Lecture 4

Microbial Culture Media

What is culture medium?

Culture Medium: a special medium used in microbiological laboratories for the growth, isolation and identification of microorganisms. A culture medium is composed of different nutrients (carbohydrate, lipids, amino acids, vitamins as well as inorganic compounds).

The food material or substances required for growing microorganisms in vitro (outside the body) is called **culture medium**, also provides a surface and the necessary moisture and pH to support microbial growth

Types of Culture Media

The culture media are classified in many different ways:

- Physical States of Media

1- Liquid media:

Liquid media does not contain any solidifying agents. Liquid media are sometimes referred as "broths" (e.g. Nutrient Broth, MacConky Broth).

Culturing in liquid medium can be used to obtain viable count (dilution methods).

- **media without any agar or gelatin**
- **Commonly used for: * General culture / * Biochemical test / * Susceptibility test**

2- Solid media:

Containing 1-2% solidifying agent.

Agar is the most commonly used solidifying agent, it is a polysaccharide derived from red algae, Agar also cannot metabolize by microbes. (e.g. nutrient Agar).

- **Commonly used for: * Agar plate/ * Isolation of bacterial colonies**

3- Semi solid media:

Containing 0.2 - 0.5% Solidifying agent (agar or gelatin) , this media are useful in demonstrating bacterial motility and separating motile from non-motile strains.

• Commonly used for: * Microaerophilic culture / * Motility test

What is agar?

A substance extracted from some red algae, especially those species belonging to the genus (Gelidium)

Agar is characterized by the following features

1- It melts within the boiling point of water and solidifies when cooled to 42°C.

2- Small amounts of it help in solidifying large quantities of media, if 1.5-2 grams of it is sufficient solidify 100 ml of medium.

3- This substance is not consumed or degraded by most microorganisms

-Based on nutritional composition, culture media can be classified as:

1. Simple media:

Media that provide the minimal requirements for microbes to (e.g. Peptone water, nutrient agar).

2- Enriched

• Support the growth of fastidious bacteria

• contains complex organic substances such as blood, serum, hemoglobin, or special growth factors specific vitamins, amino acids (e.g. blood agar)

3. Chemically defined media (synthetic Media): These media are use in studying the nutritional requirements of microorganisms or in studying their metabolic activities.

-Based on functional use culture media can be classified into:

1. Selective Media:

• Allow the growth of desired bacteria while inhibitory other types

• Contain inhibitory agents like antibiotic, bile salts.

• Examples :

➢ MacConkey agar [selective for gram negative]

➢ Mannitol salt agar (MSA) contains a high concentration of NaCl (7.5%) selective for *Staphylococcus*.

2. Differential media (called Indicator Media)

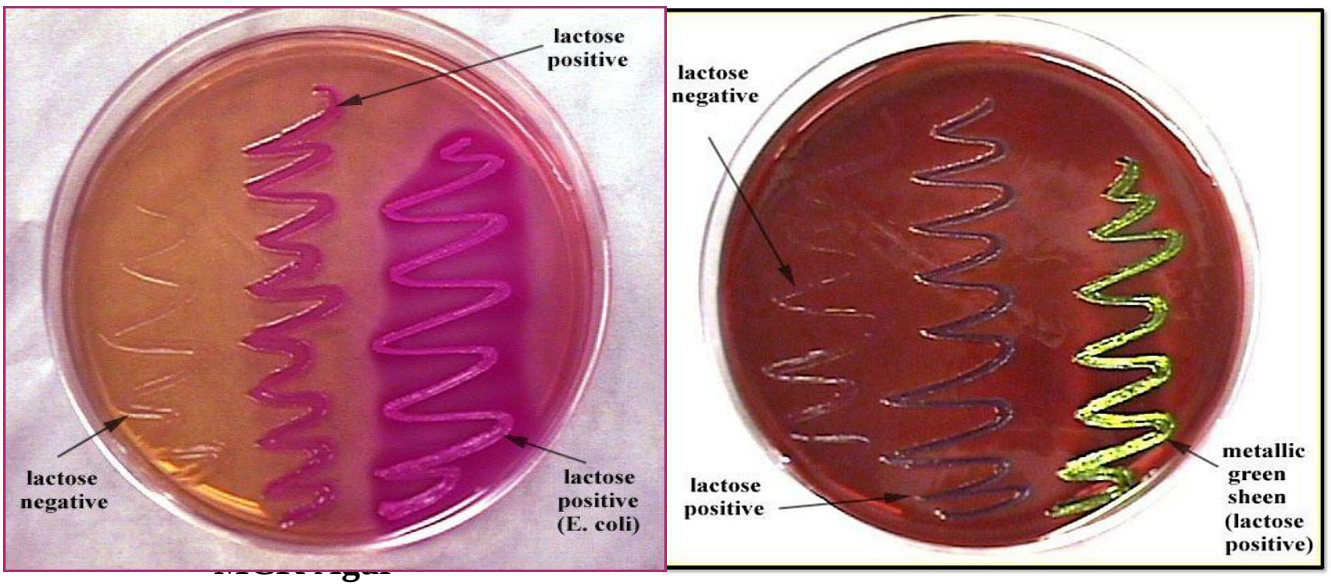
• allow multiple bacteria to grow but with distinguishable colonial characteristic.

