



MICROBIOLOGICAL CRITERIA AND SAMPLING GUIDELINES

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Microbiological Criteria (Codex)

A microbiological criterion defines the acceptability of a product or a food lot, based on the absence or presence, or number of microorganisms including parasites, and/or quantity of their toxins/metabolites, per unit(s) of mass, volume, area, or lot .

MICROBIOLOGICAL CRITERIA:

Microbiological criteria may be used to define the acceptability of a process, product or food lot.

- The criteria could be the absence, presence, or number of microorganisms and/or the quantity of their toxins/metabolites in samples.
- Microbiological criteria may be used either:
 - By an individual establishment
 - To set national baselines



Any microbiological criteria for food should have following information:

- I. A statement of microorganisms and/or toxins of concern
- II. Lab methods for their detection and quantification
- III. Sampling plan
- IV. Number of samples required to confirm to these limits
- V. Microbiological limits

Microbiological standards for different food items have been fixed where risk assessment has shown that the risk of food-borne illness associated with the consumption of certain foods is relatively high and that a standard could contribute to the management of the probable risk.

Microbiological Criteria - A criterion is fixed on which a decision can be made. A microbiological criterion will stipulate that a type of microorganisms, group of microorganisms or toxin produced by a microorganism must either be not present at all, be present in only a limited number of samples, or be present as less than a specified number or amount in a given quantity of a food or food ingredient.

A microbiological criterion should include the following:

1. A statement describing the identity of the food
2. A statement of the contaminant of concern, i.e. the microorganisms or group of microorganisms and/ or toxin or other agent
3. The analytical method to be used for the detection, enumeration, or quantification of the contaminant of concern
4. The sampling plan
5. The microbiological limits considered appropriate to the food and commensurate with the sampling plan

IMPORTANT TERMS:

- **Microbiological standards:**
Mandatory criteria with legal backing.
- **Microbiological specifications:**
Contractual agreements between a manufacturer and a purchaser to check whether the food are of required quality.
- **Microbiological guidelines:**
Non mandatory criteria usually intended as a guide to GMP.
- **Microbiological limits:**
Recommended by an authority for adoption in specific regions but not incorporated into law.



Why microbiological analysis of foods??

- ❑ One of the most important reasons for analyzing foods from both the consumers and the manufacturers standpoint is to ensure that they are safe.
- ❑ A food may be considered to be unsafe because it contains harmful microorganisms (*e.g.*, Listeria, Salmonella)
- ❑ Foods contaminated with pathogenic microorganisms do not look bad ,taste bad or smell bad.
- ❑ Disease that result from contaminated food are of two types
 - Food borne intoxication
 - Food borne infection
- ❑ Most common symptom associated with food borne illness is diarrhea.

