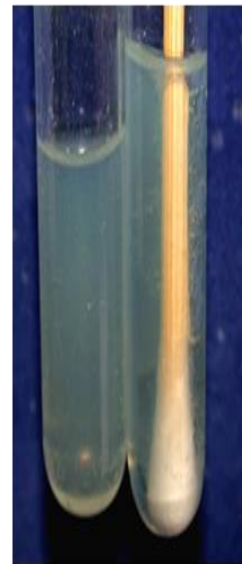




Part I- Culture media

Part II- Cultivation techniques



Cultivation of bacteria :

The macroscopic study of microorganisms requires

1- Techniques for **isolating** cells from natural sources.

2- **Growing** them in the laboratory on culture media.

- - It is different from one type of bacteria to another according nutrition requirement like energy ,carbon and nitrogen sources.

- **Microbial cultivation:** is a method of multiplying the microorganisms by letting them reproduce in predetermined culture media under controlled laboratory conditions.

Cultivation is necessary for study and diagnosis of bacteria when we get a sample .

Type of sample :

- blood .
- urine .
- Stool
- Pus.
- Sputum .
- wound swab .
- skin swab .
- csf (cerebrospinal fluid)
- Biopsy .

Why cultivate bacteria?

- 1- Isolation.
- 2- Identification and characterization.
- 3- Determine which bacteria is most likely causing infection.
- 4- Test antimicrobial susceptibility .
- 5- Bank strain for future use .



Types Of Culture Media :

Can be Classified based on :

A - **Natural** : **ex** : animal tissues , plant tissues , beef extract , etc .

B - **Artificial** : making by many company .

Artificial media divided to :

1- Supportive media (Basic media)

e.g. Nutrient agar , Nutrient broth .

Nutrient broth .(fluid culture medium)

(meat extract , peptone , Nacl)

Nutrient agar : (solid culture medium)

(Nutrient broth + a gar)

PH: 7.2 _7.4



Nutrient agar



Nutrient broth

Simple culture media

2- Selective media :

e.g. MacConkey agar , S - S agar .

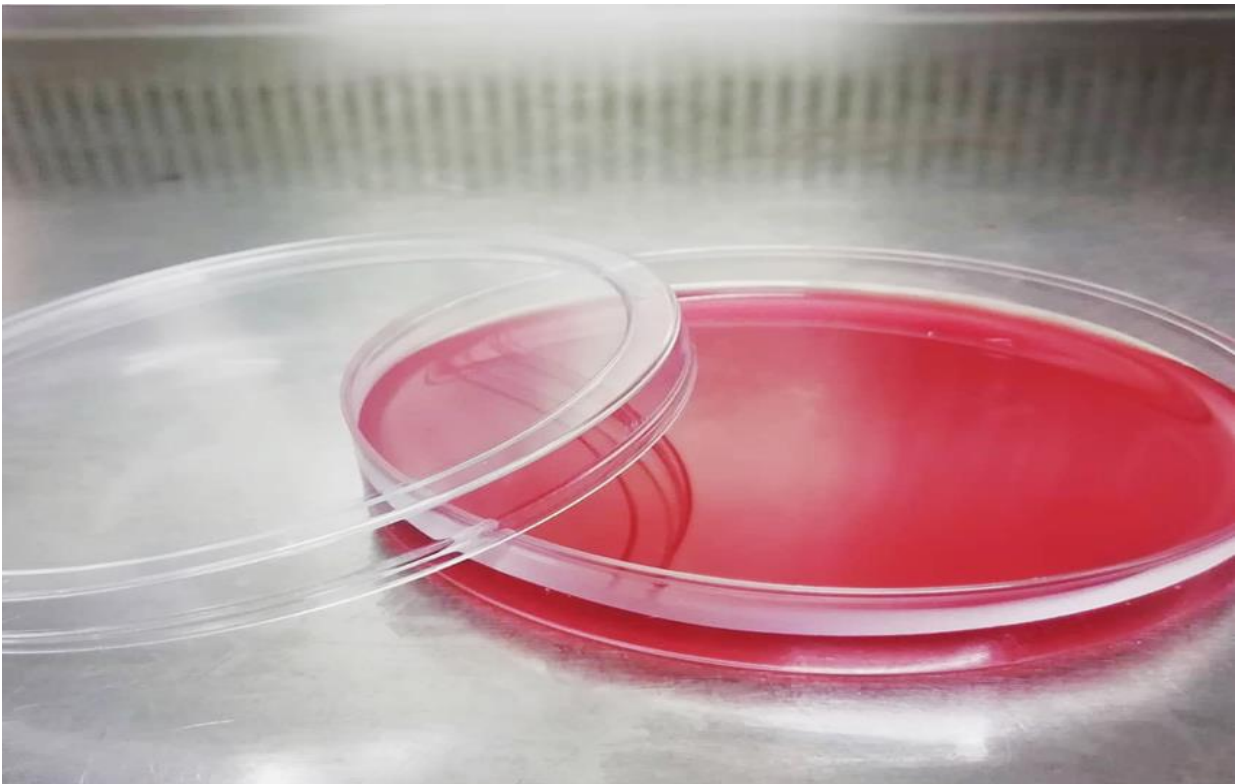
Media that use selective agents to inhibit growth of certain microorganisms thereby selecting for the growth of other microorganisms .

