Flagella Motility

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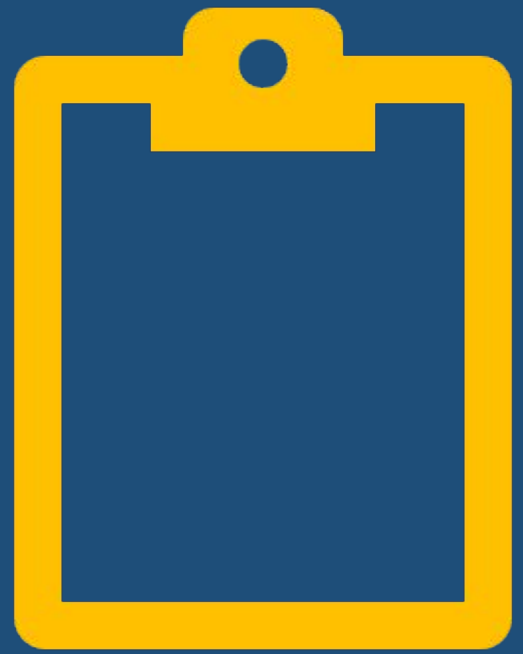
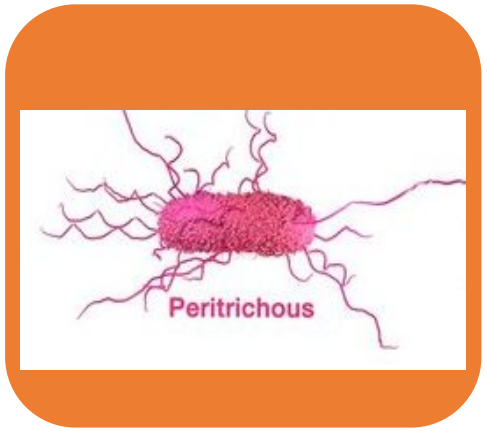
Flagellar Motility

- Motile bacteria move about with structures called flagella. Flagella are too thin to be seen by the ordinary light microscope. Nonmotile bacteria without flagella are called atrichous

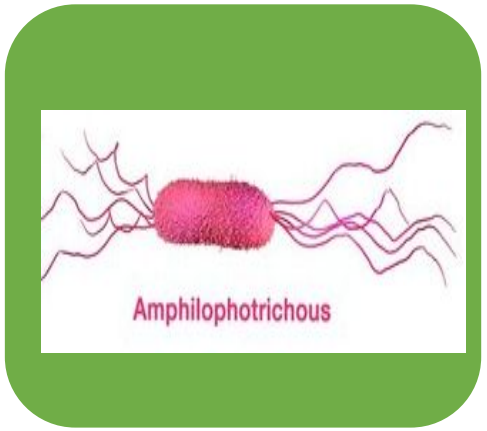


Type of Flagellar

monotrichous



peritrichous



amphitrichous

lophotrichous

Amphitrichous

Atrichous

There are a variety of ways to determine motility of a bacterium—biochemical tests as well as microscopic analysis. Microscopy is the most accurate way to determine motility, assuming that you have a fresh culture of bacteria. Not only can motility be identified, but also the organization and number of flagella.

- Wet mount slide
- Hanging Drop technique.
- Swarming
- Flagella stains
- Semi-Solid media Inoculation

Wet Mount

The aim of this experiment is to observe the natural shape, size and arrangement of bacteria in living condition. The method is also used to determine whether or not an organism is motile. The simplest method for examining living microorganisms is to suspend them in a fluid (water, saline, or broth) and prepare either a “hanging drop” or a simple “wet mount.” In wet mount, a drop of the bacteria suspension is placed on a slide, covered with a cover slip and observed under a compound microscope The slide for a hanging drop is ground with a concave well in the center; the cover glass holds a drop of the suspension. When the cover glass is inverted over the well of the slide, the drop hangs from the glass in the hollow concavity of the slide .

All bacteria have some vibrational movement, even nonmotile ones. This Brownian movement is caused by water molecules bouncing around in the solution, knocking up against each other and the microorganisms. On the other hand, those bacteria with flagella will be very apparently moving about the field of vision, although perhaps not all of the bacteria will be moving. Some cells will "run" straight across the field, others will "tumble" across the field in a slower motion.

