## **Culture Media**

Culture media: nutrient preparations used to grow microorganisms such as bacteria.

The successful cultivation of microorganisms in the laboratory is dependent upon the provision of a satisfactory nutritional environment. This nutritional environment is divided into (a) the physical environment, embracing such factors as temperature, humidity and atmosphere, and (b) the chemical environment, consisting of those compounds supplied in a culture medium to permit growth of microorganisms.

- most bacteria of medically importance require carbon, nitrogen, water, inorganic salts and source of energy for growth. Some bacteria require special growth factors, including amino acids and vitamins.

#### Common constituents of culture media

#### 1. Water

- essential for the growth of all microorganisms.
- must be free from any chemicals (high mineral conc.) which inhibit bacterial growth.
- 2. Sodium chloride, other electrolytes. (potassium, magnesium, iron, calcium)
  - NaCl<sub>2</sub> is essential ingredient of most culture media.
  - sulphates are required as a source of sulphur phosphates as source of phosphorus.
- 3. Peptone (water-soluble) (animal or plant) (pepsin, trypsin or by an acid)
  - obtained from the digestion of protein materials with proteolytic enzymes.
  - contains a mixture of amino acids, carbohydrates, mineral salts and polypeptides.

# 4. Meat extract, yeast extract:

- used to enrich media
- contain protein degradation products, carbohydrates, inorganic salts and some growth factors
- **5.** Carbohydrates (simple; arabinose, glucose, sucrose, maltose. complex; glycogen, glycerol)
  - provide bacteria with carbon and energy.
  - assist in the differentiation of bacteria. (indicator)

# 6. Agar

- a complex polysaccharide extracted commercially from a variety of seaweed (red marine algae).

- used as a solidifying agent (melt at 90°C and solidify at approximately 40°C), generally in a 1.5 to 2% concentration. (semi solid: 0.4-0.5%; transport media: Aimes)

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There are three general forms of culture media: solid, semi-solid and broth (liquid).

#### Solid culture media:

- dispensed in plastic Petri dishes (agar plates or culture plates). Its preparation can also be made into slant (slop culture or stab culture).
- allow separate colony formation.
- colonial morphology.
- quantitation.

#### Semi-solid culture media:

- prepared by using a lower concentration of agar (0.4 0.5%).
- used mainly as transport media (e.g. Stuart's transport medium) and for motility testing.

## Liquid culture media:

- dispensed in tubes with cotton-wall stoppers or in screw-capped bottles.
- growth of bacteria is recognized by turbidity.
- used mainly as enrichment media, biochemical testing media and blood culture media.

# Types of culture media

The main types of culture media are:

## 1. Simple (basic) media:

- will support the growth of microorganisms that do not require special nutrient.
- e.g. nutrient broth/nutrient agar, which contains peptone and meat extract.

#### 2. Enriched media:

- contain additional nutrients to encourage microbial growth (fastidious bacteria).
- enriched with whole blood, lyzed (heated) blood, special extract (protein, sugar) or vitamins.
- e.g. blood agar, chocolate agar

# 3. Selective media (dye; crystal violet, brilliant green, eosin – chemicals; selenite, desoxycholate, bile salts – antibiotics)

- contain one or more substances that permit the growth of certain organisms while interfere or suppress the growth of others.
- e.g. XLD, Thayer Martin agar, MacConkey agar, alkaline peptone broth.

#### 4. Differential (indicator) media

- contain one or more ingredients designed to stimulate a characteristic biochemical response, which will cause certain colonies to develop differently (differentiated) from other organisms present.
- are often based on sugar fermentation reactions which result in production of acid and the subsequent color change of a pH indicator (methyl red, phenol red, bromothymol blue).
- e.g. MacConkey agar, the indicator is neutral red. Lactose fermenting bacteria form pink colonies, whilst non-lactose fermenting bacteria form pale yellow colonies.
- other indicators may be included to detect specific bacterial products (e.g. ferric and ferrous ion for the detection of hydrogen sulphide).

#### 5. Enrichment media

- a fluid media used to enhance the growth of certain bacterial species while inhibiting the development of unwanted microorganisms.
- e.g. selenite F broth for isolation of *Salmonella* and *Shigella* species from fecal specimen.

## 6. Transport media

- mostly semi-solid media that contain ingredient to prevent the overgrowth of commensals and ensure the survival of aerobic and anaerobic pathogens.
- e.g. Amies' transport medium, Stuart's transport medium.