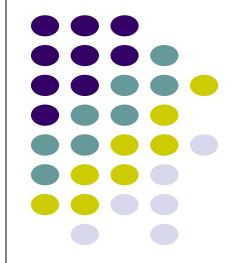
### Pure culture and selective media

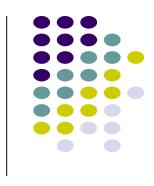


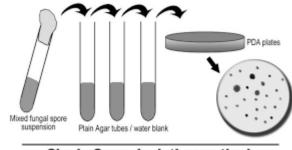


- Fungi can be purified by the following two methods
  - Single spore isolation method
  - Single hyphal tip method

# Single spore isolation method

- This method can be employed, when the fungus produces spores, which are coloured and bold.
- Procedure
- Melt 3 tubes of plain agar and cool them to 50 °C.
- Transfer a loopful of the spore suspension of the mixed culture to the first plain agar tube with an inoculation needle.
- Shake well for uniform dispersion of spores.
- Transfer a loopful from the first dilution tube to the second tube shake thoroughly. Likewise prepare third dilution also.
- Pour the media with diluted spore into three separate petriplate and allow to solidify
- Observe Petri plates under low power objective of the microscope and locate isolated single spore.
- Transfer the single spore to PDA slant to obtain pure culture





Single Spore Isolation method

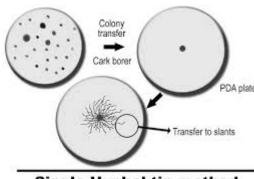
# Single hyphal tip method



 This method is employed for purifying fungi which either do not produce spores or produces small and hyaline spores.

#### Procedure

- Using cork borer, take a disc of fungal colony and place it in the middle of a plain agar plate and incubate for 1-2 days.
- Place the Petriplate in the stage of a compound microscope and locate a hyphal tip using the low power objective.
- With the cork borer, remove it and place it in a PDA slant and maintain it as a pure culture.



Single Hyphal tip method



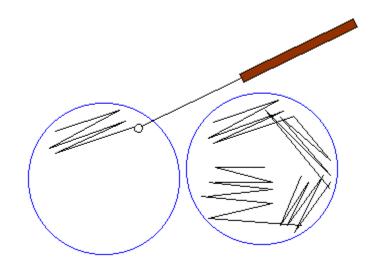
 Purification of yeast cultures: Yeast are single celled eukaryotic microorganism and produces slimy white soft colonies resembling the bacteria. They will not produce mycelial filaments. Purification can be done like that of bacterial cultures using Streak plate method, pour plate method and spread plate methods



- The methods used to isolate the bacteria in pure culture are:
  - Streaking or Plating
  - Dilution and Plating

# Streak plate technique



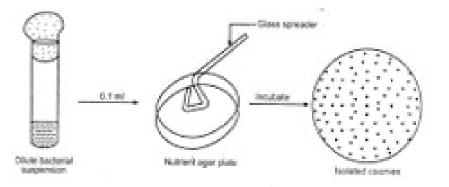




# Spread plate technique



### Spread plate technique



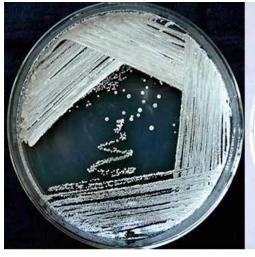




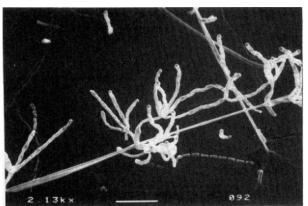
- Selective media are used for the growth of only selected microorganisms. For example, if a
  microorganism is resistant to a certain antibiotic, such as ampicillin or tetracycline, then that
  antibiotic can be added to the medium to prevent other cells, which do not possess the
  resistance, from growing.
- Examples of selective media include:
- Eosin methylene blue contains dyes that are toxic for Gram-positive bacteria. It is the selective and differential medium for coliforms.
- YM (yeast extract, malt extract agar) has a low pH, deterring bacterial growth.
- MacConkey agar is for Gram-negative bacteria.
- Mannitol salt agar is selective for Gram-positive bacteria and differential for mannitol.
- Sabouraud's agar is selective to certain fungi due to its low pH (5.6) and high glucose concentration (3-4%)

## **Actinomycetes medium**

- Yeast extract malt extract agar (ISP2)
- Actinomycetes isolation agar (AIA)
- Arginine Glycerol (AG agar)
- Glycerol asparagines agar (ISP5)







# Phytophthora isolation medium

- V8 juice agar
- Rye A agar



