2nd Professional Year

Preparation of media for cultivation of aerobic and anaerobic bacteria

Dr. Pankaj Kumar
Assistant Professor
Veterinary Microbiology

Growing bacteria in Lab

- The growth of bacteria in laboratory is based on an adequate supply of suitable nutrient material
- Along with Suitable physical conditions.
- The microorganisms need a number of elements for growth, energy source and physiological functions

Agar –Solid medium

Broth-Liquid media

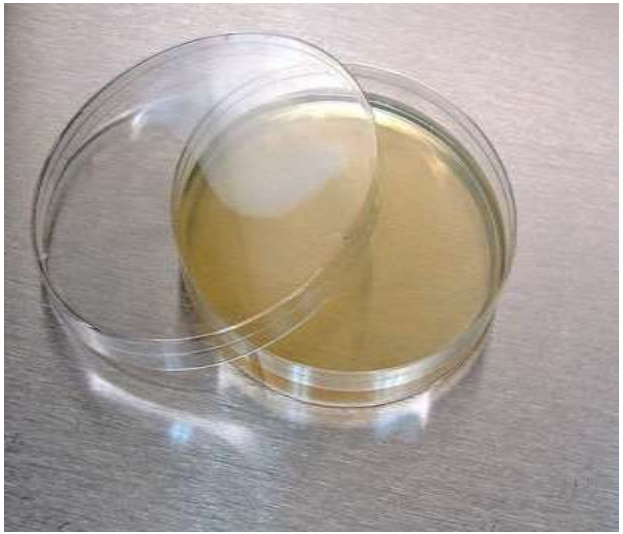
Solid media ??

- **Liquid media** are used for - propagation of microbes, fermentation studies, and various other tests

- **Solid media** has certain advantages:
  - Allows isolation of bacteria
  - It is possible to get Colonies

- The ingredients could be - pure chemical compounds to complex materials
- Generally *animal and plant tissue extracts* are used
- The more commonly used ingredients of bacteriological media are:

Meat extract, Peptone and Salt



Agar is generally used as a solidifying agent

(*Nutritional value of Agar is nil*)

Different types of bacteria media:

Basic Nutrient Media (Simple media):

- They contain only basic nutrients

  eg: Peptone broth
- Any selective or enriched ingredients – not added
- Certain pathogenic organisms having more exacting nutrition requirement

  eg: Streptococci

Peptone broth:

- Peptone 10g
- Sodium chloride 5 g
- Dist. Water 1 lit
- pH 7.6

Use: For preparing nutrient broth and nutrient agar, for Indole test, basal medium for preparation of sugar media, for inoculum preparation of bacterial culture

Nutrient broth:

- Peptone 10g
- Beef extract 10g
- Sodium chloride 5 g
- Dist. Water 1 lit
- pH 7.6

Use: For preparing nutrient agar or blood agar, to prepare liquid inoculums, to study the growth characteristics of bacteria

Nutrient agar:

- Peptone 10g
- **Beef extract** **10g**
- Sodium chloride 5 g
- Dist. Water 1 lit
- Agar 10-20gm
- pH 7.6

Use: To study the growth characteristics of bacteria

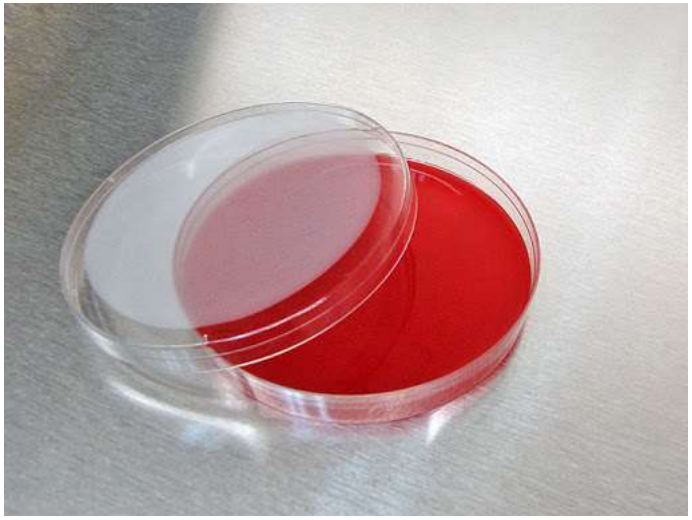
Enriched media:

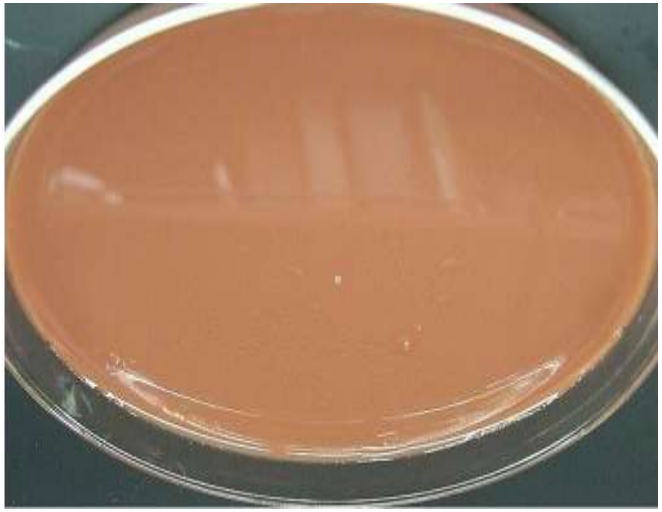
- Contains- blood, serum
- Facilitate better growth

