

The principle of Hanging drop preparation : is to determine the motility of living microorganisms from a liquid medium on an unstained slide by using a microscope.

Hanging drop is the simplest for examining motile -living microorganisms, which is to suspend them in a fluid (water, saline, or broth), then add a drop of bacterial fluid over a glass coverslip to be examined later

Objectives of Hanging Drop Technique :

- To determining the motility of living bacteria.
- To learn about the different methods of motility determination.

Motility could be detected by:

- 1) Hanging Drop technique.
- 2) Semi-Solid media Inoculation.
- 3) Flagella stain.

Examples of motile bacteria include:

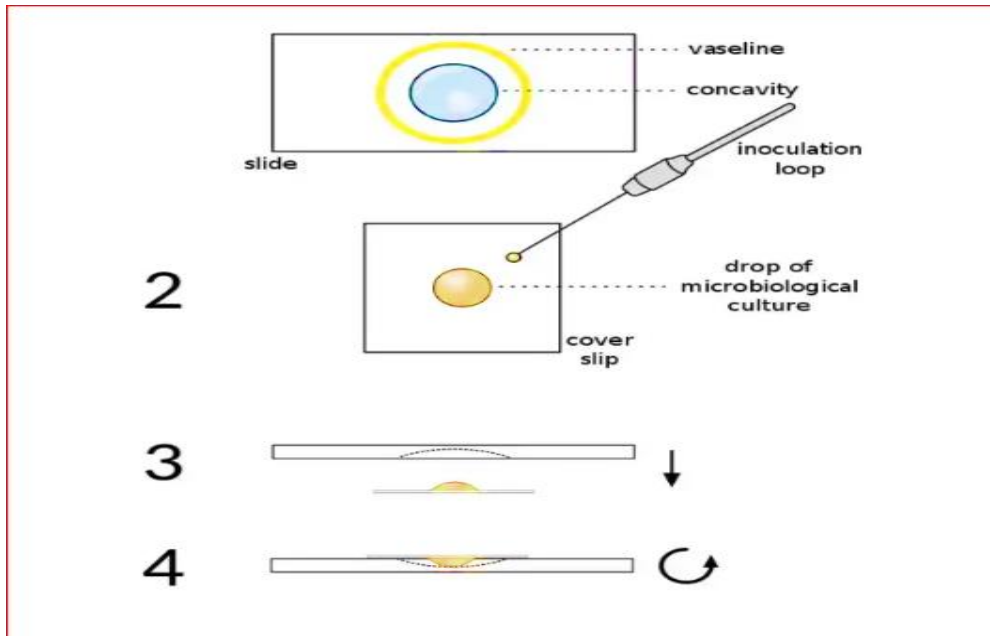
- 1- *Escherichia coli* : has swimming motility through aqueous environments.
- 2- *Pseudomonas aeruginosa*: Uses flagella for swimming and swarming motility over solid surfaces .
- 3- *Proteus species*: swarming motility.
- 4- *Salmonella species*.
- 5- *Vibrio cholerae*.
- 6- *Helicobacter pylori*.

The Materials of a technique :

- 1-24-hour broth culture of bacteria .
- 2-cavity slides & cover glasses.
- 4- Wire loop & Bunsen burner .
- 5- Petroleum jelly.
- 6-microscope .

Procedure of Hanging Drop Method :

- 1- Preparation of the Slide: A clean coverslip is prepared, and a drop of the sample (e.g., Young bacterial broth or cell suspension) is placed in its center.
- 2- Inversion and Attachment: A concave slide (a small well in the center), is then inverted and pressed onto the coverslip. This traps the liquid drop, causing it to hang from the coverslip into the concave slide.
- 3- Sealing: The edges of the depression slide are often sealed with a ring of petroleum jelly (Vaseline) to maintain humidity and prevent evaporation.
- 4-Microscopic Examination: The sealed slide is then placed under a microscope with 10x and 40x lenses, where the living organisms in the hanging drop can be viewed in their natural, unstained state.



Semi-Solid media Inoculation:

- The most commonly used test for motility in microbiology lab.
- It depends on the ability of motile bacteria to move through semi-solid media.
- Semi solid media contain about 0.5% Agar

Procedure of Motility Test

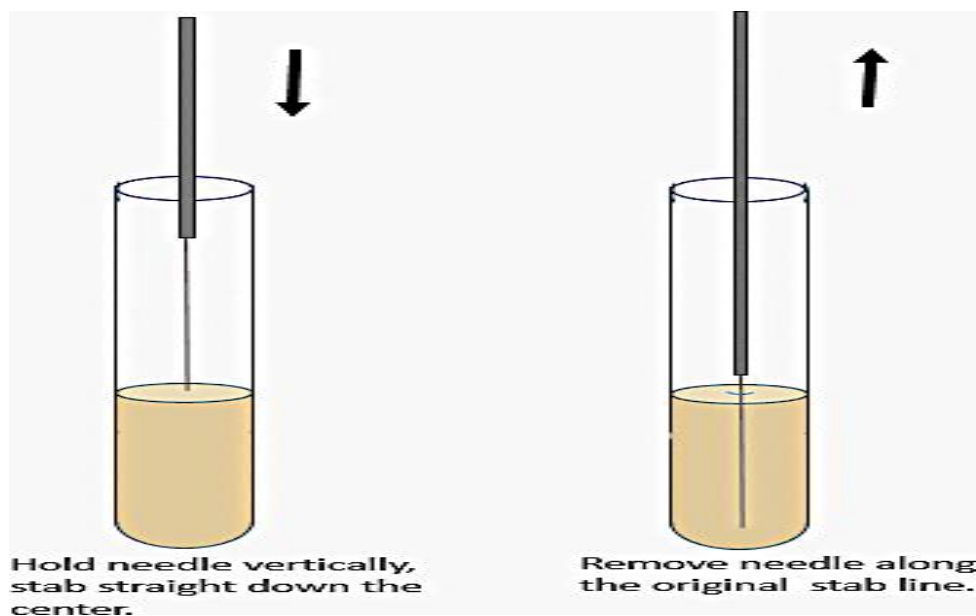
- Using a sterile bacteriological needle, pick a colony of the test organism
- Stab quickly a tube of semi solid media.
- Incubate the semi solid media for 24 hours

*Reading Results:

If bacteria is motile, there will be growth going out away from the stab line, and test is positive.

If bacteria is not motile, there will only be growth along the stab line.

A colored indicator can be used to make the results easier to see



Motility Test

