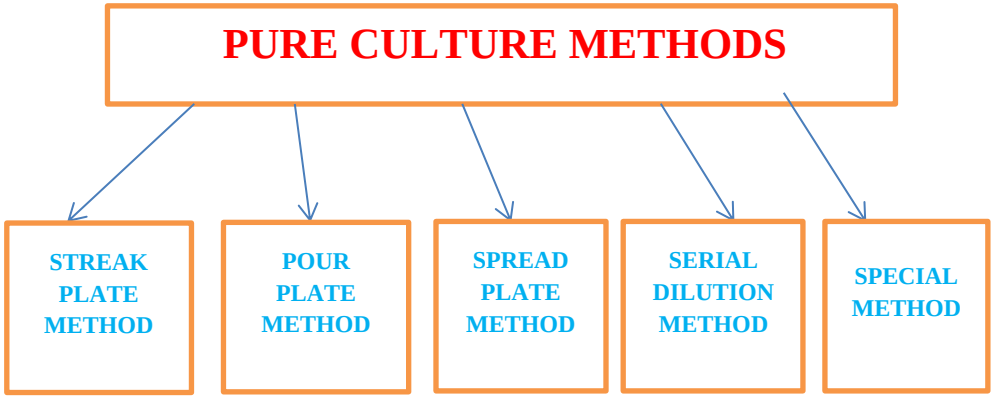
ISOLATION AND PRESERVATION METHOD

- **Pure culture technique**
- **Preservation culture method**

- **Pure culture technique:** The pure culture consists of a population of only one species of microorganism. The isolation of one kind of microorganism from a mixture of many different kinds is called the pure culture technique.



A. STREAK PLATE METHOD:

The streak plate method is a rapid qualitative isolation method. The techniques commonly used for isolation of discrete colonies initially require that the number of organisms in the inoculums be reduced. It is essentially a dilution technique that involves spreading a loopful of culture over the surface of an agar plate. Streaking is rapid and ideally a simple process of isolation dilution. The technique is done by diluting a comparatively large concentration of bacteria to a smaller concentration. The decrease of bacteria should show that colonies are sufficiently spread apart to effect the separation of the different types of microbes. Streaking is done using a sterile tool, such as a cotton swab or commonly an inoculation loop.

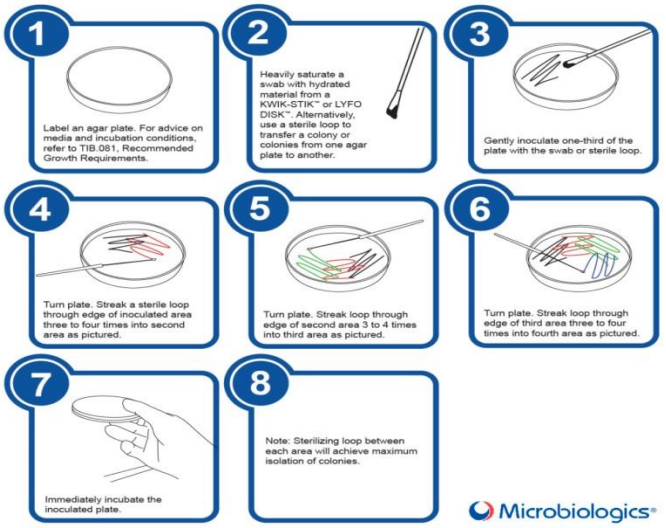


Fig no : 1 Streak plate method

B. POUR PLATE METHOD:

Pour plate method is usually the method of choice for counting the number of colony-forming bacteria

present in a liquid specimen. Because the sample is mixed with the molten agar medium, a larger volume can be used than with the spread plate. In this method, a fixed amount of inoculum (generally 1 ml) from a broth/sample is placed in the center of a sterile Petri dish using a sterile pipette. Molten cooled agar (approx. 15mL) is then poured into the Petri dish containing the inoculum and mixed well. After the solidification of the agar, the plate is inverted and incubated at 37°C for 24-48 hours.

