

# bacterial identification

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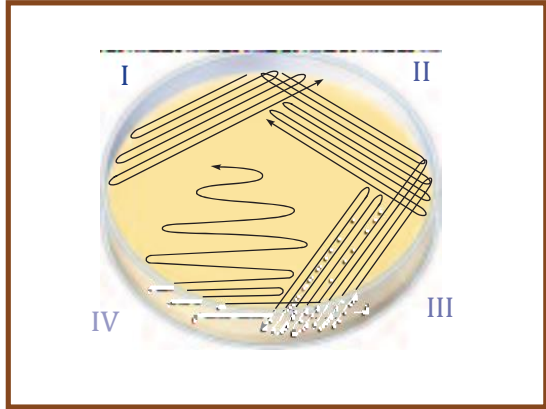
# Step 1.

- ▶ **Samples** of body fluids (e.g. blood, urine, cerebrospinal fluid) are streaked on culture plates and isolated colonies of bacteria (which are visible to the naked eye) appear after incubation for one - several days .
- ▶ Recognizing different bacterial growth morphologies on agar plates is a useful and often crucial step in the identification process.
- ▶ Each **colony** consists of millions of bacterial cells. Observation of these colonies for **size, texture, color,** and (if grown on blood agar) **hemolysis** reactions, is highly important as a first step in bacterial identification.