• Allow for preliminary characterization of bacteria.

In which more than one microorganism grows, However, one of these types is distinctive, so it can be differentiated from the other types in terms of: Color and shape of colonies

- For example:

- Eosin methylene blue (EMB), which is differential for lactose and sucrose fermentation.
- MacConkey (MCK), which is differential for lactose fermentation



3- Assay Media:

These are used to estimate the amount of antibiotics or -Vitamins produced from microorganisms or present in the samples under study

Colony is a group of bacteria, fungi, and other microorganisms grown on a solid agar medium. The cells plated on this medium grow to form a mass, which can then be duplicated for further use in the lab, which vary in terms of size, shape, and color depending on the type of microorganisms that make it up, each colony usually originates from a cell at least one.

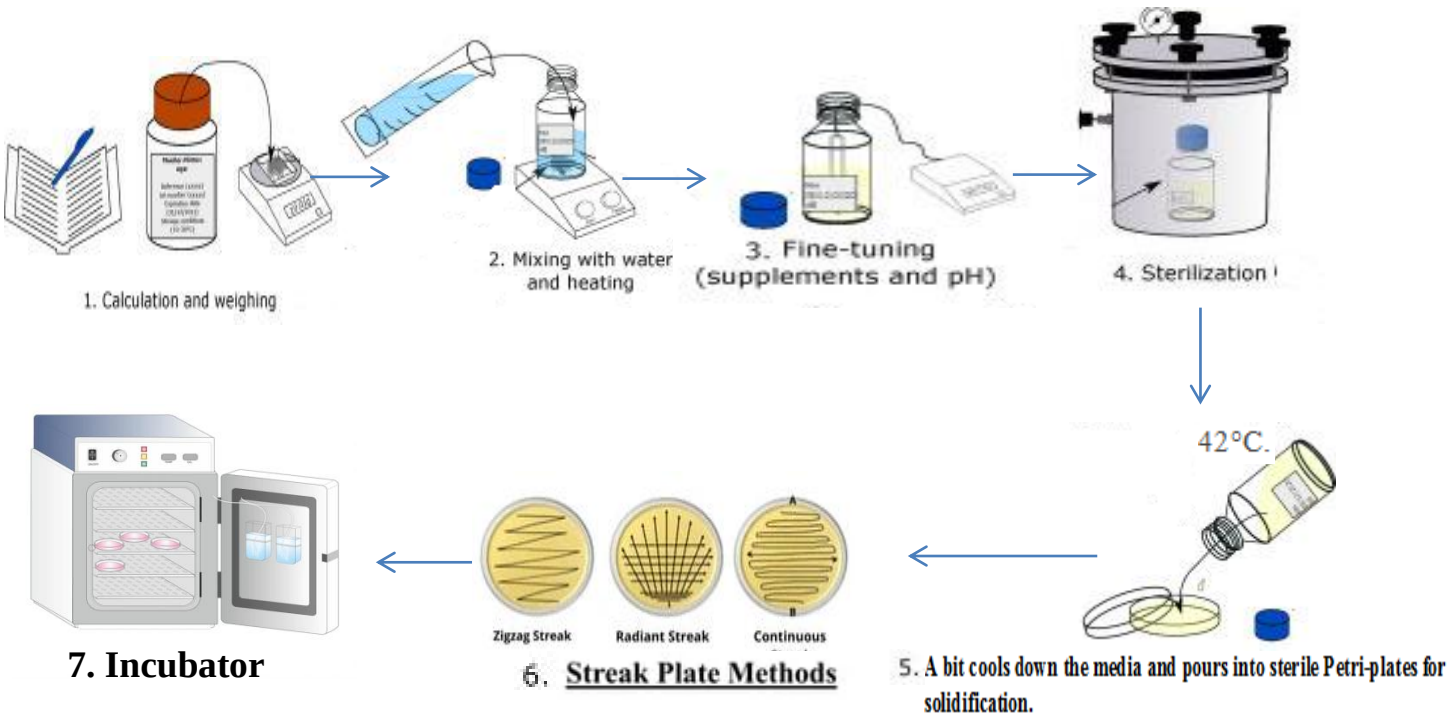
Preparation of microbial media

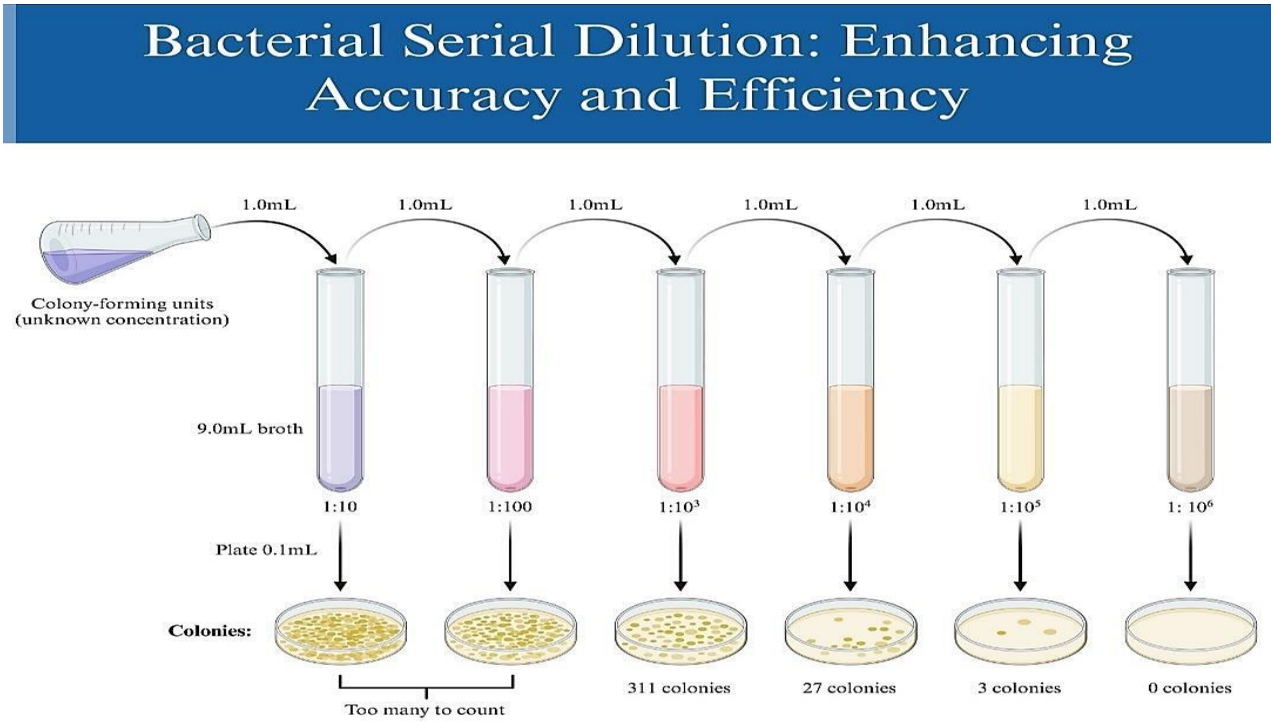
The basic requirements in microbial media preparation:.

The procedure:

1. **Weigh the amount of ingredients powder on weighing machine.**
2. **Dissolve the ingredients in distilled water.**
3. **Adjust PH of the medium if needed.**
4. **Add agar and boiled it to dissolve.**
5. **Pour the media into flask.**
6. **Autoclave the media when ingredients fully dissolve.**
7. **Sterilization is done in autoclave to prevent from contamination, at 121°C for 15 min at 15lbs.**
8. **A bit cools down the media and pours into sterile Petri-plates for solidification.**
9. **Then sample is ready to spread(spreader) / streak**

10. (Inoculation loop) on the medium for identification or isolation of microbes





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