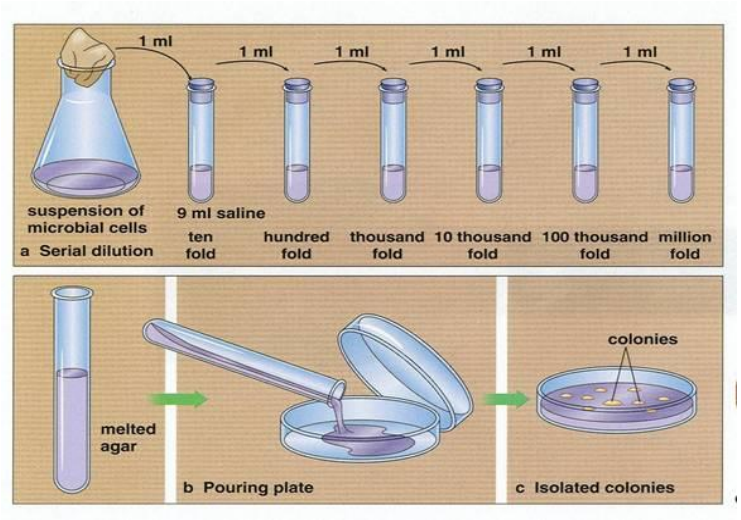


Fig no : 2 Pour plate method

C. SPREAD PLATE METHOD :

Spread plate technique is the method of isolation and enumeration of microorganisms in a mixed culture and distributing it evenly. The technique makes it easier to quantify bacteria in a solution. The spread plate technique involves using a sterilized spreader with a smooth surface made of metal or glass to apply a small amount of bacteria suspended in a solution over a plate. The plate needs to be dry and at room temperature so that the agar can absorb the bacteria more readily. A successful spread plate will have a countable number of isolated bacterial colonies evenly distributed on the plate.

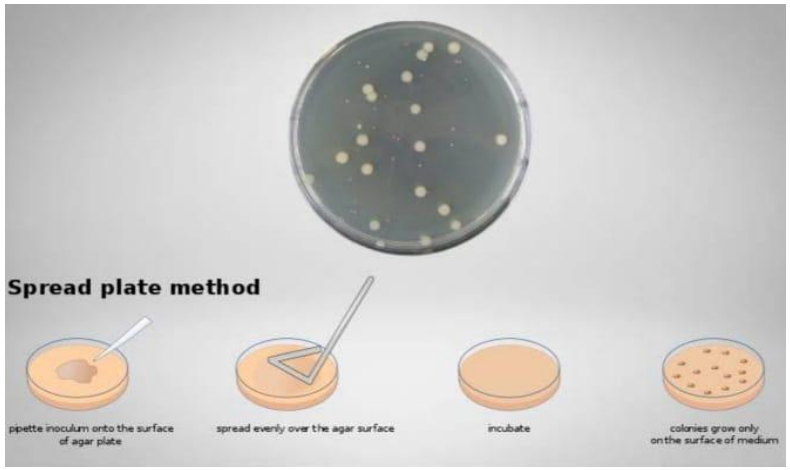


Fig no: 3 Spread plate method

D. SERIAL DILUTION METHOD:

Serial dilution, as the name suggests, is a series of sequential dilutions that are performed to convert a dense solution into a more usable concentration. In

simple words, serial dilution is the process of **stepwise dilution** of a solution with an associated dilution factor. In biology, serial dilution is often associated with reducing the concentration of cells in a culture to simplify the operation.

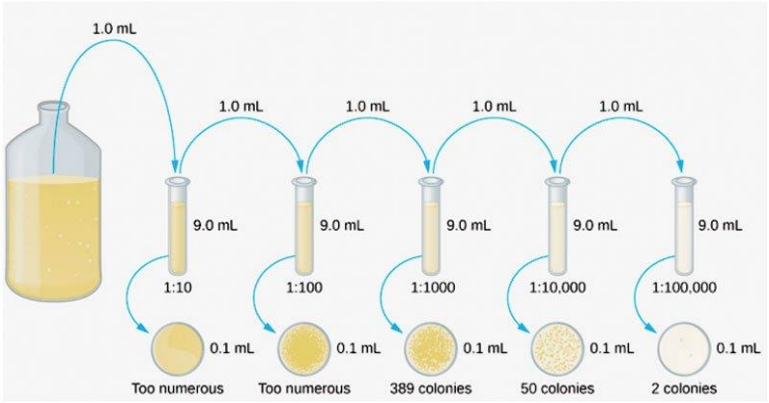
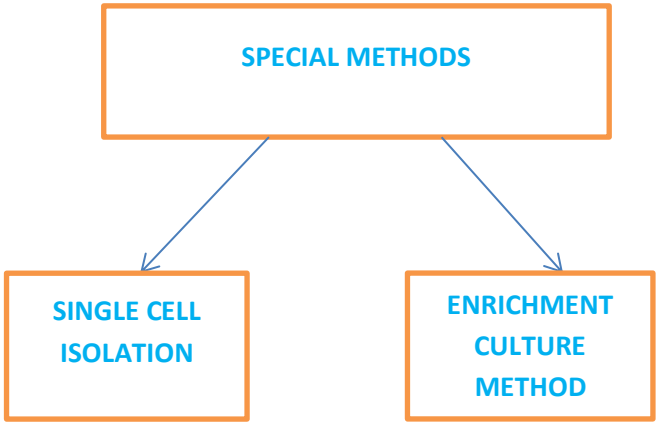