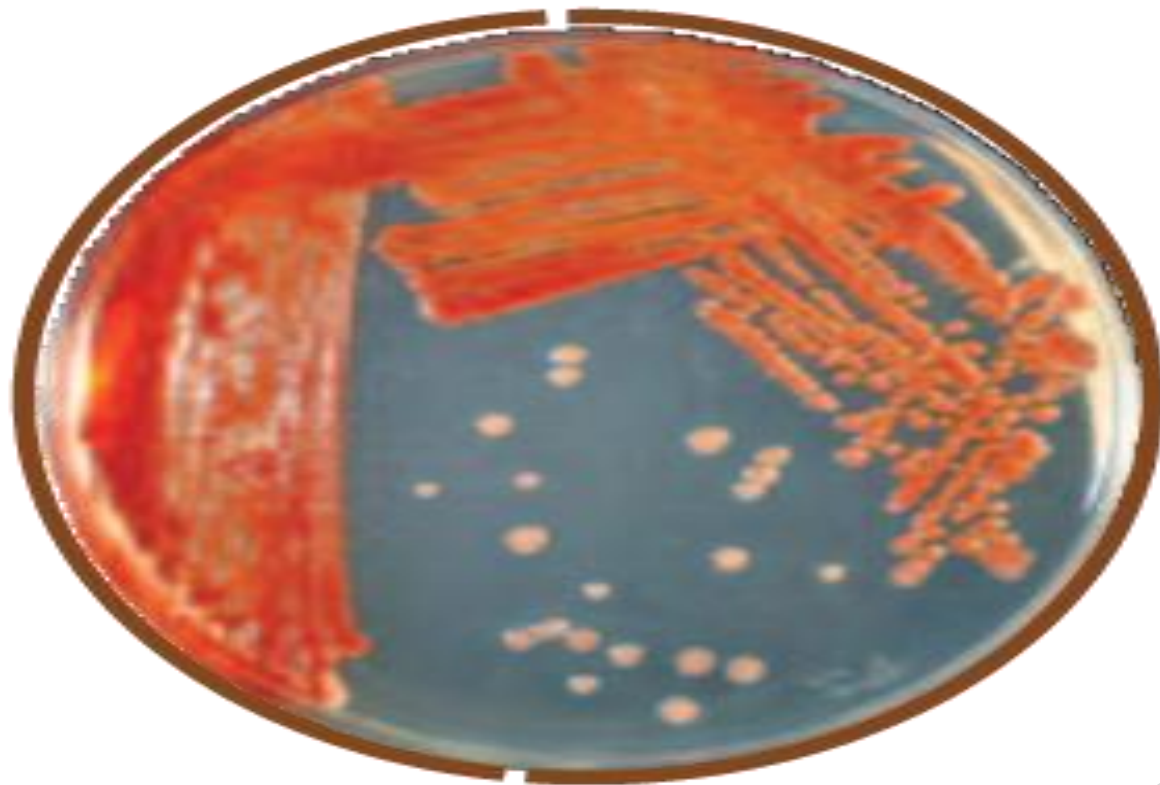
# Streak Plate Methods of Isolation

- ▶ The identification process of an unknown microbe relies on obtaining a pure culture of that organism. The streak plate method produces individual colonies on an agar plate. A portion of an isolated colony then may be transferred to a sterile medium to start a pure culture.