Necessity of Quality control in food testing laboratory

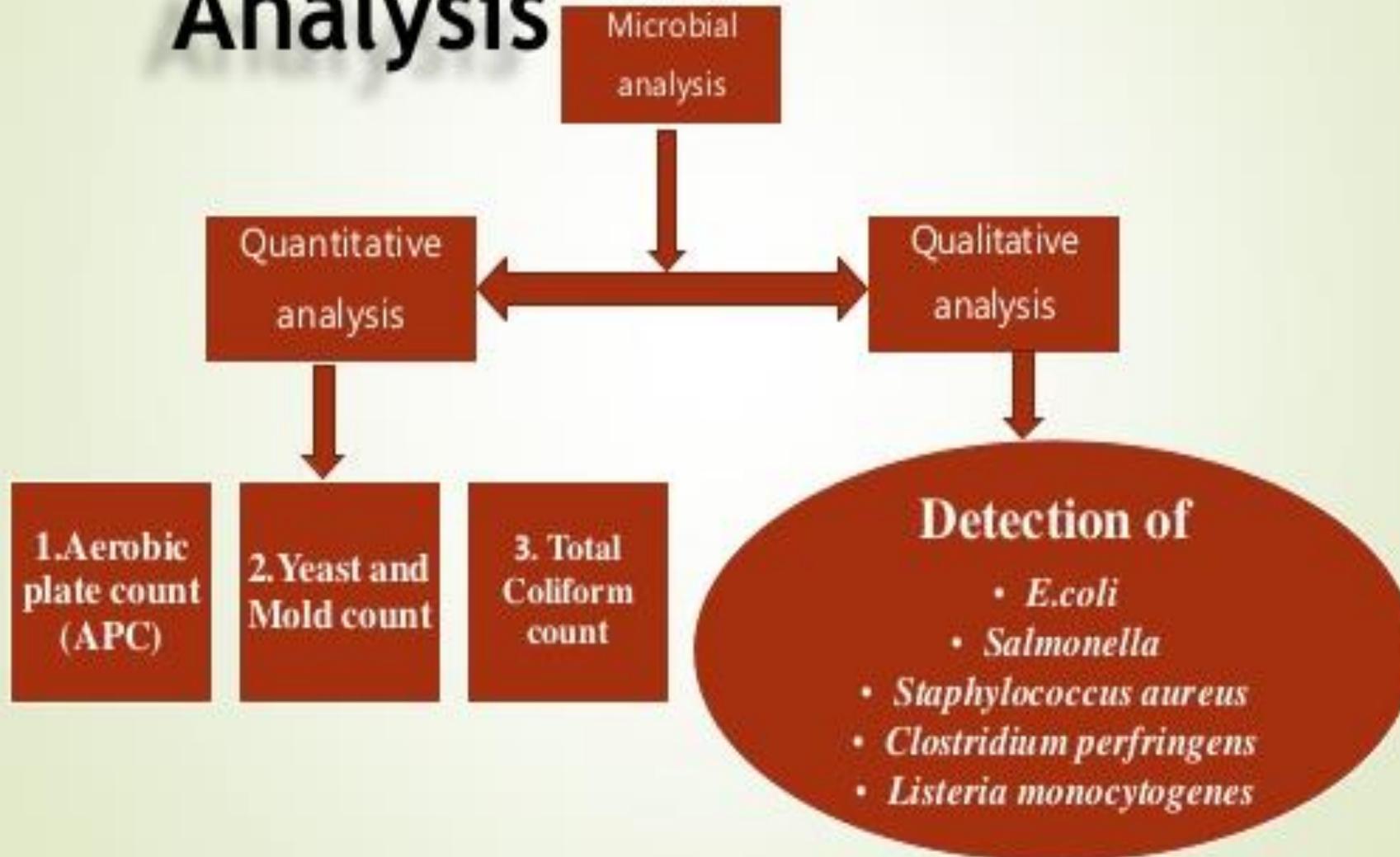
- A specific activity whose purpose is to monitor a discrete laboratory task to ensure that it meets a predefined criterion.
- Safety and wholesomeness are the most important attributes of food quality.
- The lack of quality as it relates to safety and wholesomeness can result in personal injury, sickness or death.
- Food-borne illness is an example of sickness or even death when unsafe foods are produced and eaten.
- The purposes of quality control is To protect the customers from dangers (eg contaminated foods).



Scope

- ▶ To meet out the challenges in the food processing industry, laboratories play a vital role.
- ▶ The scope of testing activity in laboratories encompasses the quality and safety attributes.
- ▶ agriculture and food products, dairy products, oil seeds, oils, spices and condiments, animal products, plantation products and flavours, functional foods, processed foods and beverages

Methods for Microbial Analysis



Field Sample Collection



Laboratory Analysis and Reporting



Data Analysis and Interpretation



Environmental Concentration

Considerations

- Data quality objectives
- Representative samples
- Spore characteristics
- Environmental characteristics
- Human activities
- Sample collection techniques
- Sample transport and storage

- Method recoveries and other challenges
- Data reporting

- Available software
- Selecting appropriate statistical data distributions
- Options for interpreting non-detects

Sample Collection Techniques:

- The FSO/ Authorized Officer must obtain the following information;
 - name of the food;
 - lot number;
 - container size or sizes;
 - product code numbers;
 - labelling information;
 - condition of the lot, i.e., broken packages, evidence of rodent or insect infestation, debris, etc.;



Rejection criteria

- Leaking container
- Unlabelled / wrongly labeled / mismatched samples
- Specimen received in fixative
- Insufficient quantity
- Specimen unsuitable for request
- Prolonged transport
- Dried specimen
- Duplicate specimens on the same day for the same test (except blood, CSF, tissue, sterile body fluids exc. urine)



Microbiological Criteria for Acceptance or Rejection of Sample Lots

1. the food which must comply with the microbiological limits set in relation to that food;
2. the microorganism or group of microorganisms of concern;
3. the number of sample units to be taken and tested;
4. the level of microorganisms considered acceptable, marginally acceptable or critical (depending on the sampling plan specified) and
5. The number of samples that should conform to these limits.

