Indian Institute of Technology Delhi
Department of Biochemical Engineering and Biotechnology

I SEMESTER
BBL132 – GENERAL MICROBIOLOGY
LAB

EXPERIMENT # 12

AIM

To study the morphotypic diversity in environmental samples

BACKGROUND

Microbes grow on solid media as colonies. A colony is defined as a visible mass of microorganisms originating from a (single) mother cell, therefore a colony constitutes a clone of bacteria all genetically identical (except mutations that occur at low frequency). The number of cells within a colony can even reach a few billion. On a given medium, a colony's shape, colour, consistency, surface appearance and size - for a given incubation time - are often characteristic, and these features are often used in the identification of particular bacterial strains (Fig. 1).

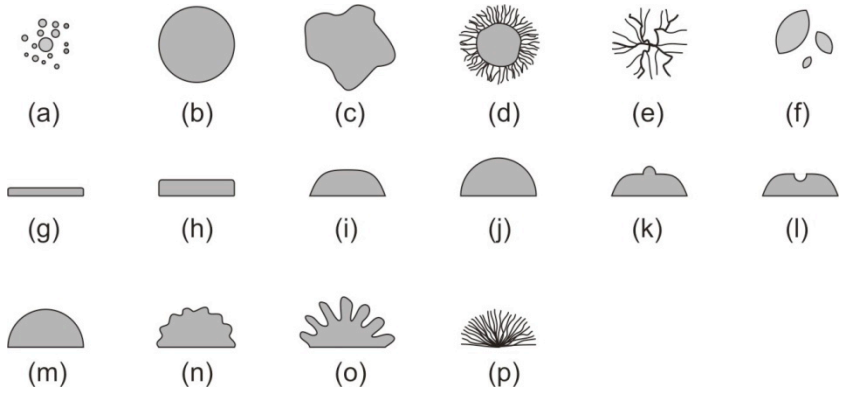


Fig. 1. Colony morphology of bacteria. Form: (a) punctiform (b) circular (c) irregular (d) filamentous (e) rhizoid (f) spindle. Elevation: (g) flat (h) raised (i) convex (j) pulvinate (k) umbonate (l) crateriform. Margin: (m) entire (n) undulate (o) lobate (p) filamentous.

Characteristics of cell morphology have great importance in the classification of bacteria using traditional taxonomical methods. Microorganisms cannot be identified solely by morphological characteristics, since bacterial cells can only be assigned to a limited number of categories

Bacteria are µm-sized organisms, where cell size is an important aspect of a thorough morphological characterization. The size and shape of the cells are usually determined following staining. The circumstances of culturing, the age of the culture and the physiological condition of bacterial cells can alter cell size and shape. According to their shape, bacteria can usually be identified as rods, cocci or spirals. An average rod-shaped bacterium is 2-5 µm long and 0.5-0.8 µm wide in diameter. The average diameter of a sphere-shaped bacterium is 0.8 µm. The size of some bacterial groups deviates from average values: spirochetes include some extremely thin (0.2 µm) bacteria, while there are some giants: *Thiomargarita namibiensis* (100-300 x 750 µm) and *Epulopiscium fishelsoni* (50 x 600 µm).

OBJECTIVE OF STUDY

Agar plates with colonies originating from an environmental sample

MATERIAL

Ruler, Magnifying glass, petridishes, saline, spreader, test tubes ,marker pen

PROCEDURE

1. Collect various environmental samples like water, soil in a sterile container from different location near to your lab

2. Dilute samples obtained from soil to 10^{-6} to 10^{-8} and water to 10^{-2} in saline buffer and plate them on Nutrient agar by spread plate method

3.To know the morphotypic diversity in air simply expose your petriplates at different location like under laminar, outside laminar, inside lab or out side of lab in corridor

4. Incubate the plates for 24 to 48hrs at 28 °C and 37 °C

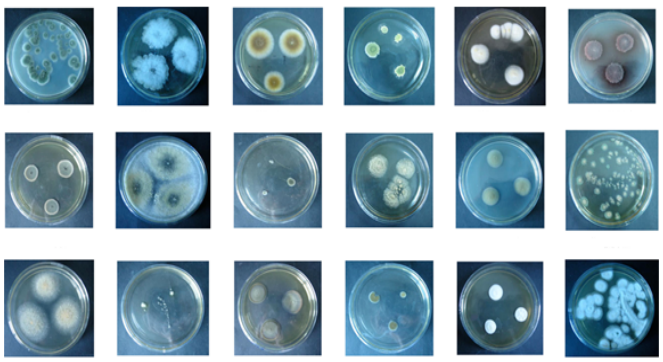
5. Select 5 different discrete colonies from the surface of a Petri plate and characterize them as follows:

- size of the colony (diameter in mm),
- shape or form of the colony (punctiform, circular, irregular, filamentous, rhizoid, spindle),
- elevation of the colony (flat, convex, pulvinate, umbonate, crateriform),

- margin of the colony (entire, undulate, lobate, filamentous),
- pigmentation of the colony (diffusible water-soluble or water-insoluble pigments),
- surface of the colony (smooth, glistening, rough, dull, wrinkled),
- density of colony (transparent - clear, opaque, translucent - almost clear, but distorted vision–like looking through frosted glass, iridescent - changes colour in reflected light),
- consistency of colony by touching it with an inoculating loop (butyrous, viscid - sticks to loop, hard to get off, brittle - dry, breaks apart, mucoid),
- Presence or absence of diffusible pigment in the medium around the colony.

RESULTS

Results will be observed as mentioned in the pictures and will be recorded in tabular form



Precautions

1. Use sterile containers for collecting your samples

2. Make proper dilution for samples obtained from soil and water

3. Avoid any contact with cultures