**STREAKING A PLATE** Hold the plate comfortably and streak with the edge of the loop. Be careful not to cut the agar.

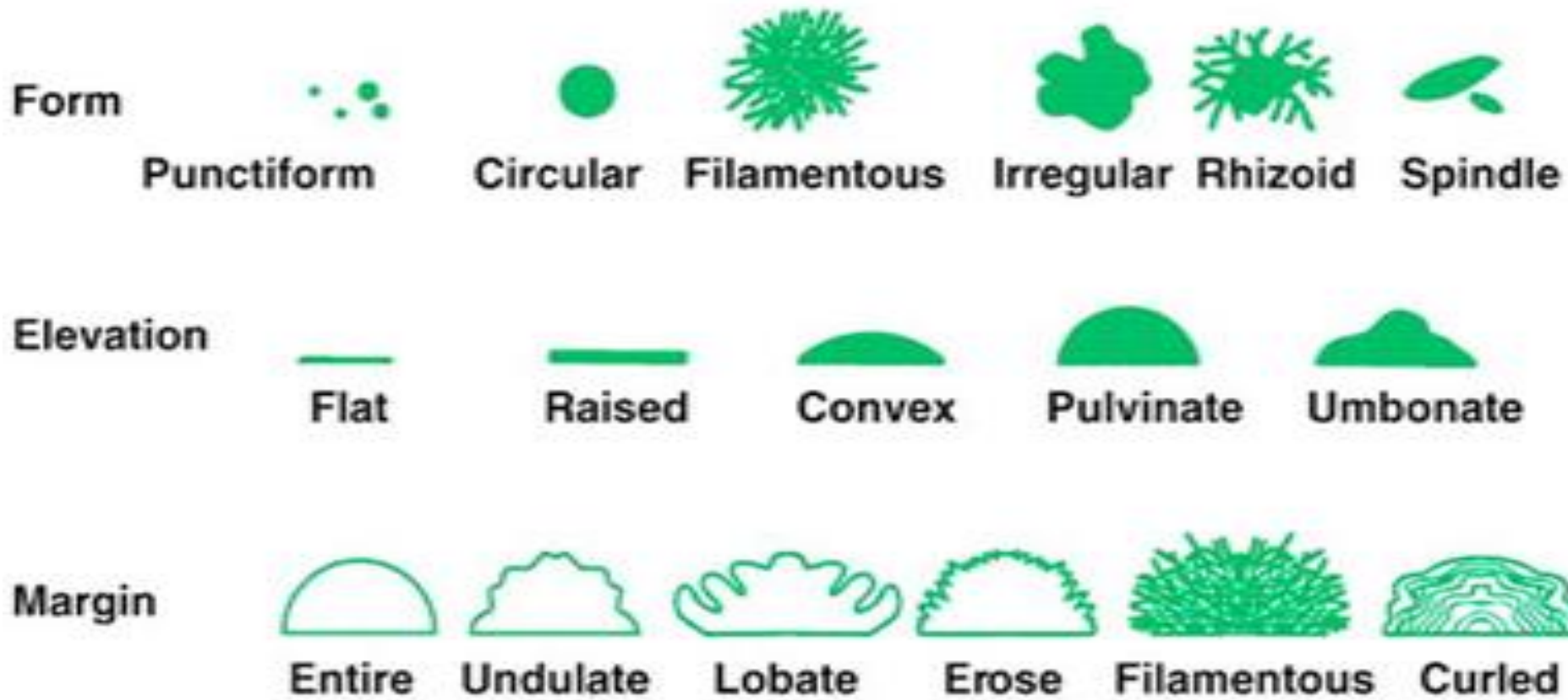


the decreasing density of growth in the four streak patterns. On this plate, isolation is first obtained in the fourth streak. A portion of an individual colony may be transferred to a sterile medium to start a pure culture.

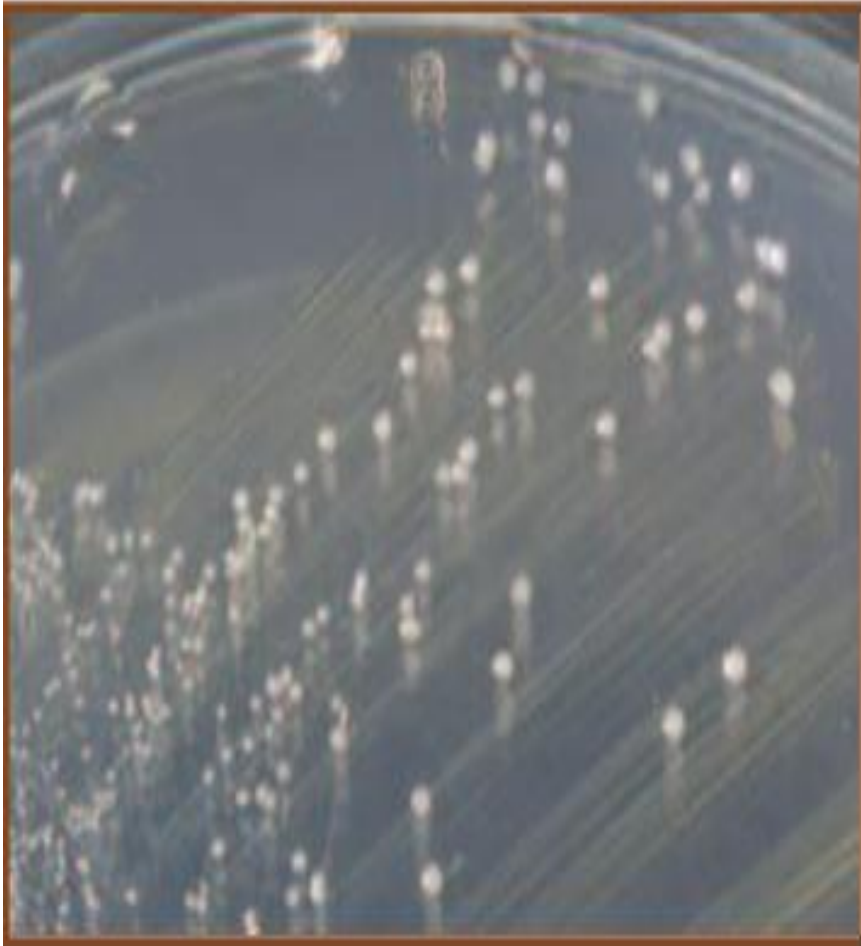


The basic categories of growth include colony shape, margin (edge), elevation, color, and texture .