Guidelines for sample size, storage & transport temperature for milk & milk products

S. No	Product	Temperature before and during transport (°C)	Minimum sample size #
1	Pasteurized milk, Cream & Flavored milk	0 to 4	100 ml or g
2	Sterilized & UHT Milk, Cream, Flavored Milk, Evaporated Milk	Ambient, max. 30	100 ml or g
3	Sweetened Condensed Milk (SCM)	0 to 4	100 g
4	Pasteurized Butter	-18 or lower	50 g
5	Dried Products: Milk Powder, Whey, Edible Casein/Ice Cream Mix	Ambient, max. 30	100 g
6	Ice Cream, Frozen Desserts, Milk Lolly, Ice Candy	-18 or lower	100 g
7	Processed Cheese & Cheese Spreads	4 to 8	100 g
8	Other Types Of Cheeses	4 to 8	100 g
9	Yoghurt, Dahi, Chakka & Srikhand	0 to 4	100 g
10	Paneer	0 to 4	100 g
11	Khoa	0 to 4	100 g

a large sample size may be necessary according to the tests required and the type of product

Reference test procedure for different microbiological parameters proposed in draft standards

Microbiological requirements		Reference test procedure	Reference
Total Plate Counts (TPC)		General guidance for the enumeration of microorganisms – Colony Count Technique at 30°C	IS 5402; 2002/ ISO:4833:1991
Coliform Count		General guidance for the enumeration of Coliforms	IS: 5401: PART-1; 2002/ ISO: 4831:1991
<i>E. coli</i>		Detection of bacteria responsible for food poisoning part – isolation, identification and enumeration of <i>E. coli</i>	IS:5887 (PART I) -1976 (Reaffirmed as 1995)
<i>Salmonella</i>		Methods for detection of bacteria responsible for food poisoning	IS:5887:PART-3; 1999/ ISO: 6579; 1993
<i>S. aureus</i>		Methods for detection of bacteria responsible for food poisoning – part - 8 horizontal method for enumeration of coagulase - positive staphylococci (<i>S. aureus</i> and other species) - section 1 technique using Baird – Parker agar medium)	IS:5887 :PART 8; SEC 1 :2002 /ISO 6888- 1:1999
Yeast & Mould Count		Method for Yeast and Mould Count of foodstuff and animal feeds.	IS:5403-1999
Spore Count	Aerobic	Specification for sterilized milk	IS:4238-1967 (Reaffirmed 1999)
	Anaerobic	Methods for detection of bacteria responsible for food poisoning part-IV Isolation and identification of <i>C. welchii</i> , <i>C. botulinum</i> and <i>B. cereus</i> and enumeration of <i>C. welchii</i> , <i>C. botulinum</i> and <i>B. cereus</i>	IS:5887 PART IV; 1976
<i>Listeria monocytogenes</i>		Microbiology of Food and Animal Feeding Stuffs –Horizontal Method for the Detection and Enumeration of <i>Listeria monocytogenes part-1 detection method</i>	IS: 14988:PART1; 2001 ISO: 11290-1; 1996

Microbiological Criteria for milk and cream

SI. No.	Requirements	Sampling Plan	Pasteurized milk/ Pasteurized cream
1	Total plate count	m	30,000/g
		M	50,000/g
2	Coliform Count ²	m	< 10 /g
		M	
3	<i>E.coli</i> ³	M	Absent/ g
4	<i>Salmonella</i> ⁴	M	Absent/ 25g
5	<i>Staphylococcus aureus</i> ⁵ (coagulase positive)	m	< 10/ g
		M	
6	Yeast and mould count ⁶	m	-
		M	-
7	Spore Count:		-
	(a) Aerobic ^{7a} (<i>B. cereus</i>)	m	-
		M	-
	(b) Anaerobic ^{7b} (<i>Clostridium Perfringens</i>)	m	-
		M	-
8	<i>Listeria Monocytogenes</i> ⁸	M	Absent/g
Sampling Guidelines ⁹		n ¹⁻⁸	5
		c	20
			0 ^{1-5,8}
		Storage & transport	0 to 4°C
Sample size	100ml or g		

Satisfactory: if the test values of m or M or both applicable within the sampling plan are conforming the specified limits, the microbiological quality of products considered satisfactory and no action is required.

m = Represents an acceptable level and values above it are marginally acceptable in terms of the sampling plan
M= A microbiological criterion which indicate unsatisfactory or potentially hazardous quality. Values above M are unacceptable in terms of the sampling plan and detection of one or more samples exceeding this level would be cause for rejection of the lot and will attract prosecution by the concerned food safety authorities.

