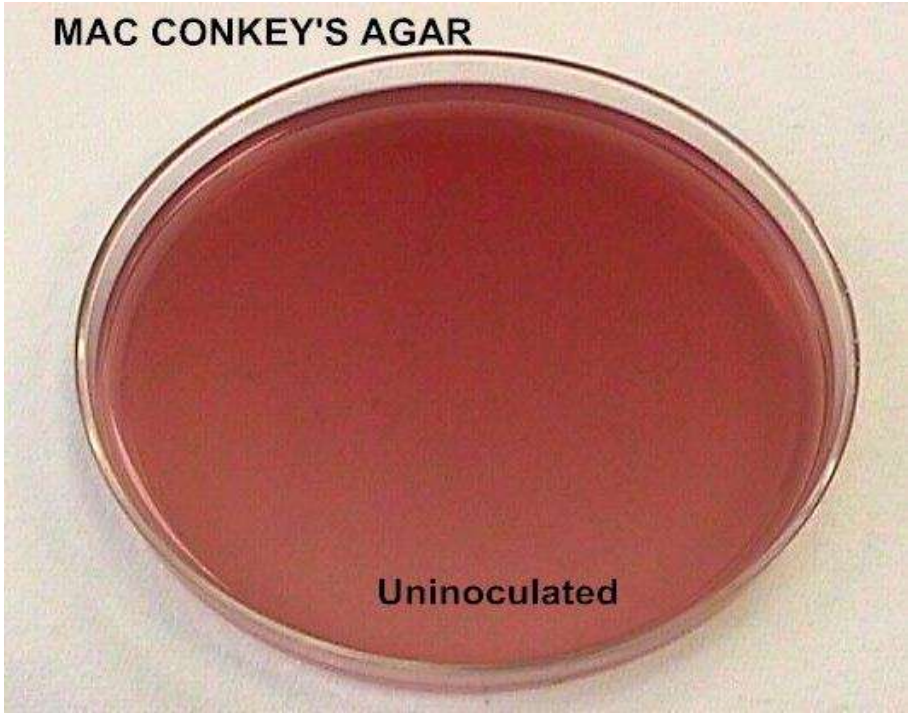
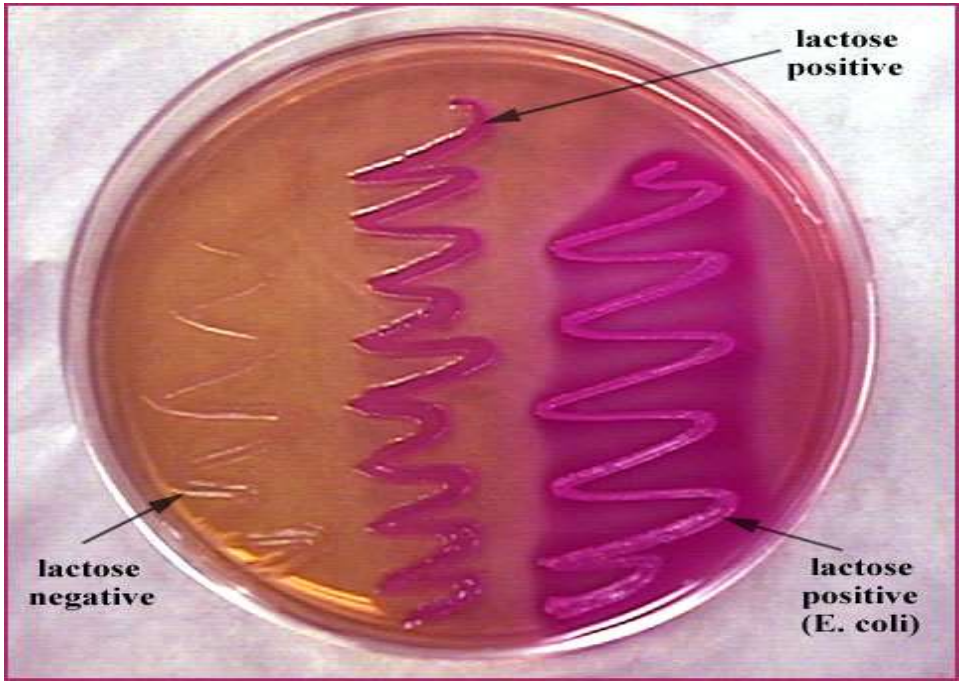
