

FE 305 Experiment 4

Bacteriological Examination of Yogurt

Contents

- Total Aerobic Mesophilic Bacteria Count
- Mold and Yeast Count
- Total Lactic Acid Bacteria Count

Experiment No from Book: **33-35**

Samples needed for this experiment

- **Home made yogurt**
- **Pasteurized yogurt**

Purpose and Importance

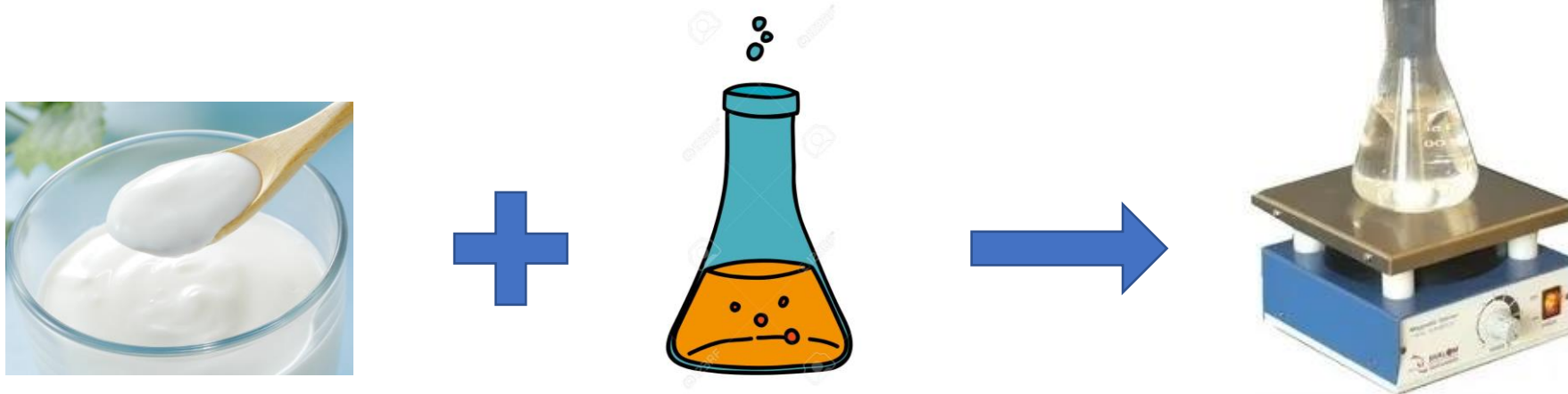
- Yogurt is a semi solid fermented milk product which is very popular in Turkey.
- Lactic acid bacteria in milk with equal ratio mixed starts the fermentation and produces yogurt with a lactic acid fermentation.
 - *Streptococcus thermophilus*
 - *Lactobacillus bulgaricus*

Symbiotic growth of these two bacteria produce lactic acid reduce pH and cause protein coagulation in milk to produce yogurt. Fermentation ends at pH \approx 4.0

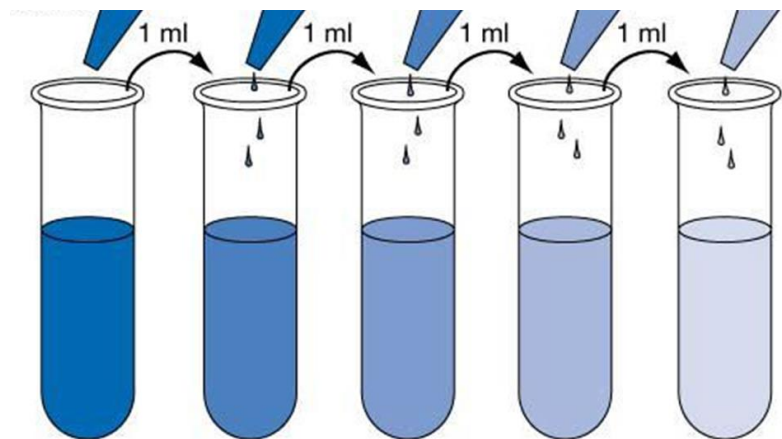
Main spoilage of yogurt is caused by mold growth on the surface.

Sampling and Preparation

- Because yogurt is a semi-solid food product, sample and dilution preparation needs additional attention.