Microbiological criteria for pasteurized butter

SI. No.	Requirements	Sampling Plan	Pasteurized Butter
1	Total plate count	m	10,000/g
		M	50,000/g
2	Coliform Count²	m	10/g
		M	50/g
3	<i>E.coli</i> ³	M	Absent/g
4	<i>Salmonella</i> ⁴	M	Absent /25g
5	<i>Staphylococcus aureus</i> ⁵ (coagulase positive)	m	10/g
		M	50/g
6	Yeast and mould count ⁶	m	20/g
		M	50/g
7	Spore Count: (a) Aerobic ^{7a} (<i>B. cereus</i>)	m	-
		M	-
	(b) Anaerobic ^{7b} (<i>Clostridium Perfringens</i>)	m	-
		M	-
8	<i>Listeria Monocytogenes</i> ⁸	M	Absent/g
Sampling Guidelines ⁹		n ¹⁻⁸	5
		c	2 ^{1-2, 5-6}
			0 ^{3,4,8}
		Storage & transport	-18°C
Sample size	100ml or g		

Microbiological criteria for sweetened condensed milk

Sl. No.	Requirements	Sampling Plan	Dried milks
1	Total plate count	m	40,000/g
		M	50,000/g
2	Coliform Count ²	m	10/g
		M	50/g
3	<i>E. coli</i> ³	M	Absent/g
4	<i>Salmonella</i> ⁴	M	Absent /25g
5	<i>Staphylococcus aureus</i> ⁵ (coagulase positive)	m	-
		M	Less than 10/g
6	Yeast and mould count ⁶	m	-
		M	-
7	Spore Count: (a) Aerobic ^{7a} (<i>B. cereus</i>)	m	100/g
		M	1000/g
	(b) Anaerobic ^{7b} (<i>Clostridium Perfringens</i>)	m	10/g
		M	100/g
8	<i>Listeria Monocytogenes</i> ⁸	M	Absent/g
Sampling Guidelines ⁹		n ¹⁻⁸	5
		c	2 ^{1&3}
			0 ^{2-4,6,7b,8}
		Storage & transport	0-4°C
		Sample size	100ml or g

Microbiological criteria for dried milks

Sl. No.	Requirements	Sampling Plan	Dried milks
1	Total plate count	m	40,000/g
		M	50,000/g
2	Coliform Count ²	m	10/g
		M	50/g
3	<i>E. coli</i> ³	M	Absent/g
4	<i>Salmonella</i> ⁴	M	Absent /25g
5	<i>Staphylococcus aureus</i> ⁵ (coagulase positive)	m	-
		M	Less than 10/g
6	Yeast and mould count ⁶	m	-
		M	-
7	Spore Count: (a) Aerobic ^{7a} (<i>B. cereus</i>)	m	100/g
		M	1000/g
	(b) Anaerobic ^{7b} (<i>Clostridium Perfringens</i>)	m	10/g
		M	100/g
8	<i>Listeria Monocytogenes</i> ⁸	M	Absent/g
Sampling Guidelines ⁹		n ¹⁻⁸	5
		c	2 ^{1&3}
			0 ^{2-4,6,7b,8}
		Storage & transport	0-4°C
		Sample size	100ml or g

Microbial pathogens of major concern

Microbial pathogens of concern with dried milk and infant formulae are similar mainly *B. cereus*, *C. perfringens*, *E. sakazakii*, *L. monocytogenes*, *Salmonella*, *Shigella* and *S. aureus*. However, control over the microbiological status of these products is essential because of the vulnerable status of infants. Global surveys of infant formulas have indicated the presence of *B. cereus* and *E. sakazakii*. While *Salmonella* is rarely found in surveys of powdered infant formula, low-level contamination of powdered infant formula with *Salmonella* has been epidemiologically and microbiologically associated with infections in infants. Illness has also been attributed to *S. aureus* contamination and to abuse of reconstituted infant powdered milk.



WHAT IS SPOILAGE?

CHANGES

Not microbiological in origin

Result of:

- Insect damage.
- Drying out.
- Discolouration.
- Staling.
- Rancidity.

Microbial activity (most food spoilage)

- Microbial growth in the form of surface slime or colonies.
- Degradation of structural components of food can cause loss of texture.
- Chemical products of microbial metabolism, gas, pigment, polysaccharides, off-odours and flavours (most common).