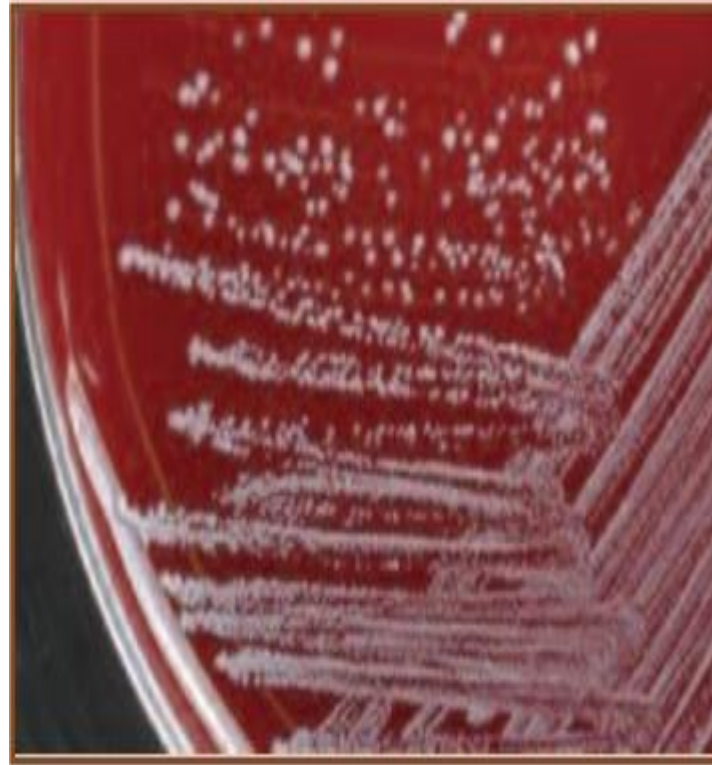
- ▶ **Colony shape** may be described as circular, irregular, or (tiny).
- ▶ The margin may be entire (smooth, with no irregularities), wavy , lobate (lobed), filamentous, or rhizoid (branched like roots).
- ▶ **Colony elevations** include flat, raised, convex, or (very convex), and umbonate (raised in the center).
  
- ▶ **Colony texture** may be moist, mucoid, or dry.
- ▶ **Pigment** production is another useful characteristic and may be combined with **optical properties** such as opaque, translucent, shiny, or dull.



(a)

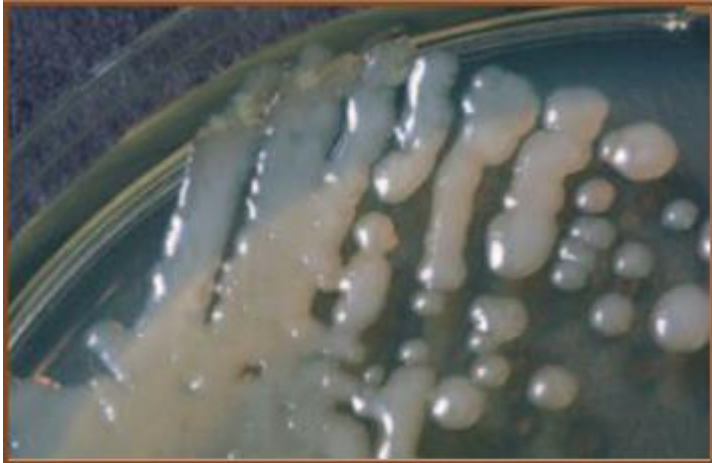


**ENTEROCOCCUS FAECIUM** GROWN ON NUTRIENT AGAR The colonies are white, circular, convex, smooth, is found in human and animal feces.



**STAPHYLOCOCCUS EPIDERMIDIS** GROWN ON SHEEP BLOOD AGAR The colonies are white, raised, circular, *S. epidermidis* is an opportunistic pathogen.





