Methods of isolation of pathogenic bacteria

The identification is required so as to cure the illness or the infection caused due to these bacteria, using appropriate antibiotics. Identification also holds significance for epidemiological purposes.

Isolation of bacteria

Isolation of bacteria forms a very significant step in the diagnosis and management of the illness. Various methods used for isolation of bacteria.

Isolation of bacteria involves various steps:-

1. Specimen collection.
2. Preservation and transportation of specimen.
3. Microscopic examination of sample

1. Specimen collection

Many different specimens are sent for microbiological examination from patients with suspected bacterial infection. Common specimens include urine, faeces, wound swabs, throat swabs, vaginal swabs, sputum, and blood. Less common, but important specimens include cerebrospinal fluid, pleural fluid, tissue, bone …etc.

Some types of specimen are normally sterile e.g. blood, CSF. These samples are usually obtained via a percutaneous route with needle and syringe, using appropriate skin disinfection and an aseptic technique. The culture of bacteria from such specimens is usually indicative of certain infection.

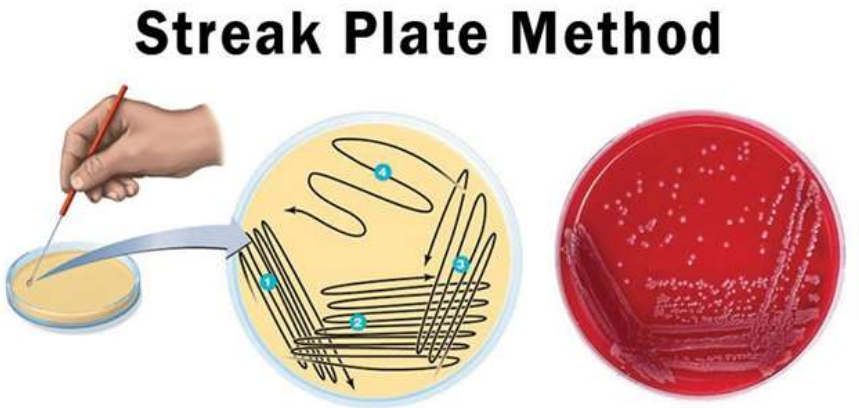
Many microbiological specimens are obtained from non-sterile sites e.g. vaginal or throat swabs, urine sample, stool sample. Specimens must be accurately labelled and indicating patients full name, the nature of the specimen, the source of sample, and the date and time of sample collection.

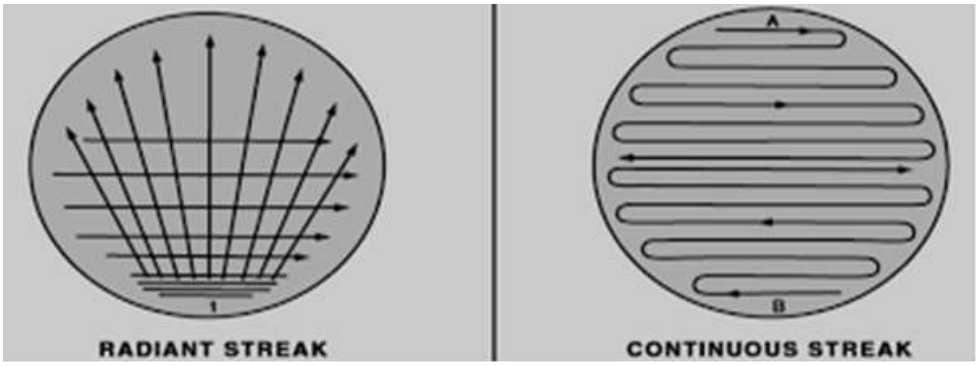
2. Preservation and Transport of specimen

Most specimens are sent to the laboratory in sterile universal containers. Swabs are placed in a suitable transport medium (eg. charcoal medium) otherwise it leads to false negative reporting. In case a delay is expected the specimen should be stored at 4° C.

Method of inoculating the solid culture media

Method used for inoculating the solid media depends upon the purpose of inoculation- whether to have isolated colonies or to know the bacterial load of the sample (quantitative analysis). For obtaining the isolated colonies streaking method is used, the most common method of inoculating an agar plate is streaking.





Culture in liquid media

Bacteria can also be grown in liquid media (broth). Like agar plates, broth cultures may be non-selective or selective. Bacterial growth is easy to detect as the clear liquid turns turbid, usually within 24–48 hr.

The advantage of broth culture is that it is significantly more sensitive than direct culture on agar.

The disadvantage is that, by itself, it is not easy to determine the type of bacteria present or whether a mixed growth has occurred, and in most cases the broth must be subcultured onto solid agar plates.