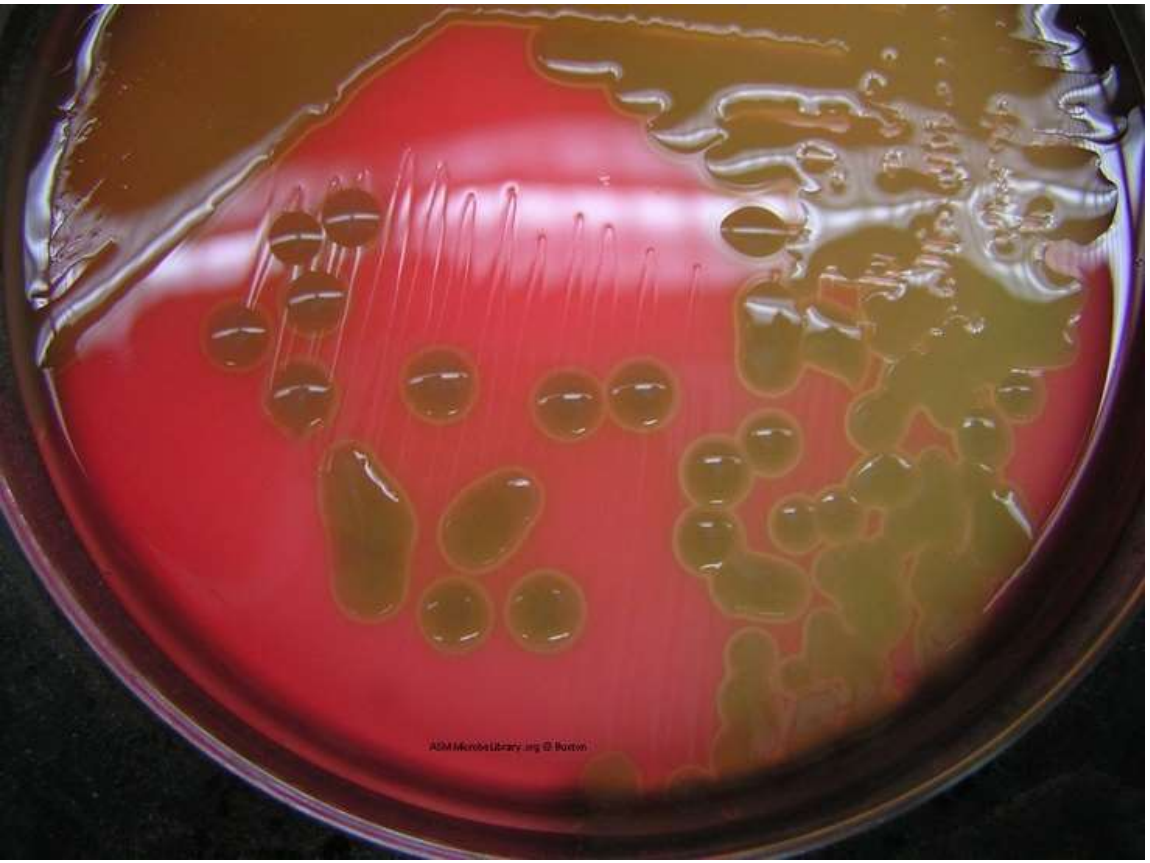