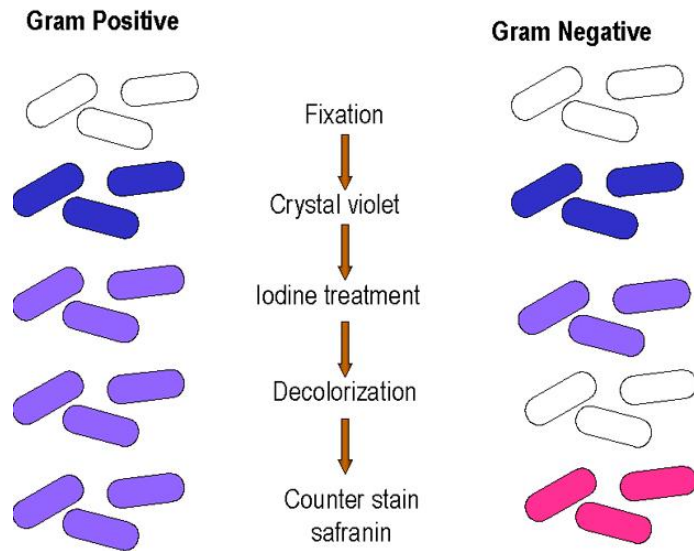
**KLEBSIELLA PNEUMONIAE GROWN ON NUTRIENT AGAR** The colonies are mucoid, raised, and shiny. While it is a normal inhabitant of the human intestinal tract, it is associated with community-acquired pneumonia and nosocomial urinary tract infections.



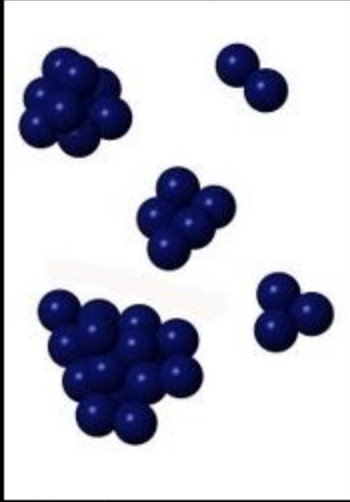
**MUCOID COLONIES** *Pseudomonas aeruginosa* grown on Endo agar a mucoid texture. *P. aeruginosa* is found in soil and water, and can cause infections in burn patients.

## Step 2.

- ▶ Colonies are Gram stained and individual bacterial cells observed under the microscope.



