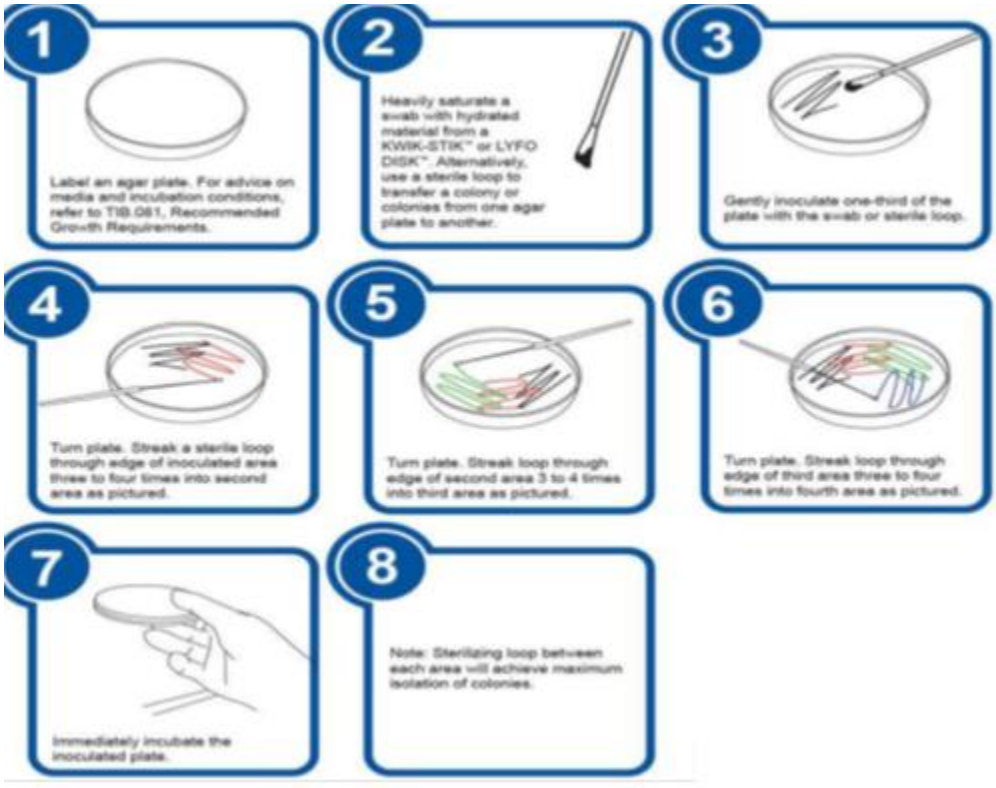
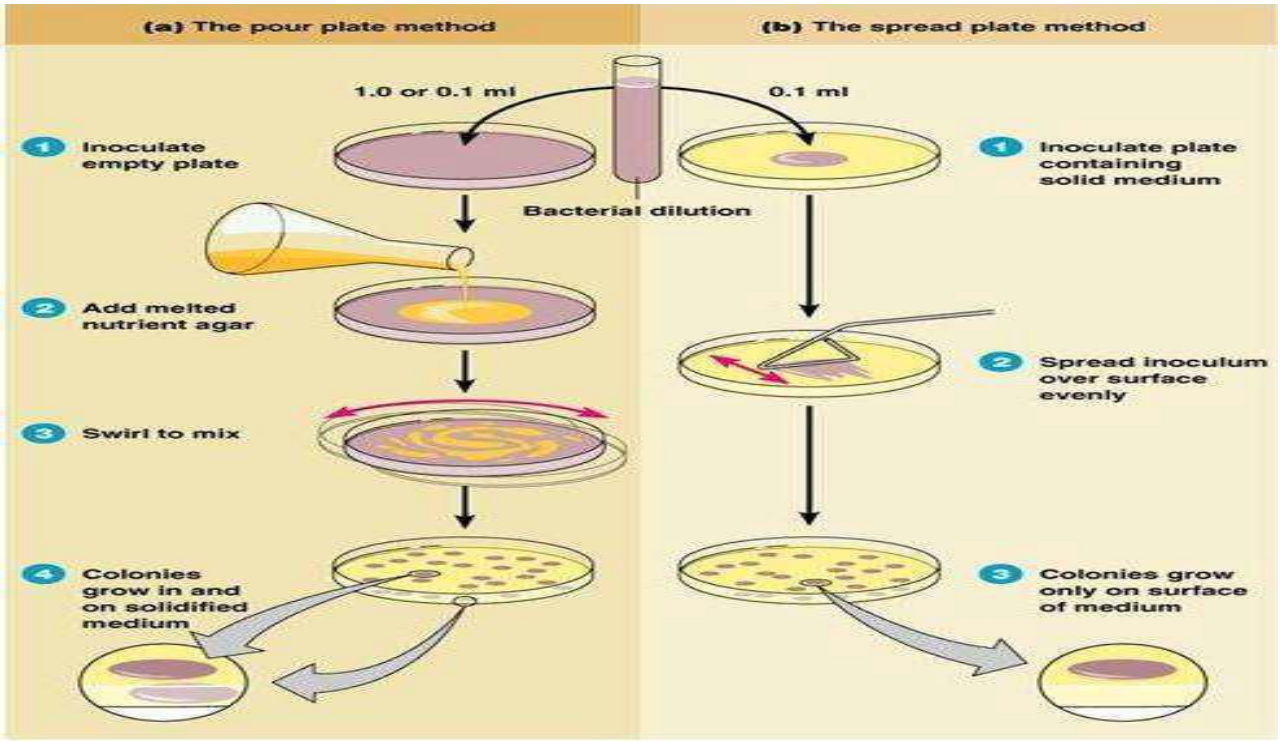
LAB:3 CULTURE METHODS