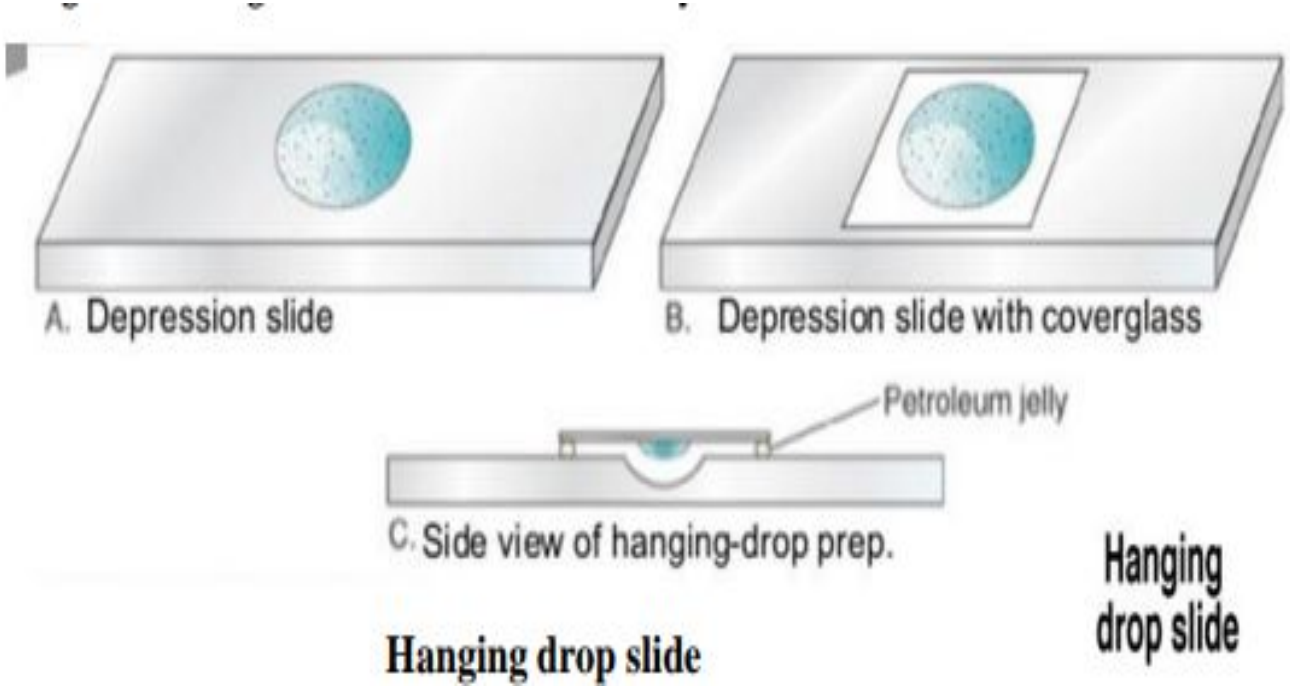
Procedure Motility of bacteria

1. Place a drop of the bacterial culture (optimally from a young broth culture) in the middle of a cover slip.

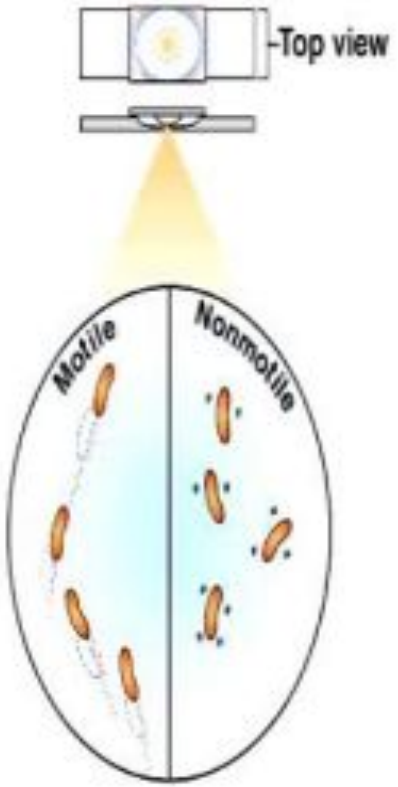
2. Place a thin line of petroleum jelly around the edge of the cover slide, or at the 4 corners of cover slip.