Example of selective agent (high salt ,high or low PH ,other chemical additives)

-**Ex:** macConky agar select gram negative bacteria on gram positive bacteria

. **Ex:** *Salmonella* - *Shigella* agar (S-S agar) .

macConky agar

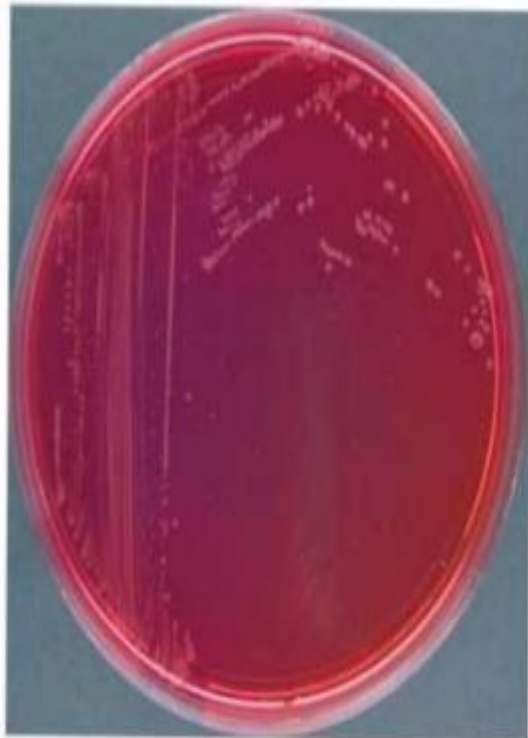


3-Enrichment media:

- ❑ In this media add special nutrition requirement for grow special type of bacteria for example (growth factors ,blood ,additional vitamins or nutrients).
- ❑ **Ex :** s-s agar to grow g. *Salmonella* , *Shigella*
- ❑ **Ex :** chocolate agar to grow g .*neisseria*

Enrichment media :

- o *Shigella* do not have black center in this medium as *Salmonella* .

