

Media

Culture Media (Sing. Medium).

Culture — Means Preparation of cells or bacteria grown in an artificial medium.

— To grow & develop

Medium → A preparation needed for growth and development of organisms, cell, tissue, organ etc.

- (i) Natural →
- (ii) Artificial →
- (iii) Polluted →

(i) Artificial → Prepared in laboratories and industries for the proper growth and development of microorganisms.

- (a) Synthetic / - Defined:
- (b) Complex.

(a) Defined or Synthetic → ~~A medium~~ All of its components are known e.g. Cyanobacterial and Eukaryotic culture media.

1) CO₂ as a Carbon Source → Sodium Carbonate or Bicarbonate Na_2CO_3 or $NaHCO_3$.

2) Ammonium Nitrate as an source of → Nitrogen Sodium Nitrate $NaNO_3$.

3) Sulphate _____

4) Phosphate _____

5) a Variety of Materials.

BG-II Medium for Cyanobacteria

| | |
|---|--------|
| NaNO ₃ | 1.5g/l |
| KH ₂ PO ₄ · 3H ₂ O | 0.04 |
| MgSO ₄ · 7H ₂ O | 0.075 |
| CaCl ₂ · 2H ₂ O | 0.036 |
| Citric Acid | 0.006 |
| Ferric ammonium Citrate | 0.006 |
| EDTA (Na ₂ Mg Salt) | 0.001 |
| Na ₂ CO ₃ | 0.02 |
| Trace metal solution | 1.0 ml |
| Final pH | → 7.4. |

For E. coli

(2)

| | | |
|---|---|-------------|
| Glucose | — | 1.0 g/l. |
| Na ₂ HPO ₄ | | 16.4 |
| KH ₂ PO ₄ | | 01.5 |
| (NH ₄) ₂ SO ₄ | | 02.0 |
| MgSO ₄ · 7H ₂ O | | 0.200.0 mg |
| CaCl ₂ | | 0010.00 mg |
| FeSO ₄ · 7H ₂ O | | 0000.50 mg. |
| Final pH | — | 06.8 — 7.0. |

- x) Photoautotrophs are mainly grown on such simple media.
- x) Many chemoorganotrophic heterotrophs can be grown on synthetic medium ^{supplemented} with glucose as a carbon source & Ammonium salt as Nitrogen source.
- x) These medium may contain few or a large number of compounds, but all are known as their constituents.
- x) Defined media are widely known in research as it is desirable to know what the experimental microorganism is metabolizing.

Complex Media :- They contain such ingredients ~~that~~ ^{are of} some of them unknown chemical composition.

(eg. Peptones; meat extract (Beef extract); Yeast extract etc as they contain undefined components).

x) They are very useful as they fulfil the requirements of a large number of microbes and they we can grow on the ~~the~~ unknown microbes. (Microbes which nutritional requirements are unknown) in such case we cannot construct a defined medium).

Many fastidious (1. Payin a lot of attention to detail bacteria. 2. Very concerned about cleanliness). Some of them even require Blood or Serum e.g. Serum albumin.

x) Peptones → Proteins hydrolyzates prepared by partial proteolytic digestion of meat, caesin, soya meal, gelatin or some other protein sources.

They serve as Source of carbon, nitrogen & energy.

- v) Beef extracts } → Aqueous extract of lean Beef.
- v) Yeast extracts } → " " " Brewer's Yeast.

v) Beef extract → posses Amino acids, peptides, nucleotides Organic acids, Vitamins & Minerals.

(4)

Yeast Extracts :- B. Excellent source of Vitamin B and Carbon and Nitrogen.

e.g. Commonly used Complex media are:

- (i) Nutrient Broth.
- (ii) Tryptic soy broth.
- (iii) Mac Conkey agar.

Nutrient Broth

Peptone (Gelatin hydrolysate) → 0.5 gm/l.
Beef extract → 0.3 gm

Tryptic Soy Broth

Tryptone (~~Gelatin~~ by Pancreatic digestion of Casein) - 17 gm/l
Pepton (soyabean digest) 0.3 gm
Glucose 0.25 gm
Sodium chloride 0.50 gm
Dipotassium Phosphate 0.25 gm

Mac Conkey Agar

- i) Pancreatic digestion of ge

MacConkey Agar

| | |
|--------------------------------|-----------------|
| Pancreatic digest of gelatin | 17.0 g/ml litre |
| " " " Casein | 01.5 |
| Peptic digest of animal tissue | 01.5 |
| Lactose | 10.0 |
| Bile salts | 01.5 |
| Sodium chloride | 05.0 |
| Neutral Red | 00.03 |
| Crystal Violet | 00.001 |
| Agar | 13.5 |

For solidifying 1.5 of Agar may be used if needed.

(x) Agar is a sulfate polymer composed mainly of
D-galactose,
3,6-anhydro L-galactose
D-glucuronic acid

Extracted from a red algae

Most of Microbes cannot digest it.

— Melted in Boiling water

— cooled to $40 - 42^{\circ}\text{C}$ before hardening.

→ Melt again about $80 - 90^{\circ}\text{C}$.

(x) Silica gel is other solidifying agent,
used usually to grow autotrophic bacteria in absence
of organic matter for determination of Carbon source of
heterotrophic bacteria, supplementing with various organic compounds.

Types of Media on the basis of Use. ⁽⁶⁾

① General purpose Media e.g. tryptic soya broth.

They support a large number of microbes

Blood and other special nutrients ~~are~~ added may be added to these general purpose media to increase the growth of fastidious heterotrophs.

Such an specialized media is called (Blood)

② enriched Media or Supplemented media

③ Selective media:- They favour the growth of a particular microorganism

e.g. Bile salts ^{dyes}, Basic Fuschin and Crystal violet favour the growth of gram (-)ve bacteria by inhibiting the growth of gram (+)ve bacteria without affecting ~~so~~ gram ~~+~~ bacteria

Endo agar, Eosin methylene blue agar, MacConkey Agar

→ For detection of E. coli and related bacteria in water supplies, ~~it~~ and other sources.

They contain certain dyes ~~and~~ which ~~suppress~~ suppress gram (+)ve bacterial growth.

For selection of Cellulose digestion bacteria we use only cellulose as both carbon and energy source. (7)

④ Differential Media → They can distinguish between different groups of bacteria

They even permit tentative identification of microorganisms based on their biological characters.

e.g. Blood agar → Distinguishes between hemolytic and non-hemolytic bacteria (Both enriched & selective).
(Differential media).

Hemolytic Bacteria → Strepto & Staphylococcus extracted from throat, produces clear zones around their colonies due to destruction of R.B.C.s.

MacConkey Agar → Differential & selective.

It possess lactose & neutral red dye, hence Lactose forming colonies appear pink to red in colour hence can be easily distinguished from non fermenter colonies.

Complete Media & Minimal Media

- Q. Describe media. Their types and uses ?
- Q. Describe
- Q. What are peptones ? What are their uses ?

