

Colony Morphology & Identification



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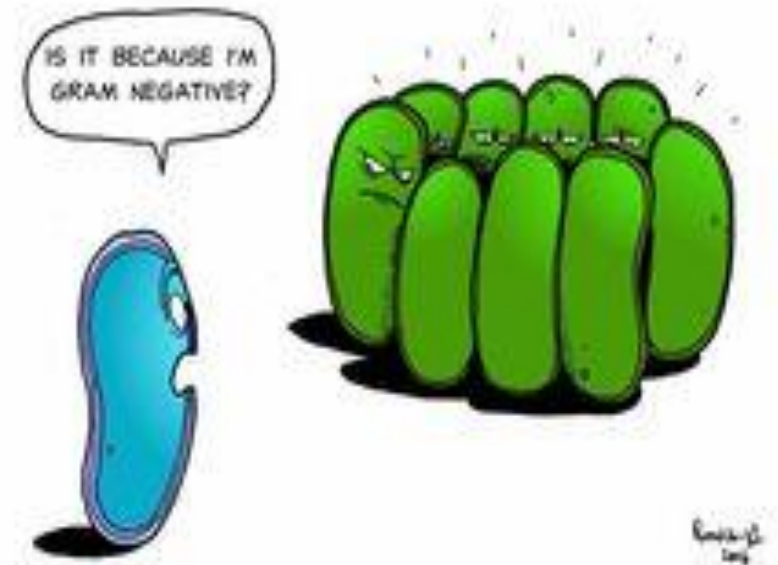


Lab. No. 8

Objectives

Methods used in bacterial identification :

1. Microscopic appearance
2. Cultural characters
 - a. Colony morphology
 - b. Haemolysis
3. Biochemical test
4. Serological test
5. Polymerase chain reaction(PCR)



Identification

- ❖ Accurate and definitive microorganism identification, including bacterial identification and pathogen detection, is essential for correct disease diagnosis, treatment of infection
- ❖ Scientists have devised an array of tests that help to identify bacteria. These tests range in complexity from viewing bacteria under a microscope to sequencing DNA



1- MICROSCOPIC APPEARANCE:

The use of a microscope is a very important skill for a microbiologist.

Microscopic methods used for unstained and stained preparation commonly used in the study of microorganisms.



