Macroscopic Appearance

Of Culture

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* Colony morphology in petri dish A colony is a large number of bacterial cells on solid medium, which is visible to the naked eye as a separate unit. colony is derived from one bacterial cell. Different species of bacteria can produce very different colonies. So in the identification of bacteria and fungi much Focus is placed on how the organism grows in or on media. colony have many feature as following:

1. Form – The form refers to the shape of the colony.







FILAMENTOUS



2. Size - The diameter of a representative colony may be measured. Tiny colonies are referred to as (pin-point) Punctiform.

3. surface- smooth, shiny, rough, dull, wrinkled.







#5M Michibel Brary are C MacConsol, Adams and Smith

4. *Texture* – Several terms that may be appropriate for describing the texture or consistency of bacterial growth are: dry, moist, mucoid, hard, viscous, butyrous (buttery).





Granular colony of *mycobacterium tuberculosis*

mucoid colonies of *Klebsiella pneumoniae* are indication of the bacterium's heavy capsule. **5.** *Color* – It is important to describe the color or pigment of the colony. such as: opaque, dull, translucent, iridescent (rainbow-like).



6. *Elevation* – This describes the "side view" of a colony. These are the most common.



7. *Margin* – The margin or edge of a colony (or any growth) may be an important characteristic in identifying an organisms.





Bacillus subtilis lobate colony

Appearance in test tube



• *Broth* :growth occurs throughout the container and can then present a dispersed, cloudy, or flaky appearance.



appearance in Semisolid media(slant) the medium is stabbed carefully in the center with an inoculating needle and later observed for the pattern of growth around the stab line.



Tubes containing pure cultures of *Escherichia coli* (white), Micrococcus luteus (yellow), and Serratia marcescens (red).



Affecting factor on growth

- 1. temperature:
- Psychrophile (0-20 °C)
- Mesophile (20-50 °C)
- Thermophile (above 45-80 °C)

2. pH : most microbes are neutrophiles, living around pH
7, a few microorganisms live at pH extremes .

3. Osmotic Pressure : most microbes exist under hypotonic or isotonic conditions, a few, called **osmophiles**, requires high concentrations of salt; these organisms are called **halophiles**.

4. Gas Requirements:



- Aerobe (aerobic organism) can use gaseous oxygen in its metabolism.
- **Obligate aerobe :**is organism that cannot grow without oxygen .
- **Facultative anaerobe** is an aerobe that does not require oxygen for its metabolism and is capable of growth in the absence of it.
- **Microaerophile** does not grow at normal atmospheric concentrations of oxygen but requires a small amount of it.
- **Obligate anaerobes** : they cannot tolerate any free oxygen in the immediate environment and will die if exposed to it.
- Capnophiles grow best at higher CO2 tensions.

Culturing of anaerobic bacteria:

anaerobic bacteria need special handling and incubation and identification, usually requires special media for culturing.

• There are commercially available anaerobic transport media in vials and tubes with anaerobic atmosphere and bag systems that act by removing molecular

oxygen.



• Culturing of anaerobic bacteria should be made in anaerobic chamber.



• Incubation the streaked plates or test tubes inside gas jar then incubated in special incubator.



Bacterial growth on media

There are many types of media which divide according to physical form ,chemical composition and ,functional type. The type of media is important in identification of bacteria, and in our lecture will explain some an important type.

• **SIM agar (sulfur indole motility agar)** used to detect bacteria that H2S gas producers; indole formation and motility.



• MacConkey agar (MAC) isolation of gramnegative bacteria.



MacConkey agar differentiates between lactose-fermenting bacteria (indicated by a pink-red reaction in the center of the colony) and lactose-negative bacteria (indicated by an off-white colony with no dye reaction).

- Salmonella/Shigella (SS agar) Isolation of Salmonella and Shigella.
- Triple-sugar iron :used to detect bacteria that ferment sugars and produce H2S(black). Such as Enterobacteriacea.



• Mannitol salt agar Isolation of Staphylococcus aureus. pink turn to yellowo.



 Blood agar :have intact red types of RBC (5%sheep blood)used to differentiate between bacteria that cause blood cells damage (hemolysis).bacteria that secrete toxins cause 3 types of blood hemolysis α,β,γ.





 γ The RBC undamaged hemolysis appear



α is weak hemolysis appear as gray zone around colony.

Chocolate agar: It is a different of the blood agar plate, containing red blood cells that have been lysed by slowly heating to 80 °C. Chocolate agar is used for growing fastidious respiratory bacteria, such as *Haemophilus influenzae* and *Neisseria meningitidis*. Bacteria feed on V and X factor which are inside red blood cells.