

To review on isolation and preservation of pure cultural media

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ABSTRACT

A culture media is a special medium used in micro-biological laboratories to grow different kinds of micro-organisms. The growth of culture medium is composed of different nutrients that are easily essential for the microbial growth.

I. INTRODUCTION

A micro-biological culture medium is a substance that encourages the growth as well as support, and survival of micro-organisms. Culture media contains nutrients, growth promoting factors, energy sources, buffer salts, minerals, metals, and gelling agents (for solid media)

Principal of cultural media

Culture media are used for the quality control tests of non-sterile raw materials and finished products and microbial contamination (sterility) tests in applications such as hygiene monitoring as well as sterilization process. validation and determination of the effectiveness of preservatives and antimicrobial agents.

What is Culture Media.

The cultural media is a source of nutrients to support the grow the micro-organisms in-vitro. The media helps in the growth and counting the microbial cells, selection of the micro-organisms, and survival of micro-organisms. The culture medium can be available in liquid or gel.

Common used an ingredients of culture media

- 1) **Peptone**- source of carbon and nitrogen.
- 2) **Beef extract**- source of amino acid, vitamins, minerals.
- 3) **Yeast extract**- source of vitamin, carbon, nitrogen.
- 4) **Distilled water**
- 5) **Agar**- solidifying agent.

How are to prepare culture media?

- 1) Weight of the ingredients powder on weighing balance machine.
- 2) Dissolve in the ingredients in the distilled water .
- 3) Adjust PH of the medium..

4) Add agar and boiled.

5) Pour the cultural media into conical flask.

6) open Autoclave the media when ingredients fully dissolve.

7) Sterilization process is done again autoclave to prevent from contamination, at 121°C for the 15 minutes

8) After the autoclave placed the media into conical flask in the laminar air flow.

Sterilized the laminar air flow with 70% of the alcohol.

9) A bit cools down the media & pours into sterilized .Petri-plates for solidification

10) Then the sample is ready to spread . (Inoculation loop) on the medium for identification or isolation of the microbes.

Sealed the Petri-plates with paraffin, label in the petri plate.

11) Keep the petri plate inverted in incubator at 37°C for 24hours.

(Observe the result next day colonies formation is visible on the media)



Types of culture media based on consist of physical state

1. Solid medium
2. Semi-solid medium
3. Liquid medium

1. Solid media

Principle of Solid Media

It is for the isolation of the bacteria as a pure culture on a solid medium.

(Robert Koch) realized the use of solid media.

Agar is highly used to hardening the media at 1.5-2.0% concentration. Solid media allow to the growth of bacteria as colonies by streaking on the medium. It is solidified at the 37°C.

Agar is an un-branched polysaccharide extracted from red algae species. like the Gelidium. Colonies identification is done by the medium.

Examples of Solid Media

Nutrient agar, MacConkey agar, Blood agar, Chocolate agar.

2. Semi-solid media

Principle of Semi-solid media

This media shows the motility of bacteria and the cultivation of micro-aerophilic bacteria. This media has been agar at a concentration of 0.5% or less. It has a jelly consistency.

Examples of Semi-solid media

Stuart's and Amies media.

3. Liquid media

Principle of Liquid media

This media shows the growth of the large number of bacteria.

It is called Broth that allows bacteria to grow uniformly with turbidity. The growth occurs at 37°C in an incubator for 24hrs.

Liquid media don't have the addition of agar. it is for fermentation studies.

Examples of Liquid media

Nutrient broth,



Types of culture media based on chemical composition

There are seven routine laboratory media.

- 1) Basal media
- 2) Enriched media
- 3) Selective media
- 4) Enrichment media
- 5) Indicator media or differential media
- 6) Transport media
- 7) Storage media

1. Basal media

Basal media is simple. it enhances the growth of many micro-organisms. It's a routinely use medium in the laboratory, having Carbon and Nitrogen. This media allows the growth of non-fastidious bacteria without any enrichment source; used for sub-culturing. It's a non-selective medium. Staphylococcus and Enterobacteriaceae grow in this media.

Examples of Basal media

Nutrient Agar, Peptone water.

2. Enriched media

This media requires the addition of other substances like blood, egg, as well as serum. An enriched media allows the growth of diverse micro-organisms but inhibits other and fastidious microbes grow as they require nutrients like vitamins and growth-promoting substances.

Examples

Blood agar, Chocolate agar

3. Selective media

This selective media show the growth of selective. microbes or desired micro-organisms and inhibits the growth of unwanted microbes. The

inhibition occurs by adding bile salts, antibiotics, dyes, PH adjustments. Media is agar-based; any media is possible to transform into selective by adding

4. Enrichment media

It is a liquid medium, which also permits the growth of desired bacteria at a low density. The media provides an environment and conditions as selective media and inhibits un-wanted bacteria from growing. It is used for the isolation of the soil and fecal micro-organisms.

Examples of Enrichment

Salmonella Typhi.

5. Indicator or differential media

This media shows visible changes due to the presence of an indicator. It different bacteria based on colony color grows on the same plate. bio-chemical characteristics show organisms growth with chemical indicators like neutral red, phenol red, methylene blue.

Examples of Indicator or differential media

Mannitol salt agar (mannitol fermentation shows yellow color colonies);

6. Transport media

The media transport specimens after collection to control the overgrowth of micro-organisms. For the cultivation, this media act as temporary storage. It also maintains the viability of pathogens in specimen and prevents them from drying.

Examples of Transport media

Stuart's transport medium (lacks carbon, nitrogen, growth factors)

7. Storage media

It is maintains the longevity of bacterial culture.

Examples are cooked meat broth, NA egg saline.

Types of culture media based on oxygen requirement

Micro-organisms have different requirements for growth depending on oxygen requirements.

1. Aerobic media

In this media, it is easy to cultivate microbes, on the solid media, the growth occurs by keeping of culture in the incubator. It shows the growth of non-fastidious micro-organisms.

Examples of aerobic media are- liquid media, solid media

Peptone water- 1%peptone + 0.5% NaCl +100ml water.

Nutrient agar- nutrient broth +2% agar.

2. Anaerobic media

The media cultivates anaerobic bacteria at less oxygen, reducing oxidation-reduction potential. due to Anaerobic media contains extra nutrients like vitamin K, hemin, and oxygen that get reduced by a physical or chemical process. The medium is boil in a water bath to force out dissolved oxygen and packed with sterilized in paraffin.

Examples of Anaerobic media

RCM (Robertson cooked meat) isolation for Clostridium sp.

Application of culture media

- 1)To culture microbes.
- 2)To the identify the cause of infection
- 3)To identify characteristics of micro-organisms.
- 4)To the isolate pure culture.
- 5)To the stored the culture stock.
- 6)The observed the bio-chemical reactions.
- 7)To the test of microbial contamination in any sample.
- 8)To the check of anti-microbial agents and preservatives effect.
- 9)The observe microbe colony type, its color, shape, cause.
- 10)To differentiate between different colonies.
- 11)To create the antigens for laboratory used .
- 12)To estimate the viable count.
- 13) test of antibiotic sensitivity.

Uses of cultural media

Culture media are highly used for quality control tests of non-sterile raw materials and finished products as well as for microbial contamination tests in applications such as sterilization process validation & determination of the effectiveness of preservatives and anti-microbial agents.

II. CONCLUSION.

Culture media supported the growth of the bacterial cells and micro-organisms in labs by fulfilling their need for nutrients and minerals. However, different organisms have different nutrient living conditions.



REFRANCE -

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