Indications for culture -

- Isolate bacteria in pure cultures.
- Demonstrate their properties.
- Obtain sufficient growth for preparation of antigens & for other tests.
- Typing bacterial isolates.
- Antibiotic sensitivity.
- Estimate viable counts.
- Maintain stock cultures.





METHODS OF ISOLATION:

- Streak culture or surface plating
- Lawn or carpet culture
- Stroke culture
- Stab culture
- Pour plate method
- Anaerobic methods of culturing bacteria

1. Streaking:

- Routinely employed for isolation
- Platinum / Nichrome loops

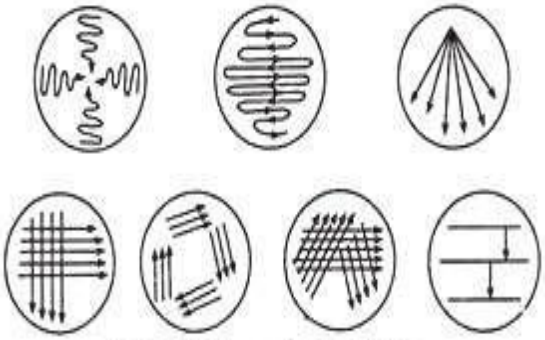
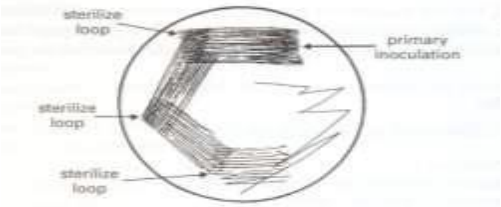
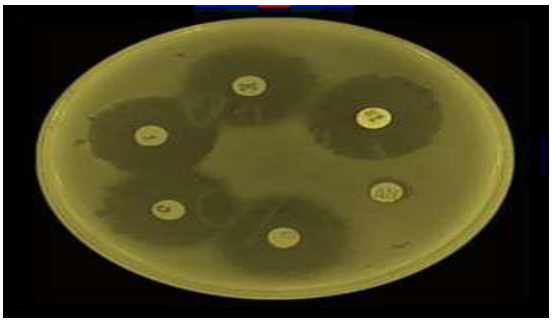


FIG. 16.13. Various methods of streaking.



2. Lawn or Carpet Culture

- Uniform surface growth
- Bacteriophage typing
- Antibiotic sensitivity testing
- Preparation of bacterial antigens & vaccines

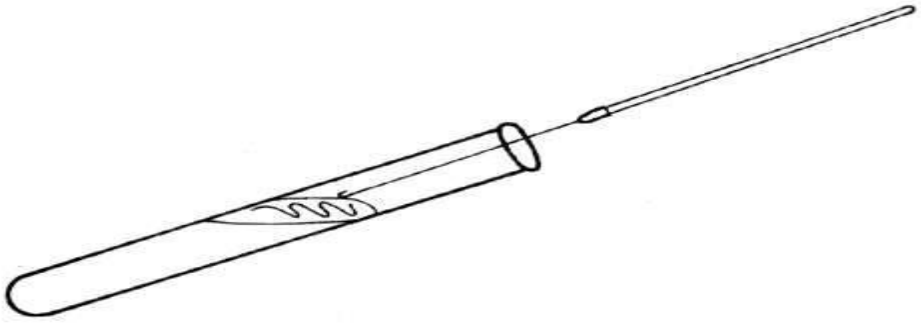


Antibiotic sensitivity testing

3- STROKE CULTURE

• Tubes containing agar slopes

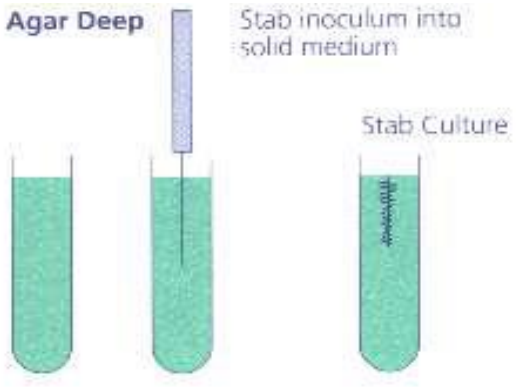
• For slide agglutination & other diagnostic tests.