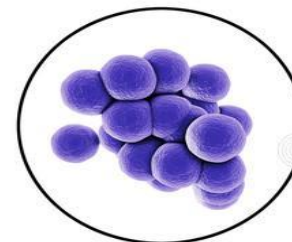
Unstained preparation

- 1- Wet Mount
- 2- Hanging drop

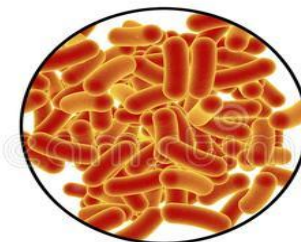
Stained preparation

- 1- Differential stain
- 2- Special stain

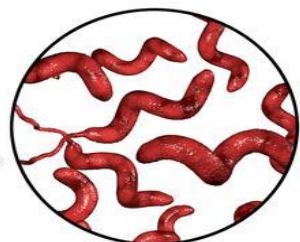
SHAPES OF BACTERIA



Spherical



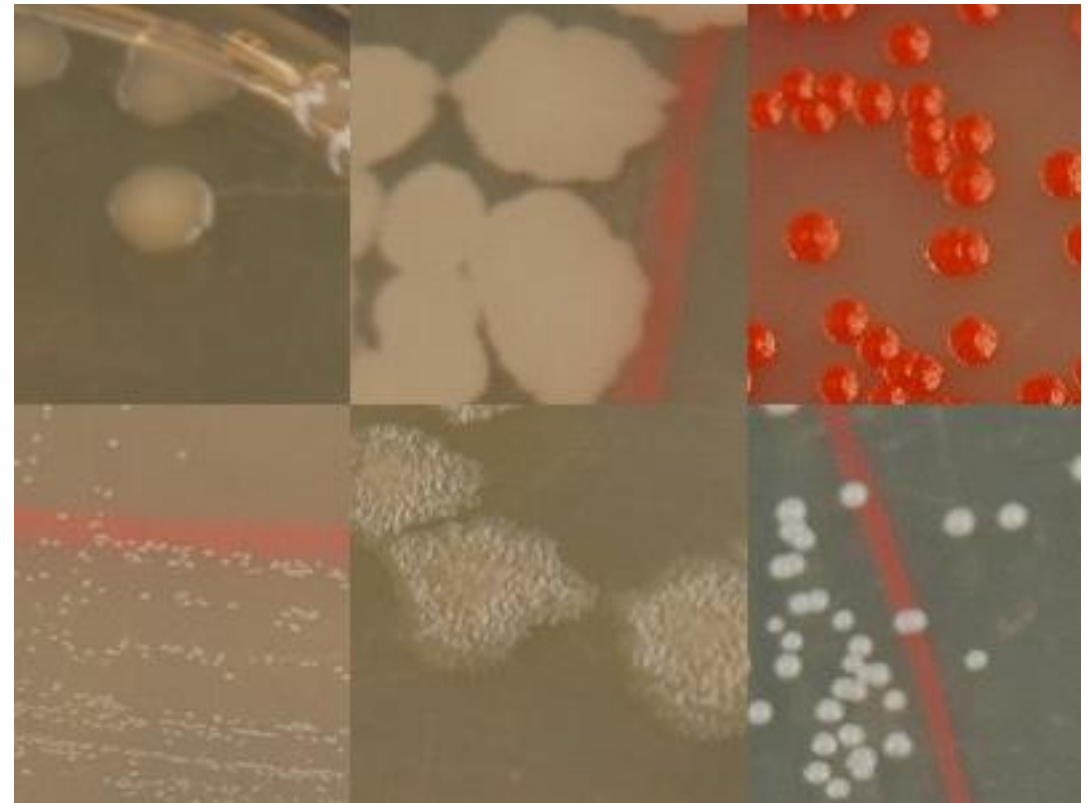
Rod-like



Spiral

2- Cultural characters:

The **macroscopic appearance** of colonies of bacteria can also be used to identify bacteria (e.g: **hemolytic** properties on agar containing blood, **pigmentation** of the colonies, **size and shape** of the colonies



A. Colony Morphology





















- Bacteria grow on solid media as **colonies**. A colony is defined as a **visible mass** of microorganisms all originating from a single mother cell.
- Each bacteria has a special colony morphology on solid media.
- The **commonly observed colonial characteristics** are helpful in making a preliminary bacterial identification.
- These **cannot be used as the single criteria for identification.**

What is the difference between colony morphology and cellular morphology?

Morphology of colonies can be defined as their color, shape, edge and elevation. ... However, **cellular morphology** shows the **difference** of the individual cells that is seen under the microscope. **Cellular morphology** of a **cell** can be cocci, bacilli, spiral etc