3. Turn the depression slide upside-down (depressed area facing down) and gently touch the cover slide. The jelly holds the cover slip to the slide and also keeps the suspension from drying out.

4. Now flip the entire microscope slide/cover slip combination over. Observe under microscope.



Hanging drop slide



Bacterial Identification Tests: Motility Test (Hanging drop method)

Some Important Motile and Non-motile Bacteria

1. All Enterobacteriaceae are motile, except Shigella species

2. Klebsiella species are non-motile.

3. Pseudomonas species are motile with the help of a unipolar flagellum

4. Vibrio colony has a shooting star motility

5. Campylobacter has a darting motility

6. Listeria monocytogenes has a tumbling motility

Hints:

1. Unstained bacteria are very transparent and difficult to see. Therefore, while focusing, reduce the lighting sufficiently to make the organisms visible.

2. For proof of true motility, look for directional movement that is several times the long dimension of the bacterium. The movement will also occur in different directions in the same field.

3. Ignore Brownian movement which is a vibrational movement caused by invisible molecules bombarding bacterial cells. If the only movement you see is vibrational and not directional, the organism is nonmotile.

4. If you see only a few cells exhibiting motility, consider the organism to be motile. Characteristically, only a few of the cells will be motile at a given moment.

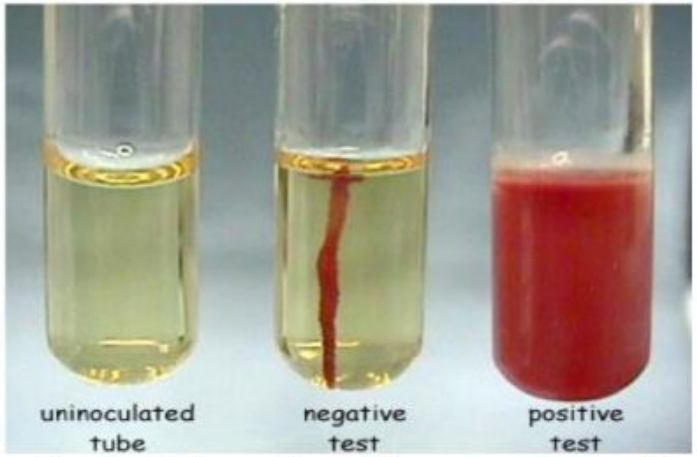
5. Don't confuse water current movements with true motility. Water currents are due to capillary action caused by temperature changes and drying out. All objects move in a straight line in one direction.

6. Always examine a wet mount immediately, once it has been prepared, because motility decreases with time after preparation.

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 0.4% Agar How to Perform Test: Using a sterile bacteriological needle, pick a colony of the test organism Stab quickly a tube of semi-solid media. (Avoid using bent needles). 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 se

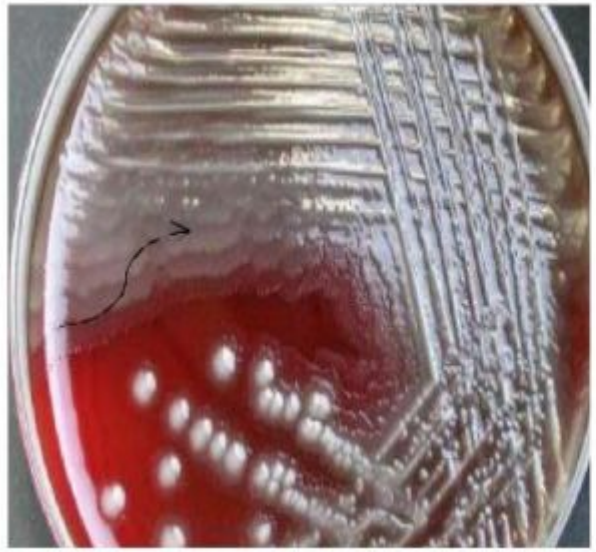


Semisolid media with tetrazolium chloride

Swarming Phenomenon;

When Proteus cells are inoculated on the surface of a suitable hard agar medium, they grow as short “vegetative” rods. After a certain time, however, cells start to differentiate at the colony margin into

long “swarmer” cells possessing up to 50 times more flagella per unit cell surface area. These swarmer cells migrate rapidly away from the colony until they stop and revert into the vegetative cell form




Safety first,
science always!