Lactose fermenter
Non Lactose fermenter


MAC CONKEY'S AGAR
Uninoculated


lactose positive
lactose negative
lactose positive (E. coli)

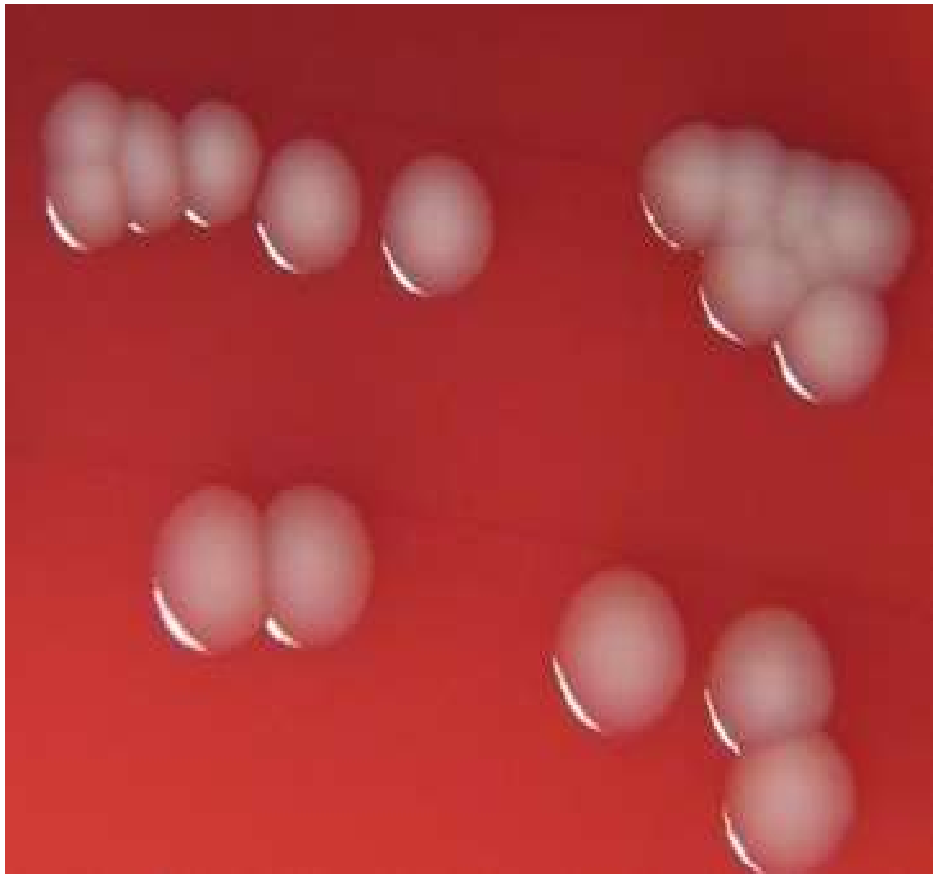
The three types of hemolysis



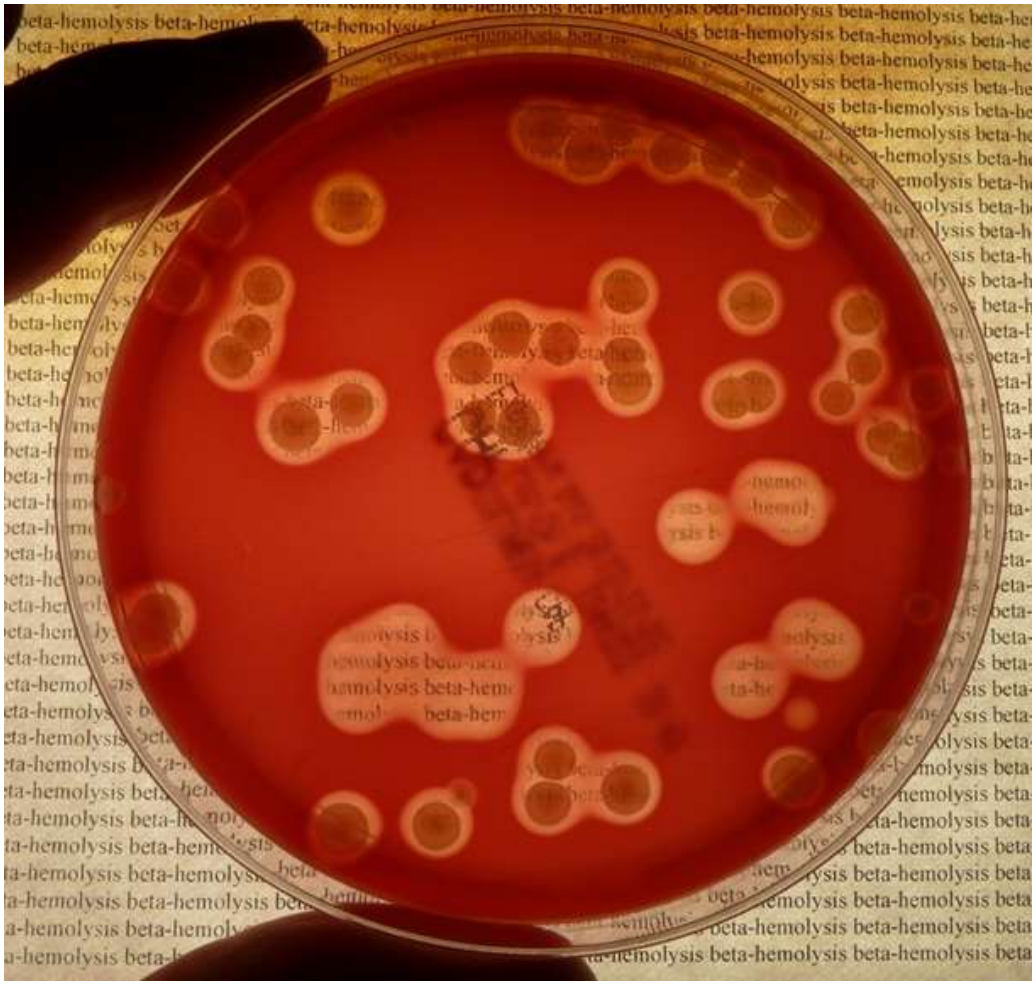
Alpha hemolysis
Incomplete (partial) lysis of RBCs