Fig no. : 4 Serial dilution method

E. SPECIAL METHOD:



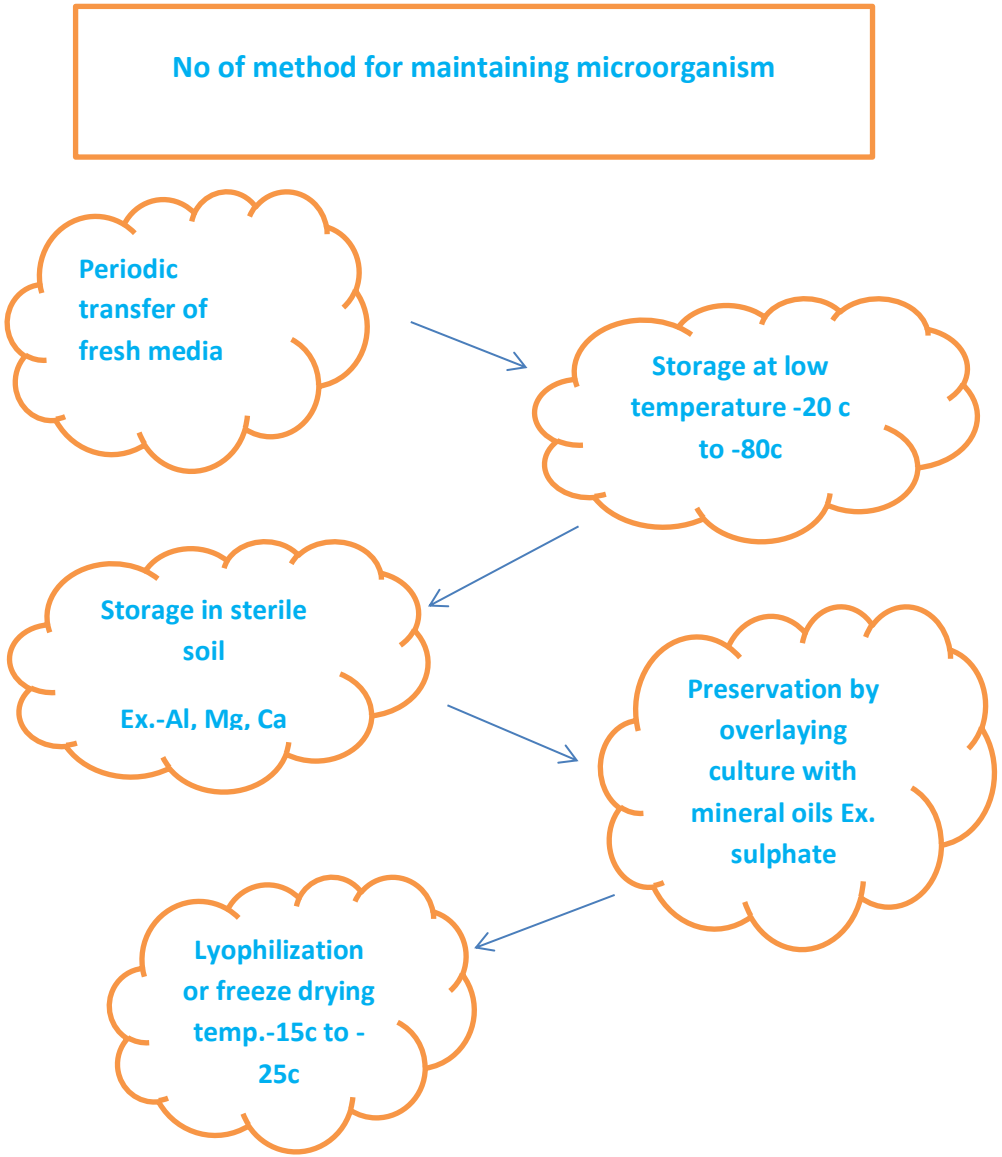
i. SINGLE CELL ISOLATION:

An individual cell of the required kind is picked out by this method from the mixed culture and is permitted to grow.

ii. ENRICHMENT CULTURE METHOD:

The enrichment culture strategy provides a specially designed cultural environment by incorporating a specific nutrient in the medium and by modifying the physical conditions of the incubation. The medium of known composition and, specific condition of incubation favours the growth of desired microorganisms but, is unsuitable for the growth of other types of microorganisms.

- **Preservation of culture:**



- **REFERENCES:**

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2. By Prof. Chandrakant Kokare textbook of Pharmaceutical Microbiology of Nirali Prakashan page no 4.1 to 4.8

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