M \) or more than \( c \) units(2) are between \( m \) & \( M \)......results are Non compliance/ Not acceptable (applicable for TPC & hygiene indicators).

• Potentially Hazardous – if one or more of the values observed are \( > M \) or more than \( c \) units(2) are between \( m \) & \( M \)......results are Non compliance/Not acceptable & indicates serious food safety concern (applicable for E.coli, staph, Salmonella, Bacillus, Clostridium & Listeria).
ACTIONS IN CASE OF NON-COMPLIANCE – PATHOGENS

• Immediate remedial action initiated
• Enforcement/prosecution by the food safety authority
• Prevent the affected lot from being released for human consumption
• Ensure recall of product if it has been released for human consumption
• Reject lot at port of entry
• Detail risk assessment study
Actions in case of non-compliance – TPC & Hygiene Indicators

• Indicates poor hygiene or poor handling practice.
• Check on the manufacturing process and controls for prevention of recontamination.
• Determine and correct the root cause of the failure and as appropriate, review and revise monitoring procedures, environmental surveillance and prerequisite programs.
• HACCP/GMP audit by the Food safety authority.
# Case Study – Salmonella in Milk Powder

## Example 1

**Criteria – n=5, c=0, m=0**

All the samples tested for Salmonella must be negative.

*Lab report – Sample 1=Absent/25g*  
Sample 2=Absent/25g  
Sample 3=Absent/25g  
Sample 4=Absent/25g  
Sample 5=Absent/25g

**Interpretation:** The test result is satisfactory and not potentially hazardous.

## Example 2

**Criteria – n=5, c=0, m=0**

All the samples tested for Salmonella must be negative.

*Lab report – Sample 1=Absent/25g*  
Sample 2=Absent/25g  
Sample 3=Absent/25g  
**Sample 4=Present/25g**  
Sample 5=Absent/25g

**Interpretation:** The test result is unsatisfactory and potentially hazardous.
### Example 1

**Criteria** – \( n=5, \ c=2, \ m=10, \ M=50 \)  
All the samples tested for Coliform count must be \(<m\) or maximum 2 values can be between \(m\) & \(M\)  

Lab report –  
Sample 1 = \(<10/g\)  
Sample 2 = \(<10/g\)  
**Sample 3 = 30/g**  
Sample 4 = \(<10/g\)  
**Sample 5 = 40/g**  

Interpretation: The test result is Satisfactory.

### Example 2

**Criteria** – \( n=5, \ c=2, \ m=10, \ M=50 \)  
All the samples tested for Coliform count must be \(<m\) or maximum 2 values can be between \(m\) & \(M\)  

Lab report –  
Sample 1 = \(<10/g\)  
Sample 2 = \(<10/g\)  
Sample 3 = 30/g  
Sample 4 = 20/g  
Sample 5 = 40/g  

Interpretation: The test result is Unsatisfactory.
CONCLUSION

• Many challenges are coming to the food industry as a result of increased food safety regulations.

• Food microbiological testing & result interpretation is definitely an important piece of the food safety, but is certainly not the only factor.

• Everyone involved in the food supply, from farmers to food processors to Food regulatory authorities all have a role to play in food safety.

• Collaboration among all of the different sectors (industry, academia, government, testing labs and consumers), improved technologies and proper education in food safety and also microbiology concepts are all important steps to take in reaching the ultimate goal:

**A Safe Global Food Supply**
Thank you for kind attention...