- 25 g yogurt + 225 ml 0.1% sterile peptone water = 10^{-1} diluted sample



Serial dilution is applied starting from 10^{-1} diluted sample

Tests and Analysis

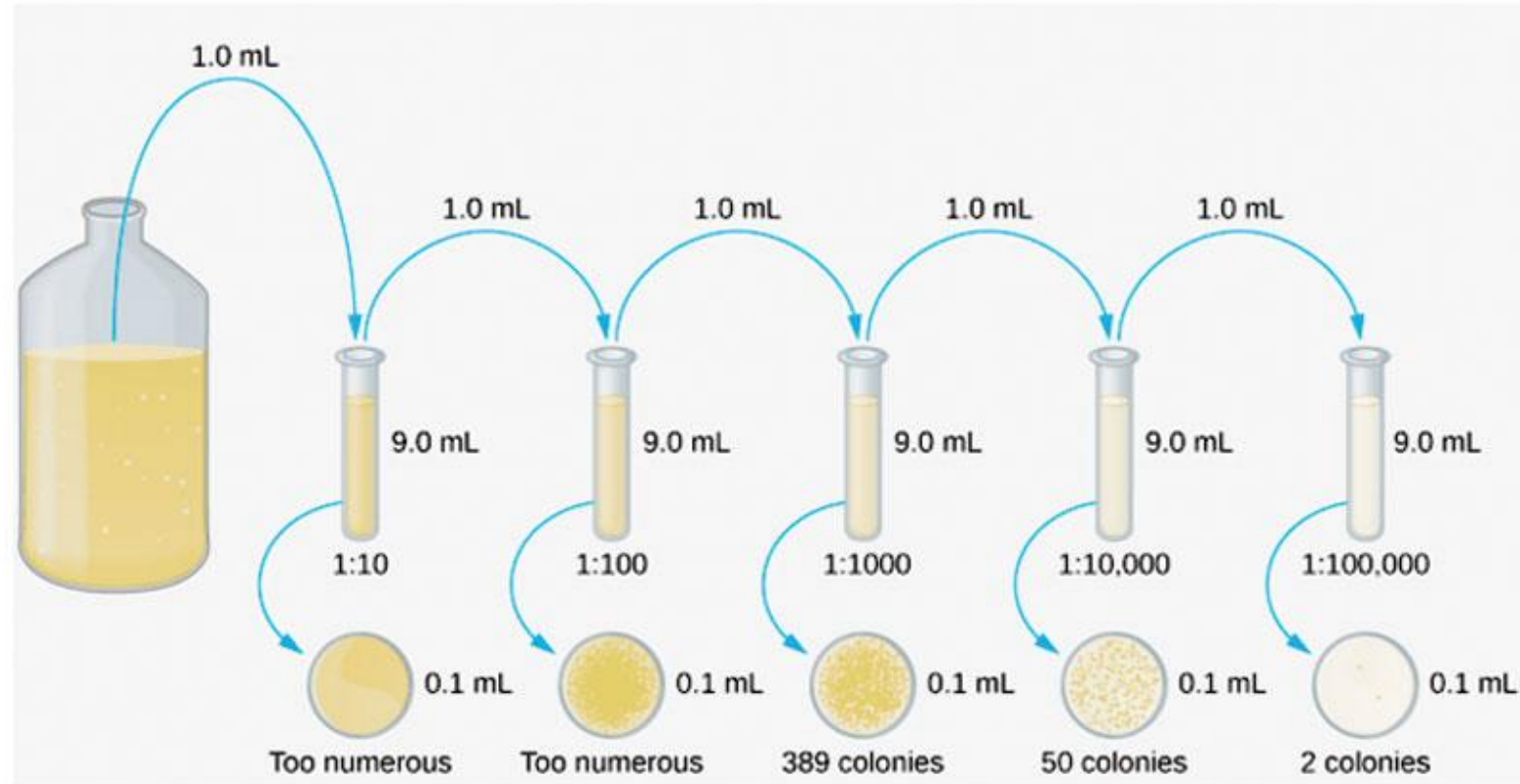
- Total Count
 - Media: PCA
 - Method: Spread Plate
 - Incubation: 37 C for 24 hours
- Mold and Yeast Count
 - Media: PDA
 - Method: Spread Plate
 - Incubation: 25 C for 3-5 days
- Lactic acid bacteria count
 - Media: MRSA
 - Method: Spread Plate
 - Incubation: 42 C for 2 days

Material and Methods

- Yogurt sample prepared earlier as 10^{-1} diluted sample and dilutions.
- Spreader
- Sterile pipette
- Incubator
- Alcohol
- Bunsen burner
- Sterile PCA, PDA and MRSA

Procedure

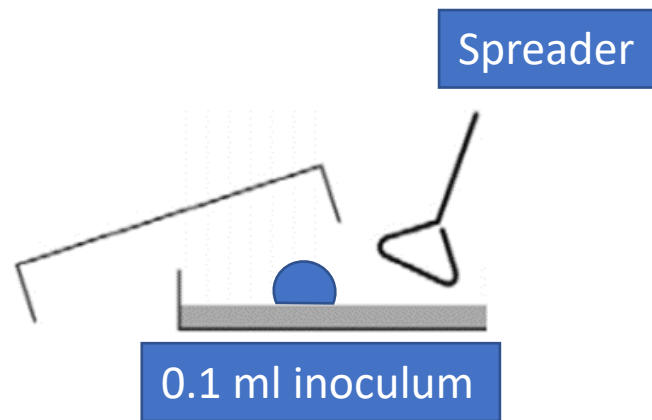
- Take 0.1 ml of sample from each dilution by using sterile pipette at Aseptic Conditions.
- Place the sample on PCA petri plate near flame.