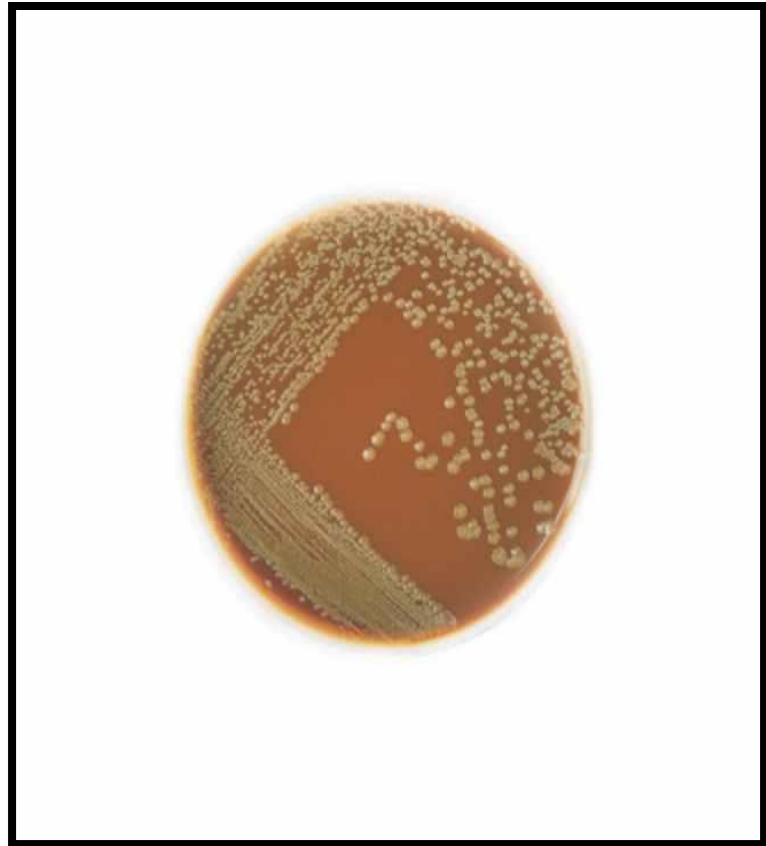


***Shigella* on XLD.**



***Salmonella* on XLD.**

Chocolate agar :



Chocolate agar : contain red blood cells that have been lysed by slowly heating to 80c .

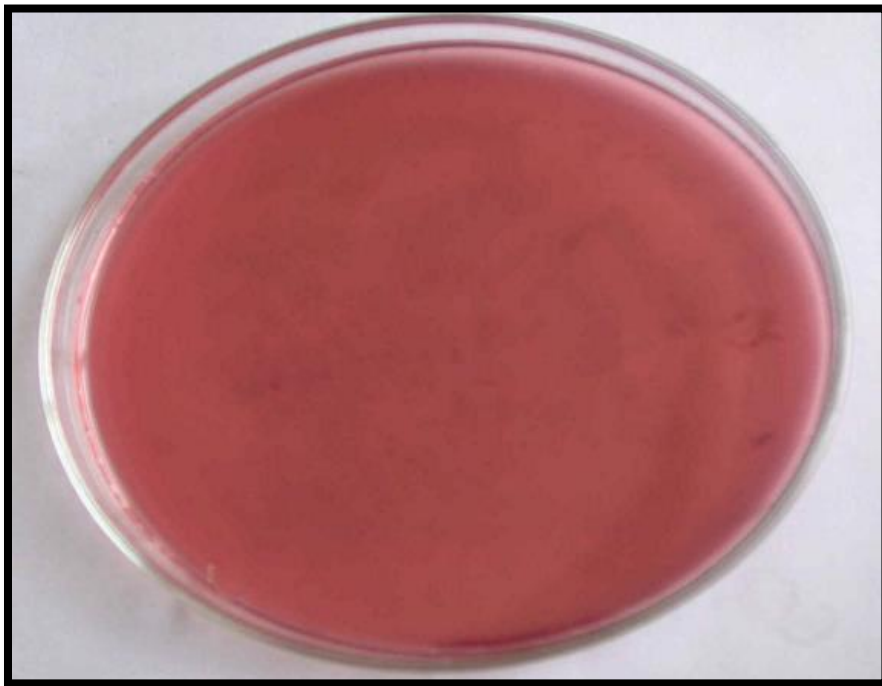
4 -Differential media :