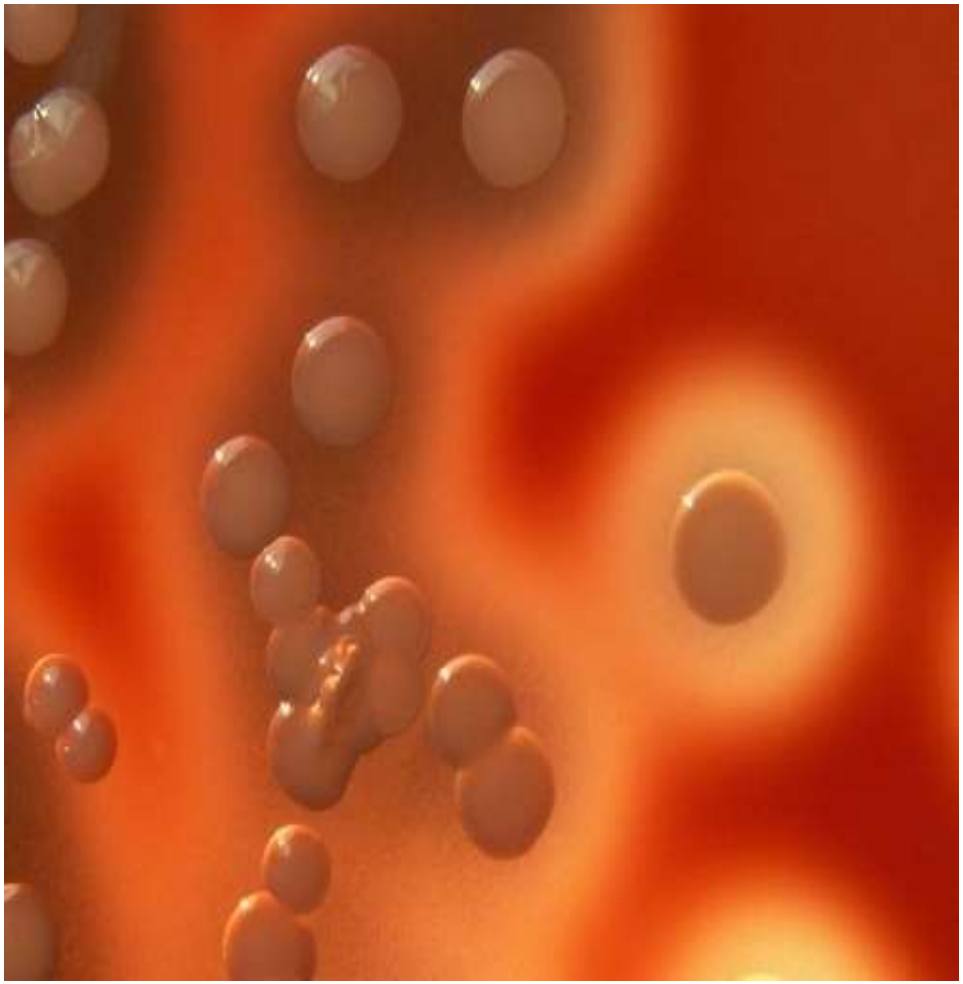
Gamma hemolysis

No hemolysis, and no change in the medium



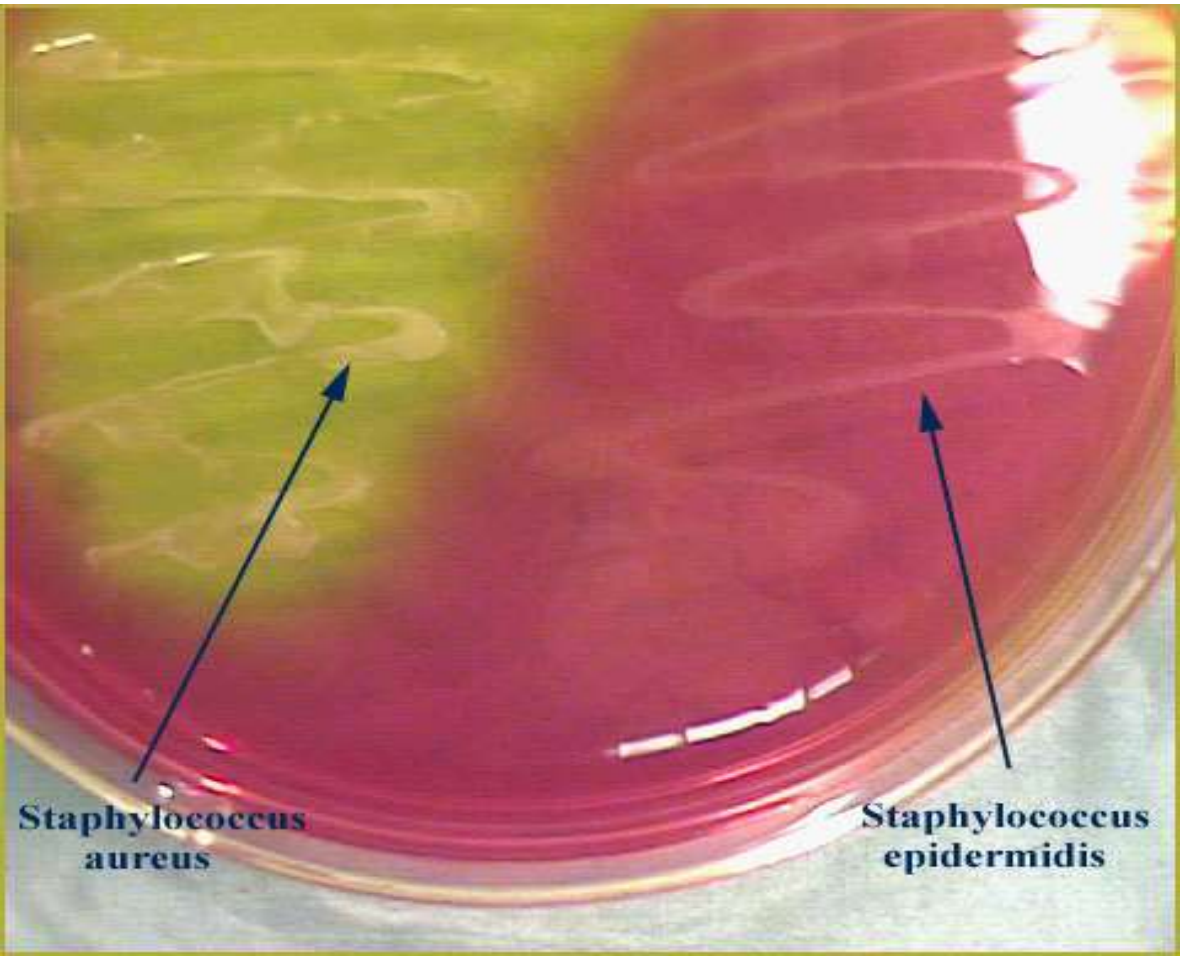
Beta hemolysis = Complete hemolysis