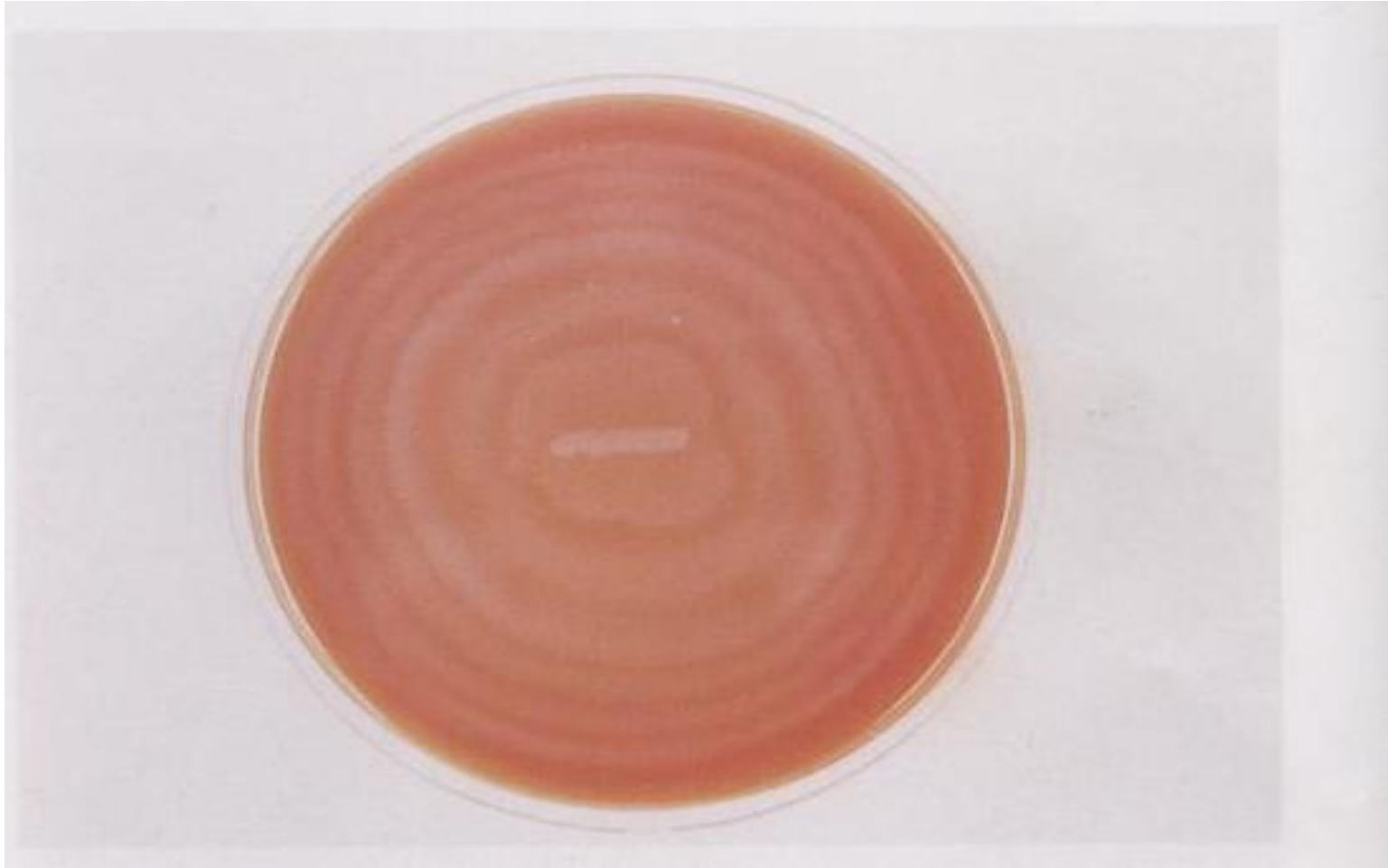


Following are a number of terms used to describe colonial morphology:

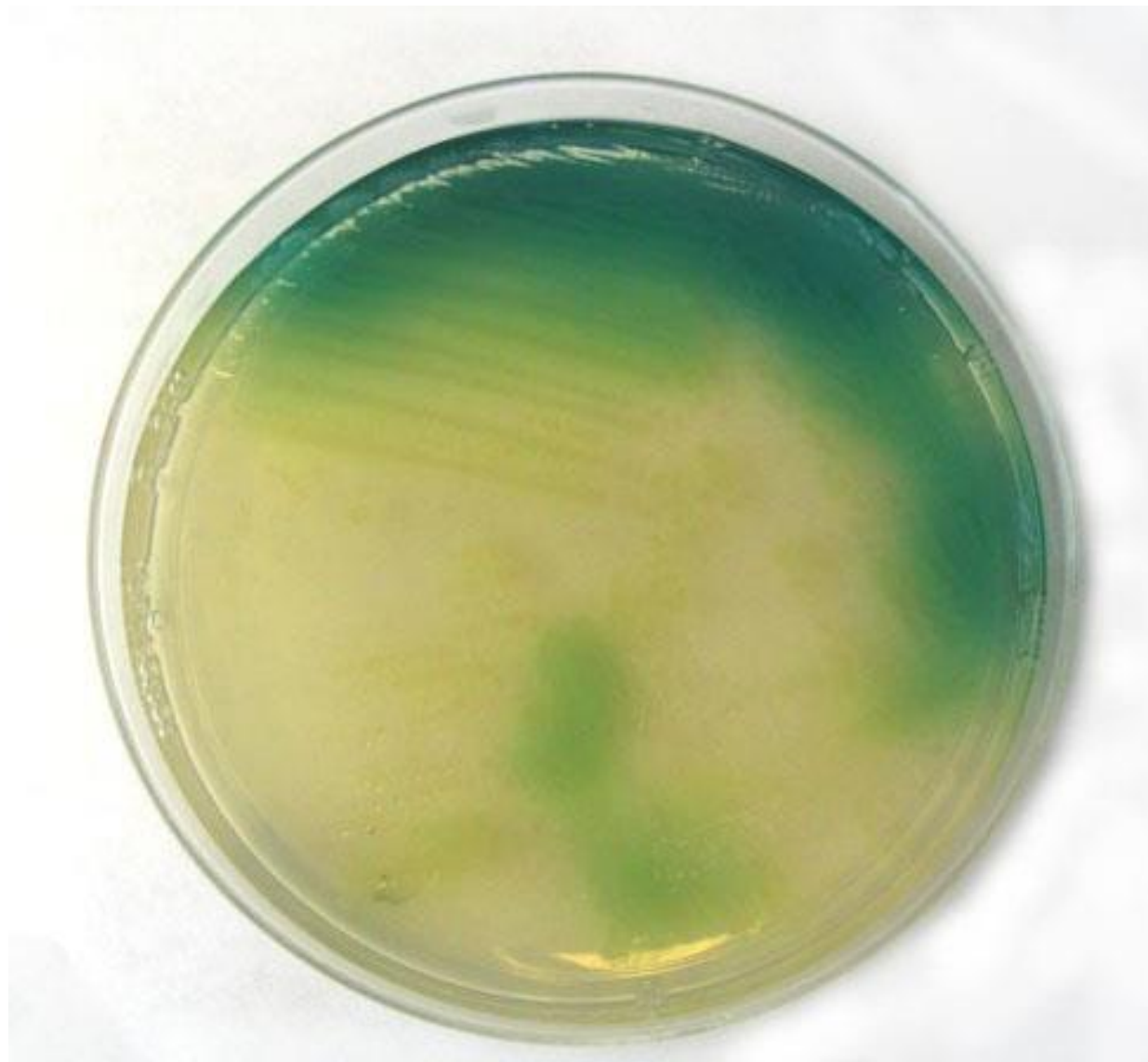
Shape	 Circular	 Rhizoid	 Irregular	 Filamentous	 Spindle	
Margin	 Entire	 Undulate	 Lobate	 Curled	 Rhizoid	 Filamentous
Elevation	 Flat	 Raised	 Convex	 Pulvinate	 Umbonate	
Size	 Punctiform	 Small	 Moderate	 Large		
Texture	Smooth or rough					
Appearance	Glistening (shiny) or dull					
Pigmentation	Nonpigmented (e.g., cream, tan, white) Pigmented (e.g., purple, red, yellow)					
Optical property	Opaque, translucent, transparent					