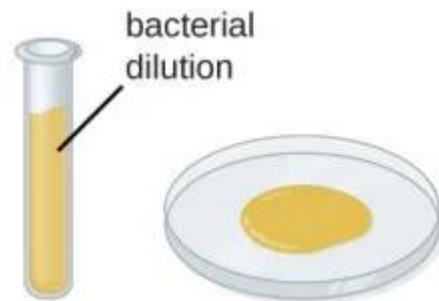
	10^{-1}	10^{-2}	10^{-3}	10^{-4}	10^{-5}
Total Count	PCA	PCA	PCA	PCA	PCA
Mold and Yeast	PDA	PDA	PDA	PDA	PDA
Total Lactic Acid Bacteria	MRSA	MRSA	MRSA	MRSA	MRSA

Procedure

- Sterilize spreader with dipping in alcohol and passing through flame.
- Spread the sample on petri plate by using spreader.



1 Sample (0.1 mL) poured onto solid medium



Spread Plate Method

2 Spread sample evenly over the surface

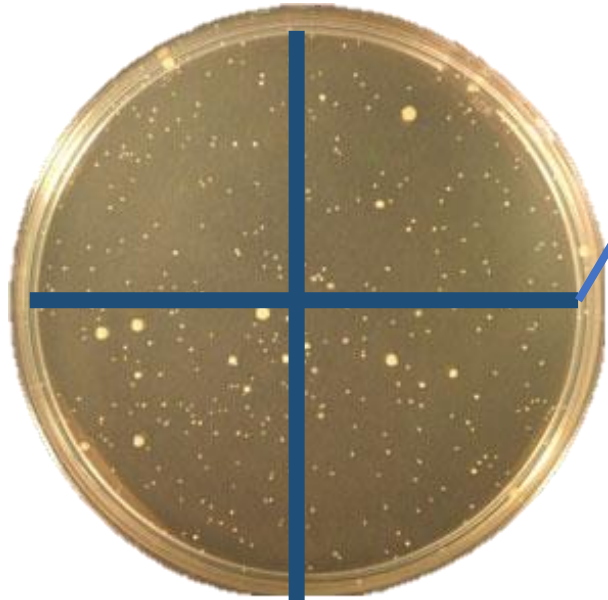


3 Plate incubated until bacterial colonies grow on the surface of the medium



Results

- Count formed colonies on every petri plate for each dilution and record results on the given table below.



- You can divide petri into equal parts to count easily.
- Ex: For Home made yogurt
 - Non → Too nuber to count → TNTC
 - -1 → 546
 - -2 → 186
 - -3 → 34
 - -4 → 0

Sample names	Dilutions					
	non	10^{-1}	10^{-2}	10^{-3}	10^{-4}	10^{-5}
Home made	TNTC	546	186	34	0	0
Pasteurized						

Calculation

- # of microorganisms / g of yogurt = $\frac{\text{Count of microorganisms in one petri}}{\text{inoculum amount in one petri}} \times \text{Dilution factor}$
- Ex: For home made yogurt take only counts between $30 < x < 300$ colonies into calculation.

- Non: TNTC

- -1: 546 → No calculation above 300

- -2: 186

- -3: 34

- -4: 0 → No calculation below 30

$$\frac{186}{0.1 \text{ ml}} \times \frac{1}{10^{-2}}$$

$$\frac{34}{0.1 \text{ ml}} \times \frac{1}{10^{-3}}$$

- Take average of these two results

- # of microorganisms / g or ml of water = $\frac{186000 + 340000}{2} = 263000$ microorganism / ml of water

Same Calculation for each Mold and Yeast and Lactic acid bacteria count

Evaluation of Results

- **Total count** → Calculate and record # of microorganisms / g of yogurt
- **Mold and Yeast count** → Calculate and record # of molds / g of yogurt
- **Lactic Acid Bacteria** → Calculate and record # of LAB / g of yogurt

Total count gives information about general microbial conditions of yogurt.

Mold and Yeast count is important for spoilage and shelf life of yogurt

Lactic acid bacteria count is important for fermentation and production conditions of yogurt.