Beta hemolytic Staphylococci

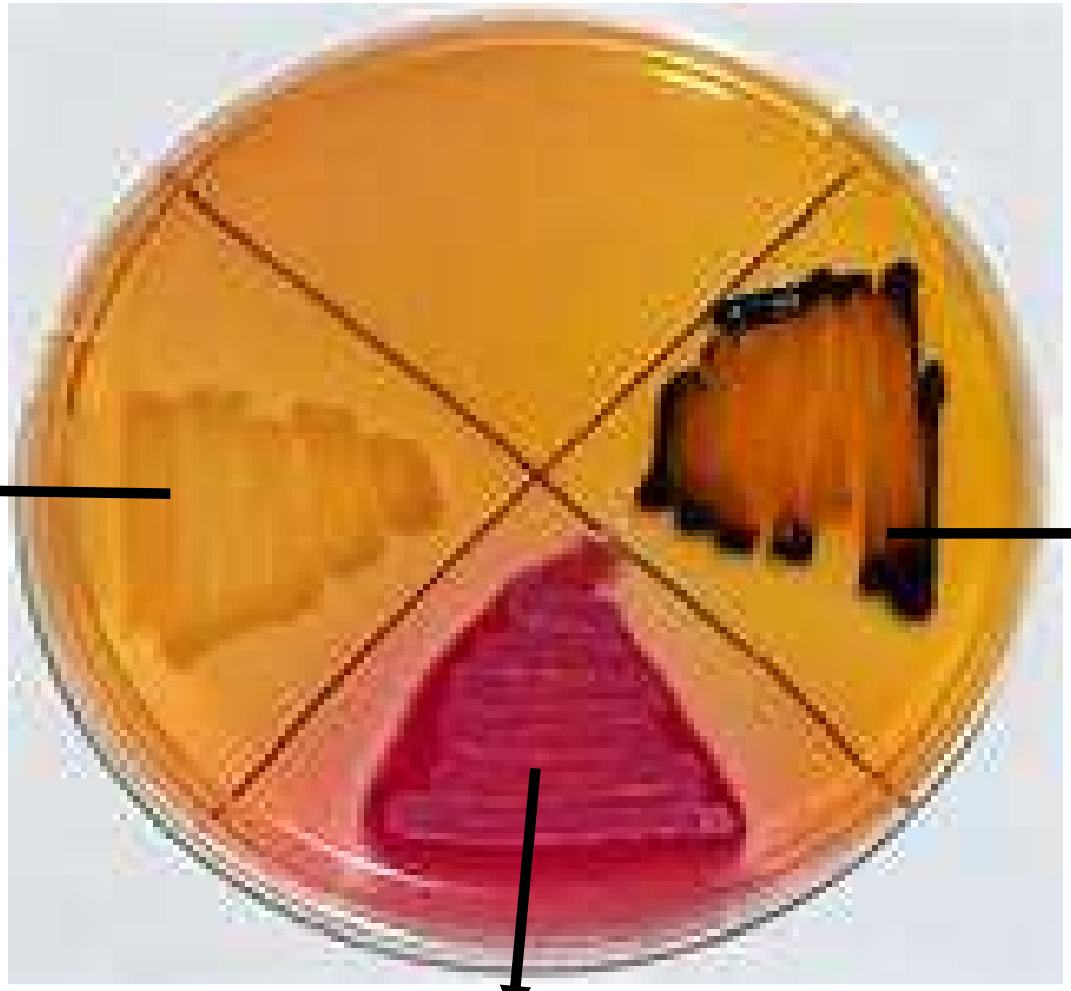
S. aureus


Staphylococcus aureus
Staphylococcus epidermidis


Salmonella