Bacillus subtilis



Proteus spp (Swarming)



Pseudomonas aeruginosa (Pigmentation)

B. Haemolysis

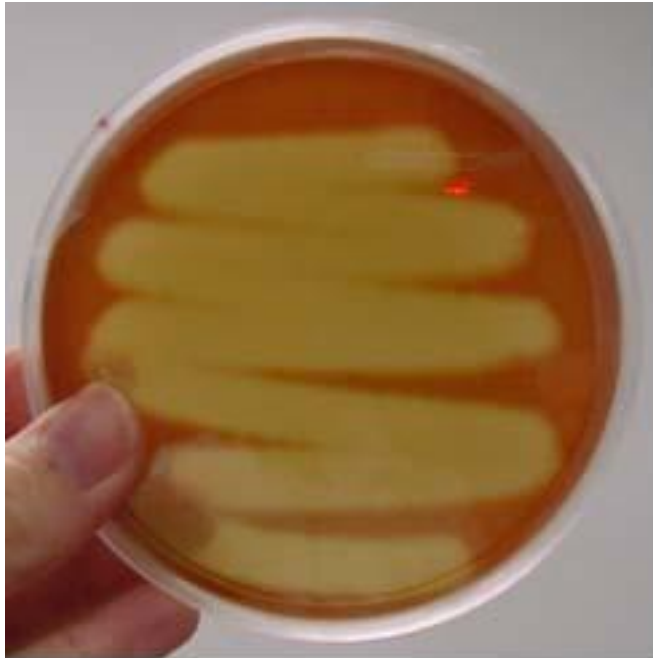
Is the breakdown of red blood cells and the ability of bacteria colonies to induce hemolysis is used to classify certain microorganisms this is particularly useful in classifying *Streptococcus spp* .

Alpha: partial clearing of blood around colonies with green discoloration of the medium.

Beta: zone of complete clearing of blood around colonies due to lyses of the RBC.