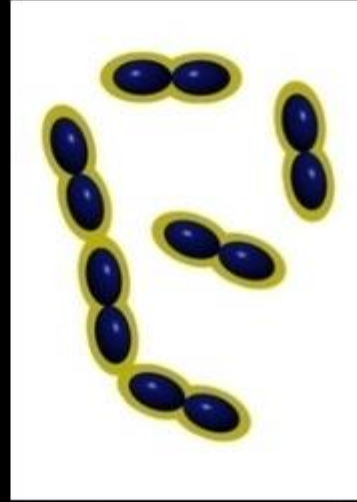
GRAM - POSITIVE



*Staphylococcus aureus*



*Streptococcus agalactiae*

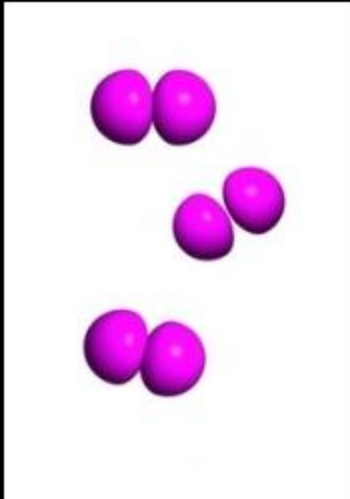


*Streptococcus pneumoniae*



*Listeria monocytogenes*

GRAM - NEGATIVE



*Neisseria meningitidis*



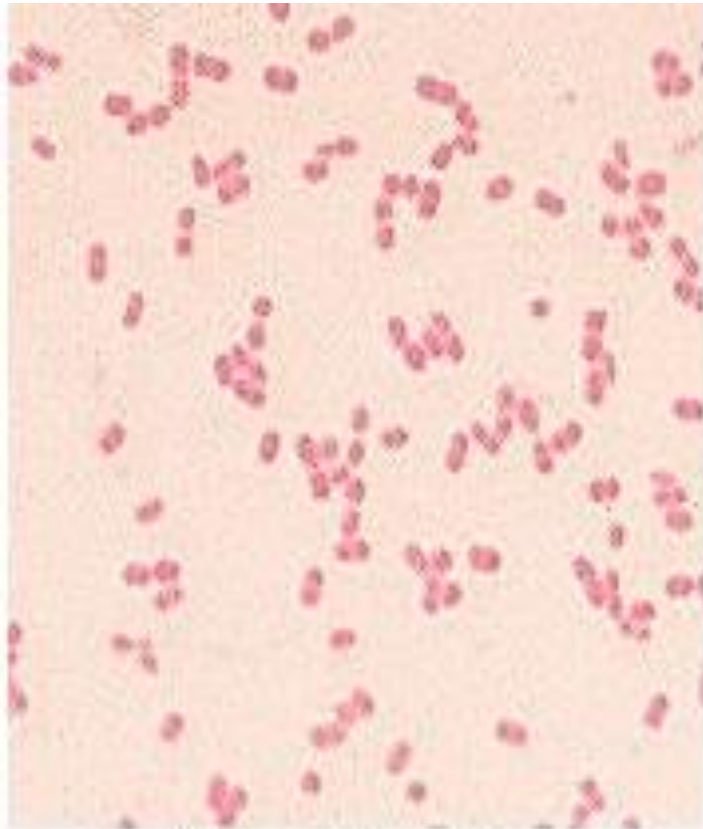
*Haemophilus influenzae*



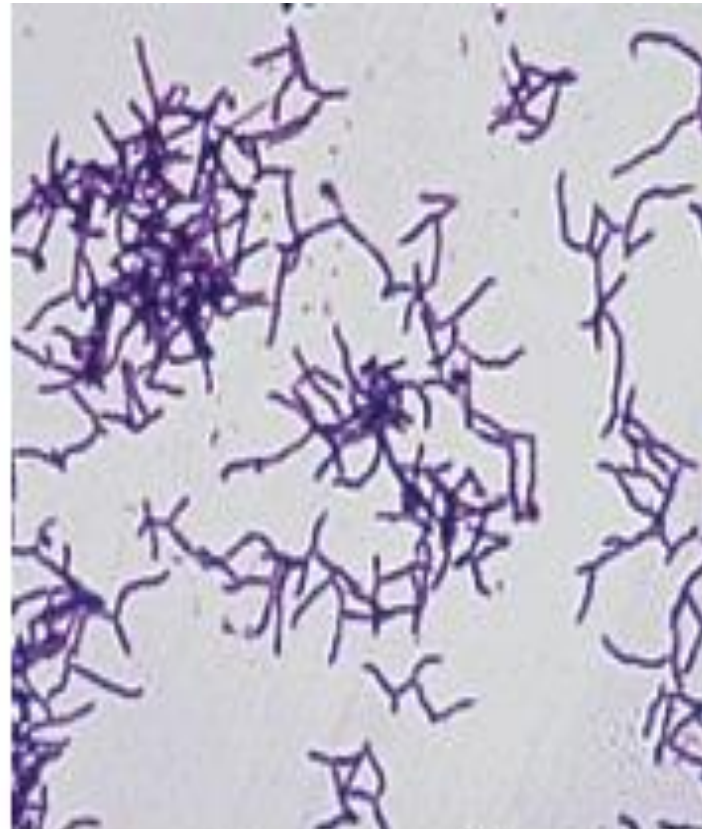
*Klebsiella pneumoniae*



*Escherichia coli*



Gram Negative



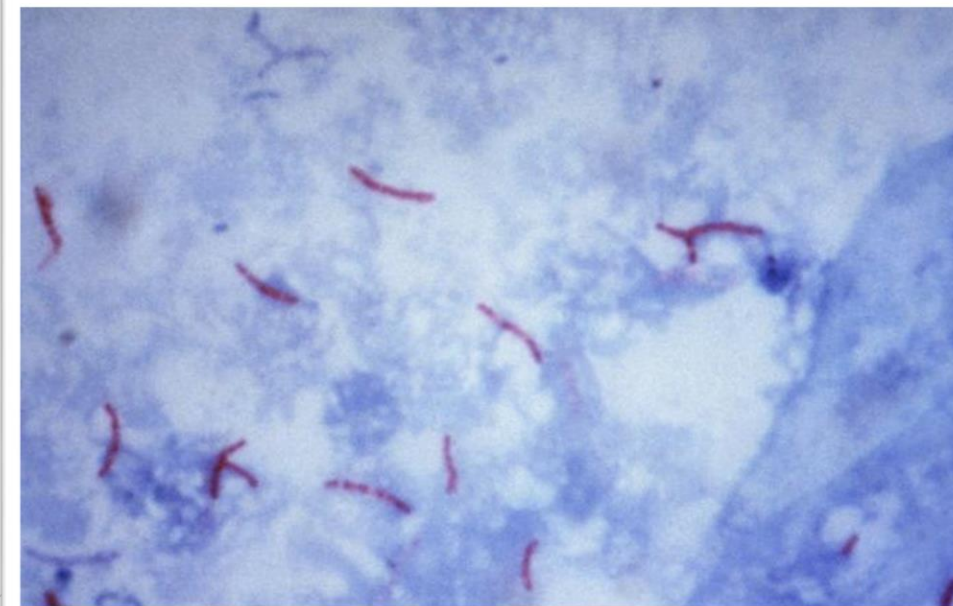
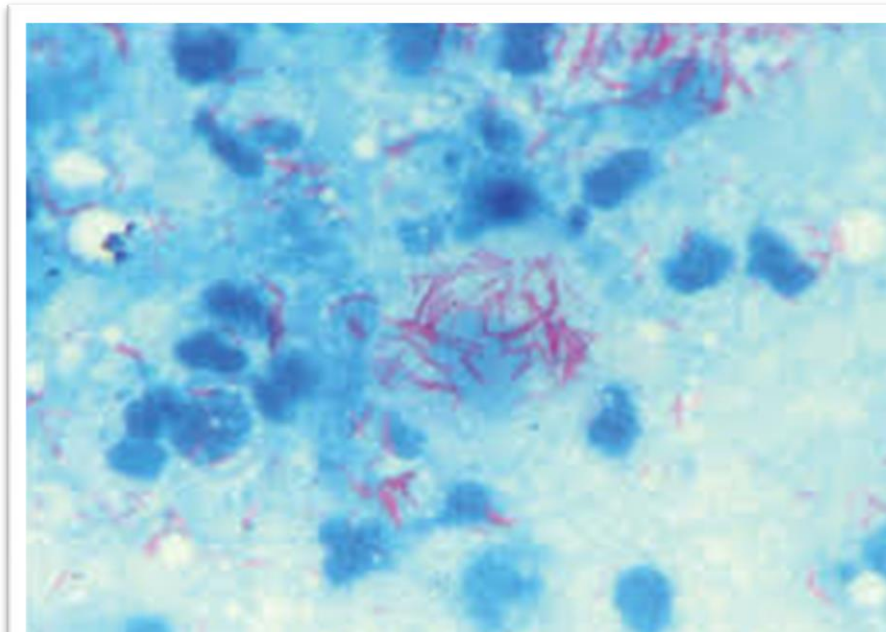
Gram Positive

# Approaches to rapid diagnosis without prior culture

- ▶ Certain human pathogens (including the causative agents of tuberculosis, Lyme disease and syphilis) either cannot be isolated in the laboratory or grow extremely poorly.
- ▶ Successful isolation can be slow and in some instances impossible.
- ▶ Direct detection of bacteria without culture is possible in some cases.

# Rapid diagnosis

- ▶ **Direct microscopic observation** of certain clinical samples for the presence of bacteria can be helpful (e.g. detection of *M. tuberculosis* in sputum).



Thank You!