©
www.microbiologyinpictures.com
Salmonella enterica subsp. enterica

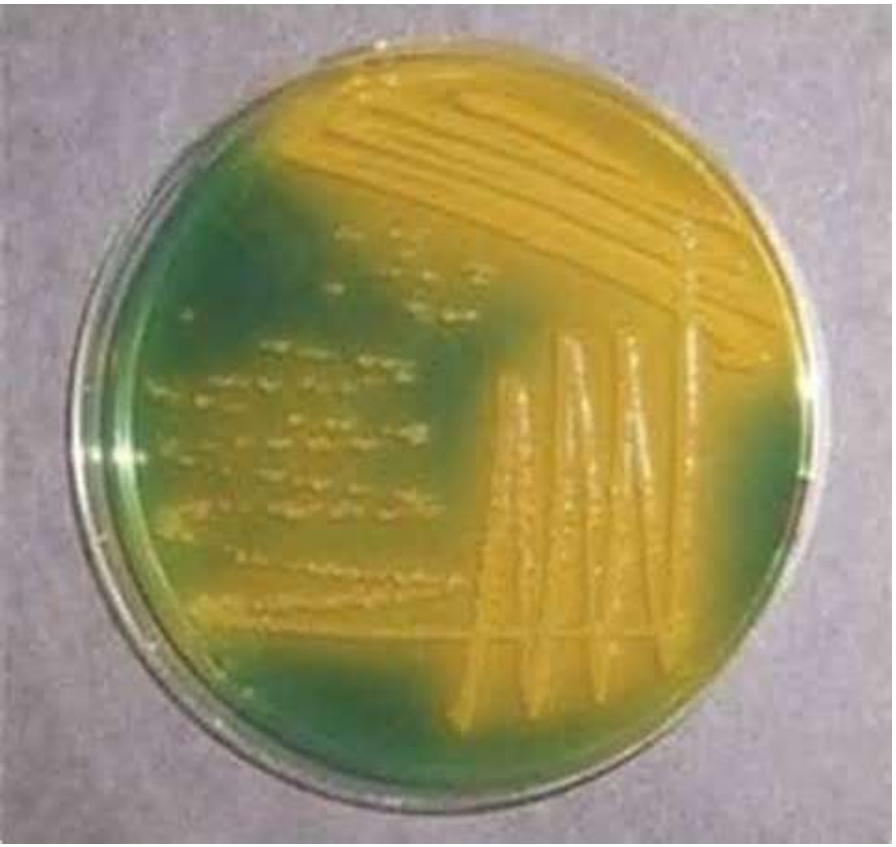
Shigella: colorless colonies without black centers