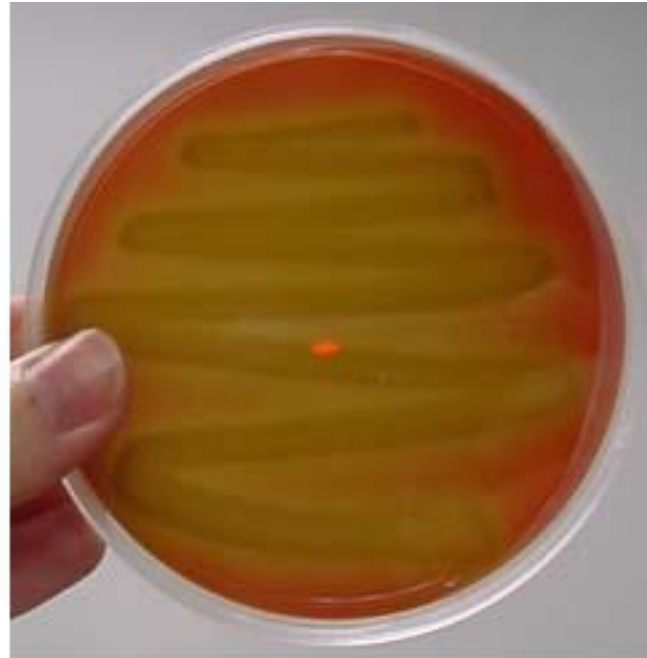
Gamma: no change in the medium around the colony; no lyses of the RBC.

Types of haemolysis



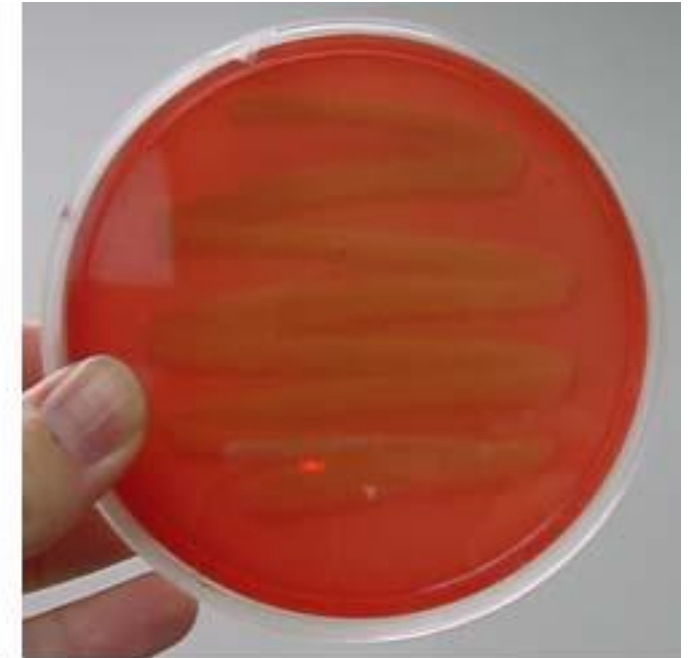
Beta Hemolysis

Complete lysis of RBC



Alpha Hemolysis

The agar is dark & greenish



Gamma Hemolysis

Unchanged no hemolysis

3. Biochemical tests :-

Biochemical tests: Are tests that identify the bacteria on the basis of the presence of **certain enzymes** and other biochemical properties. biochemical reactions are used for accurate identification

- Kligler's Iron Agar (KIA)
- Triple sugar iron
- IMViC test
- Oxidase test
- Catalase test
- Urease test

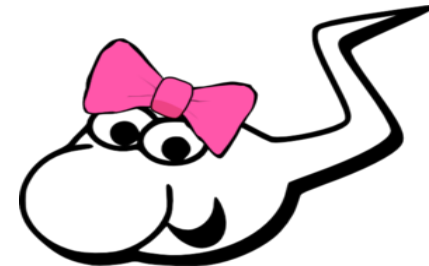


4. Serological test :

- Identifying and quantifying the specific Ab found in serum of infected patient in early and late stages of infection.
- Detecting and identifying an organisms in a specimens by its surface Ag or by the soluble Ag it produces.
- Some serological tests are not limited to blood serum, but can also be performed on other bodily fluids such as [semen](#) and [saliva](#).

Serological techniques:

- Latex agglutination (LA),
- Complement fixation (CF)
- Enzyme-linked immuno-assay (ELISA)
- Fluorescent antibody (FA)



5. Polymerase Chain Reaction (PCR)

PCR is a means to amplify a particular piece of DNA

Amplify: making numerous copies of a segment of DNA

PCR can make billions of copies of a target sequence of DNA in a few hours



References

- Luis M. de la Maza, Marie T. Pezzlo, Cassiana E. Bittencourt, Ellena M. Peterson. **2020. Color Atlas of Medical Bacteriology.** (2020, Wiley) - libgen.lc.
- Gary W. Procop, Deirdre L. Church , et al. **2017. Koneman's Color Atlas and Textbook of Diagnostic Microbiology. 7th Edition.** Jones & Bartlett Learning