1-Ex : MacConkey agar

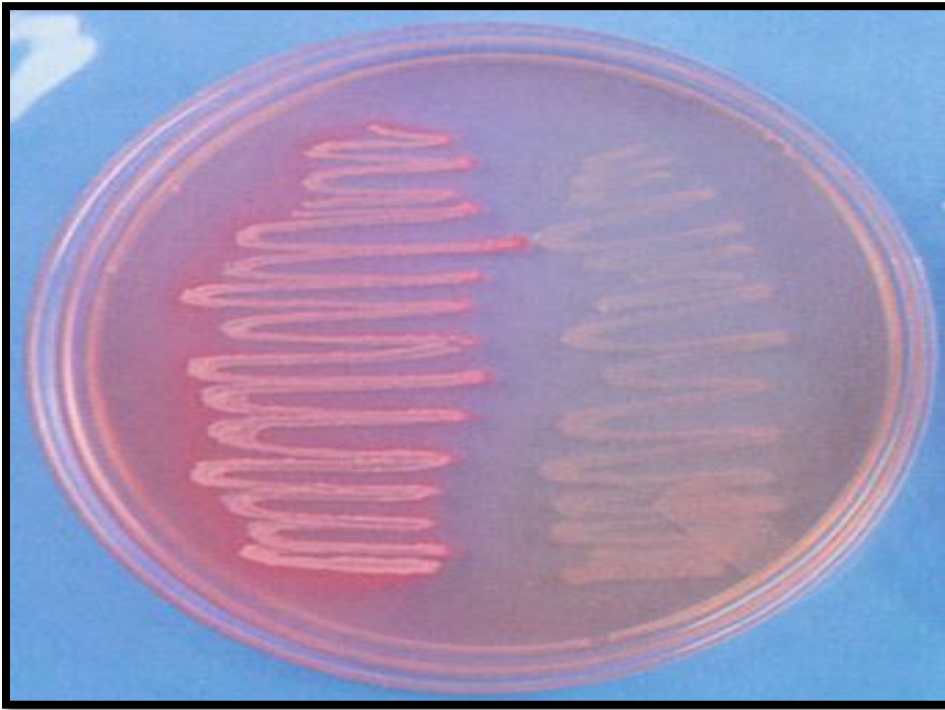
All type of bacteria are grow but each one have feature differ from anther .

Ex: all genus of family Enterobacteraceae(Gve-) are grow but *G. E.Coli* Appear pink because it (**lactose fermented**) while *G.Salmonella* and *Shigella* appear pale (**non lactose fermented**).

Fermentation decreases PH indicator in the media causes a color chang



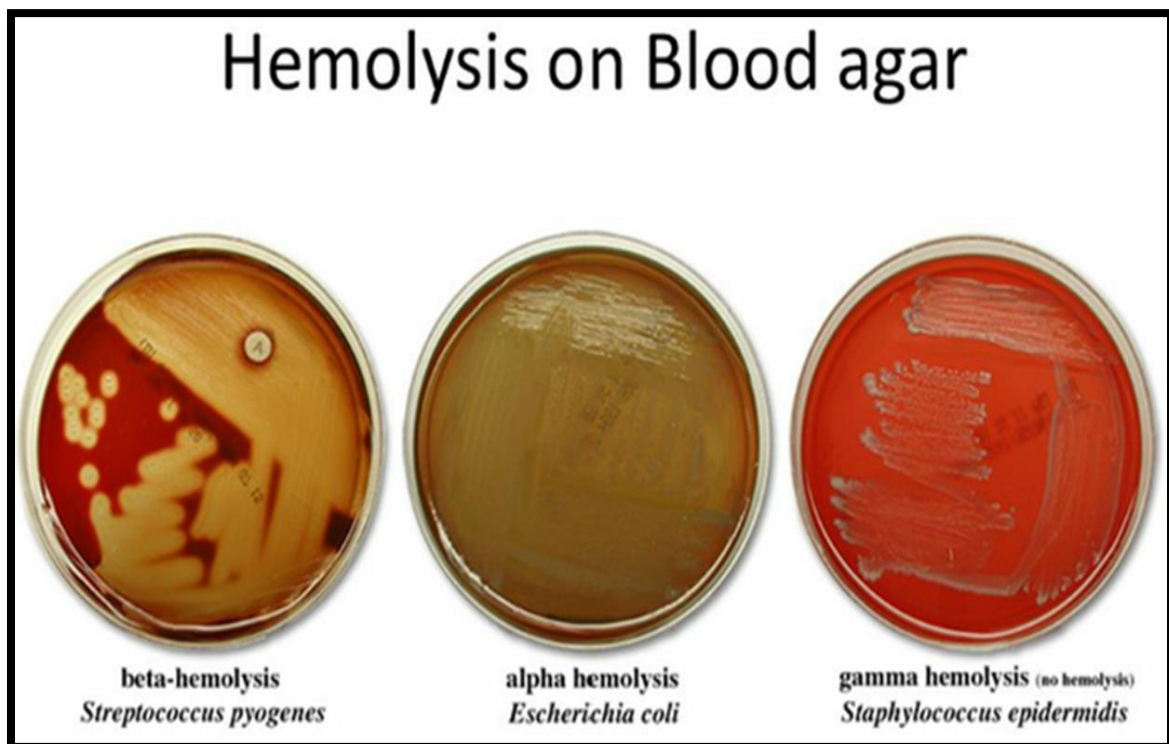
e .