Eg- Blood agar, Serum agar, Serum glucose agar, Glucose broth

Blood agar: Prepared by adding 5-10 % sterile defibrinated blood in melted nutrient agar

- at a temperature of 55^0 C temperature





Enrichment Media:

- Favours the growth of a particular organism

- Eg:
  Tetrathionate broth,
  Selenite F broth,
  Rappaport broth
- Enrichment media are liquid in consistency while selective media are solid.

used for isolation of Salmonella

Differential media:

Mac Conkey agar:

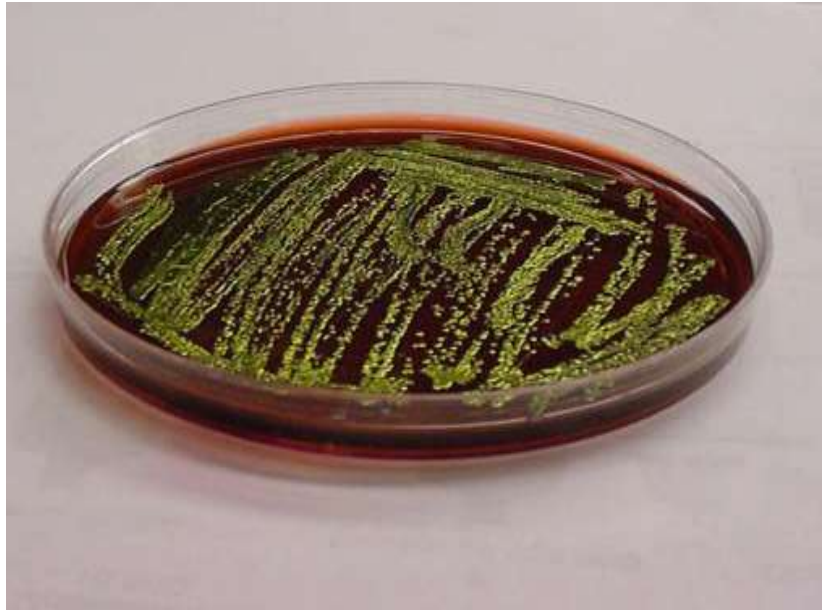
• Peptone	20g
• Sodium chloride	5g
• Bile salts	5g
• Lactose	10g
• 2 % neutral red	5ml
• Agar	20-25g
• Distilled water	1 lit
• pH.	7.4

McConkeys Agar

Brilliant green agar (BGA): For selective isolation of Salmonella other than *S. typhi*

Eosin Methylene Blue agar (EMB): For selective isolation of *E. coli.*

Characteristic metallic sheen colonies of *E. coli* on EMB



Robertson's cooked meat medium(RCM):

- Peptone 10g
- Beef extract 10g
- Sodium chloride 5 g
- Dist. Water 1 lit
- pH 7.6

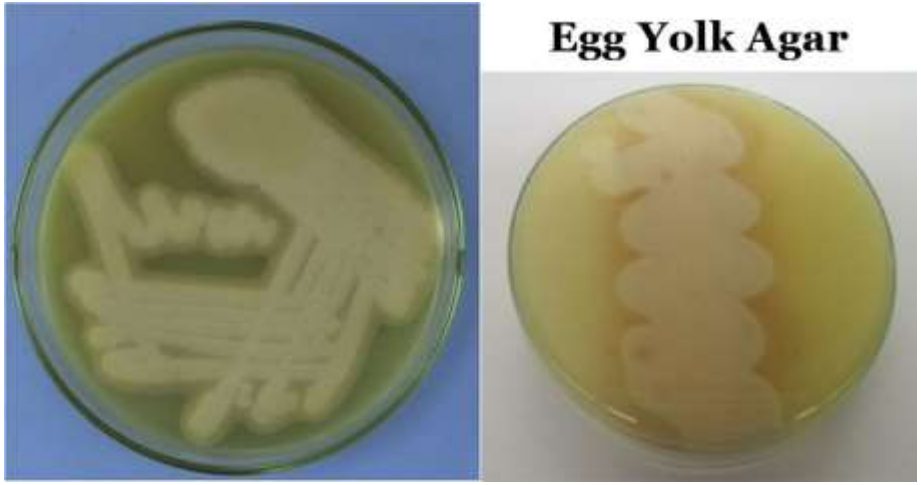
- Add pieces of heart – Nutrient broth

Use: For cultivation of anaerobic bacteria e.g. Clostridia

Dorset's egg medium:

- Fresh egg contents 75ml
- Nutrient broth 25ml
- 2 % malachite green 1.25ml

- Use: For cultivation of Mycobacteria



Sabouraud's Dextrose Maltose agar:

Peptone	1g
Dextrose or maltose	4g
Agar	2g
Distilled water	100 ml
pH	5.4

Use: the pH of the medium is acidic and is suitable for cultivation of yeast and fungi

THE END

The images for slides are taken from resources available on internet and used for the purpose of teaching students