Salmonella: colorless colonies with black centers

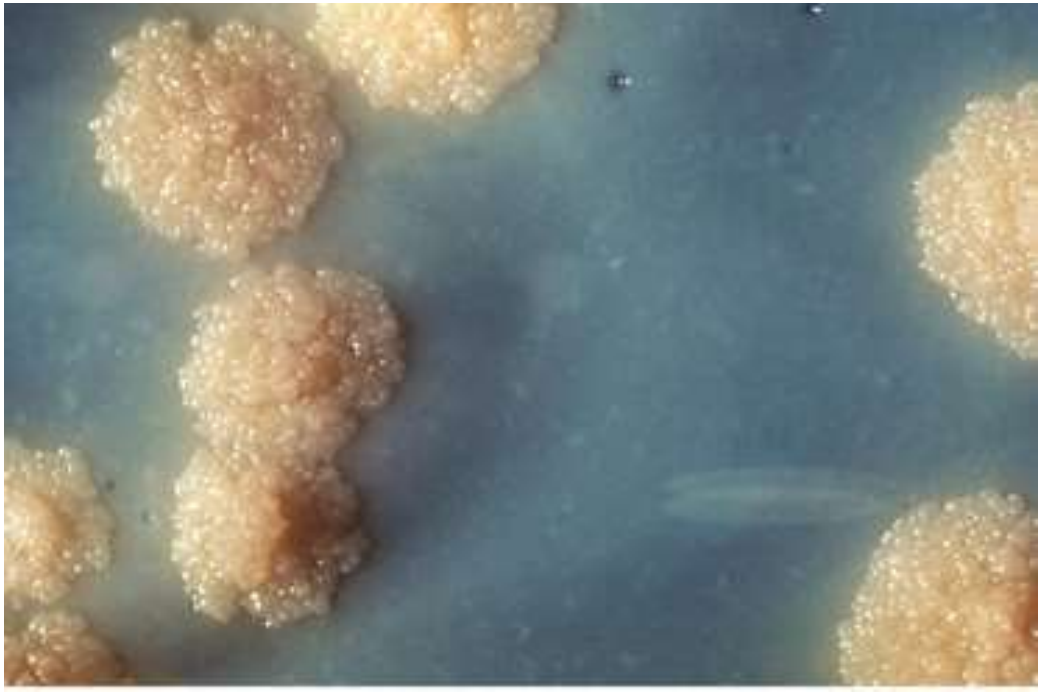
Lactose fermenter flora: pink to red colonies

✓Vibrio parahemolyticus non-sucrose fermenter, green colonies



✓Vibrio cholera
Ferment sucrose smooth yellow colonies


T.E.B.S



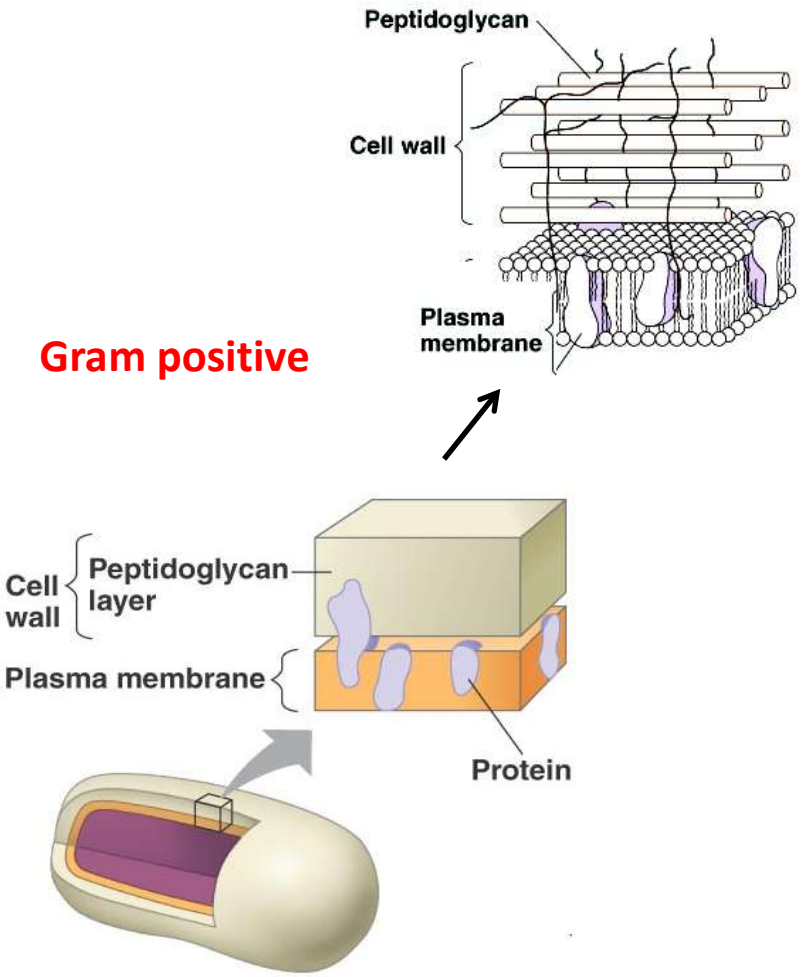
Mycobacterium tuberculosis produces rough
and tough colonies