Ex: **blood agar** :G. Sterptococcus are grow and hemolyse blood but S . Pyogen have beta hemolytic while S . Pneumonia have alpha hemolytic

Blood agar





there are another classification of culture media according solidity:

agar is the material that responsible about solidity in culture media .

1-Solid media :bacteria grow on surface of these media as colony Ex: blood agar ,nutrient agar ,macconky agar .

- These media may be as many shape .

A-in petri dish :use for bacterial growth generally for making some diagnostic tests also for sensitivity against chemotherapy agent .

B - deep :in test tube ,use for growth anaerobic bacteria.

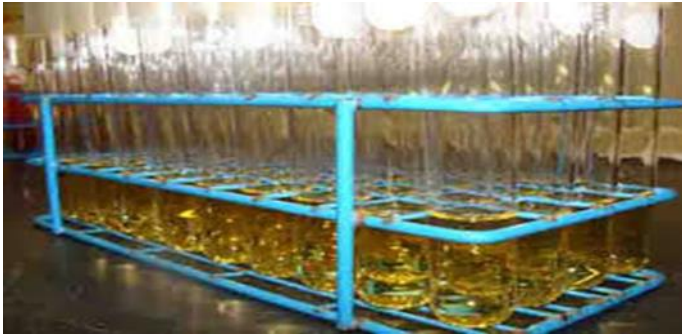
C - slant :in test tube use for increase surface area of bacterial growth .

2-semi solid media: have agar less than solid media ,use for study some bacterial activity like motility .

3-liquid media : these media have not agar , bacterial growth are seem as turbidity ,use for activating bacteria also when take sample .

Ex : nutrient broth , macconky broth .

liquid media



2- Solid form---- Nutrient agar

Plate



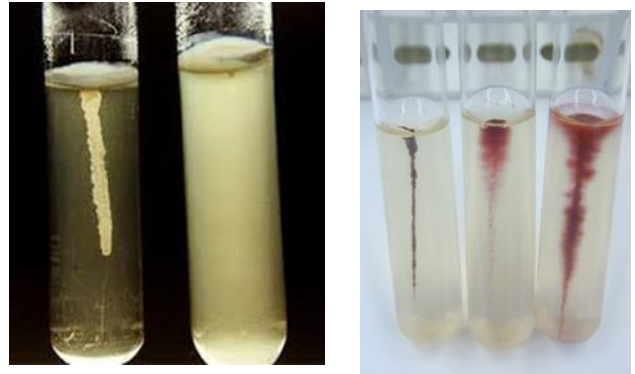
Deep



Slant



3- Semisolid – test motility



Material:

- 1-Boder of wanted media or its contains .
- 2- volum flask (100-250-500 ml).
- 3-balance .
- 4-distilled water .
- 5-brner.
- 6-steril petri dish.
- 7-coton.
- 8-alminum foil.
- 9-alcohol(80).
- 10-disinfectant .

Procedure:

1 - add suitable amount of wanted culture media to (100ml) of D.W(or its contains take it form table) then shake it until dissolution .

2 - heat up the media until boiling then closed flasks mouth by coton and cover it with aluminum foil.

3 - enter the flask to autoclave for(20 min) for sterilizing .

4 - the media to cool off (40c*).

5 - distributed this media in sterile petri dish closeness of flam in side inoculation cabinet and pass flasks mouth many time on flam .

Not make sure the media sterilized put it in incubator over night .