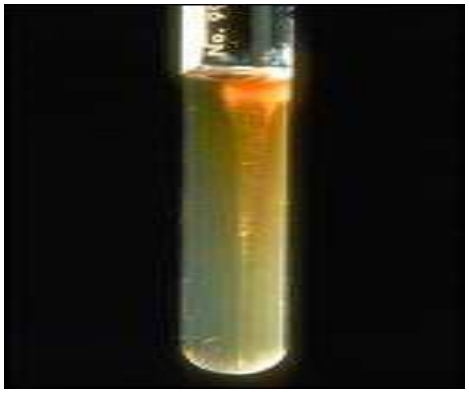


4- STAB CULTURE

• By puncturing a suitable medium with a long, straight charged wire.

• For gelatin liquefaction, stock cultures & motility





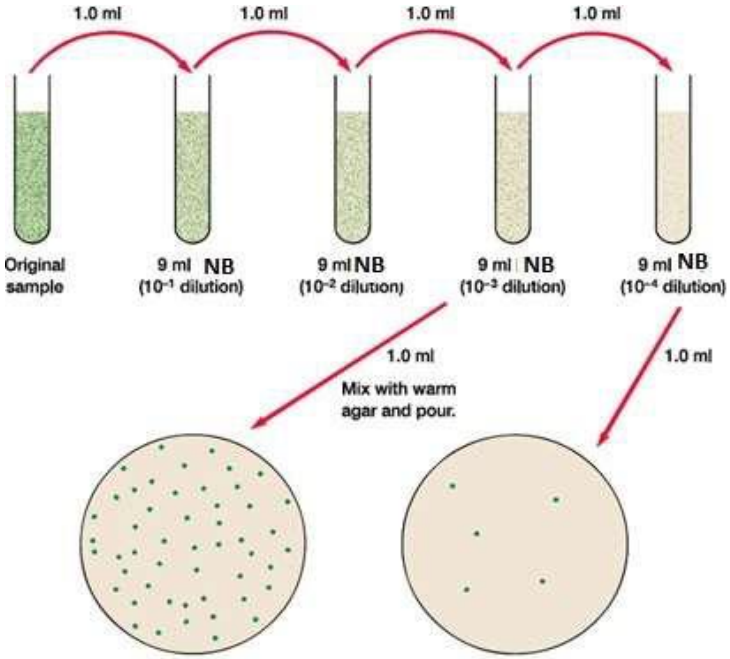
5- POUR PLATE METHOD

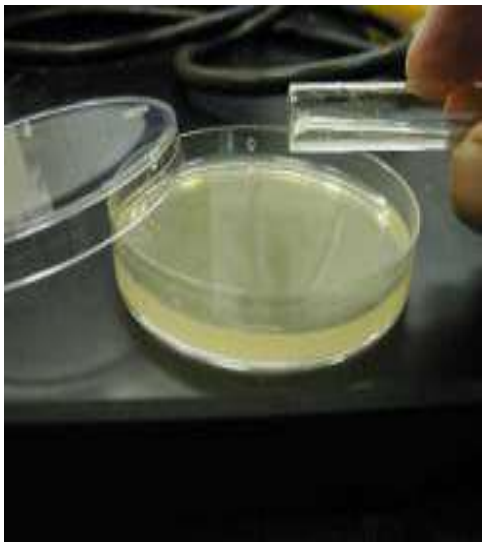
• 1 ml of appropriately diluted inoculum is added to 15 ml of molten

agar and poured on petridish.

• Colonies appear through out the depth of medium.

• Used to estimate viable count, recommended method for quantitative urine cultures.





BROTH/LIQUID CULTURE

• Inoculated by a charged loop, pipette or syringes.

• For blood cultures & sterility testing .

6. ANAEROBIC CULTURE METHODS

Anaerobic condition can be achieved by:

• Cultivation in vacuum

• Displacement of oxygen with other gases

• Chemical or biological methods

• By displacement and combustion of oxygen

• By reducing agents

• Anaerobic chamber

DISPLACEMENT METHOD

• Displacement of O2 with gases like H2 , N2 , He or CO2 .

• Rarely produces complete anaerobiosis.

e.g. Candle jar

CHEMICAL OR BIOLOGICAL METHODS

• Alkaline pyrogallol (pyrogallic acid in NaOH) absorbs O2

• Yellow phosphorous

• Rosenthal method - Mixture of chromium & sulphuric acid

• Gaspak

BIOLOGICAL METHODS

Absorption of oxygen from small closed systems has been attempted by incubation along with

- Aerobic bacteria EXAMPLE:- *Pseudomonas aeruginosa*
- Anaerobiosis produced by this method is slow and ineffective.