Haemophilus influenzae growth on Chocolate agar

Gram positive vs. Gram negative bacteria

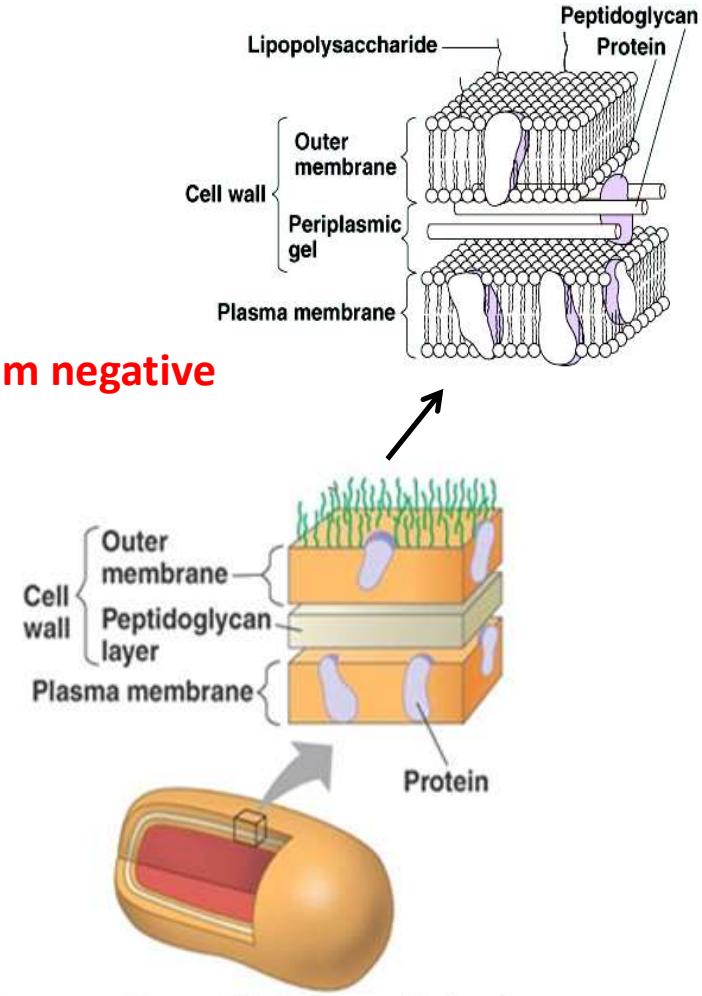
Staining Principle

Gram positive



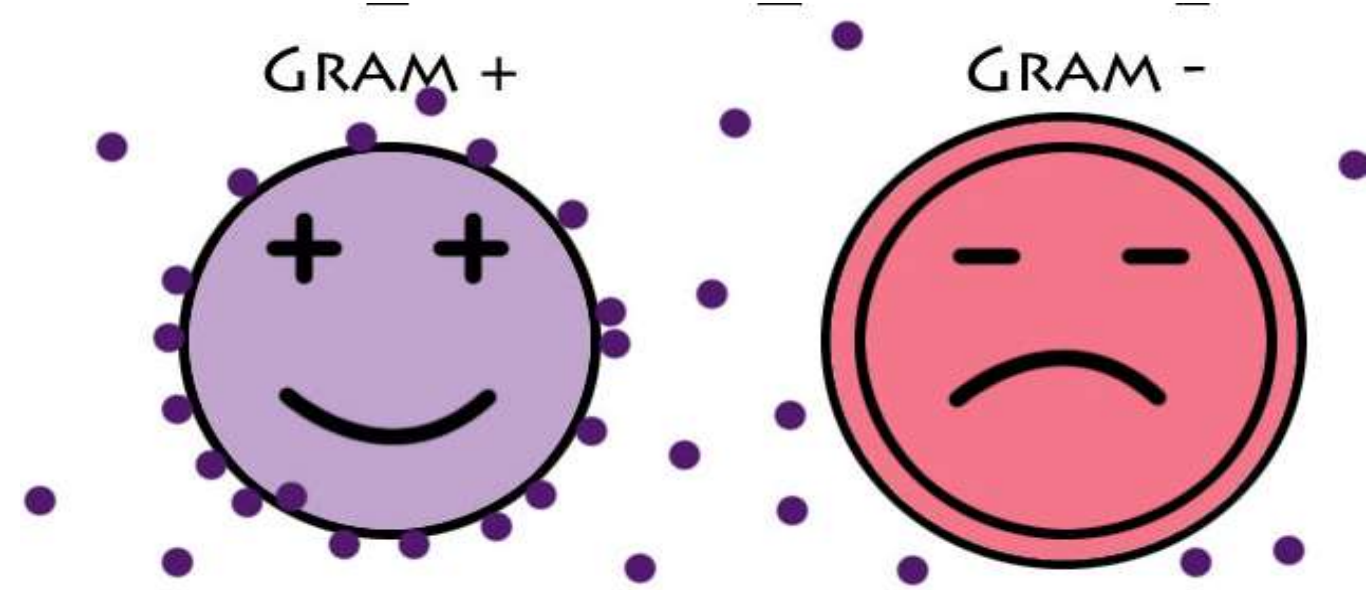
(a) Gram-positive: peptidoglycan traps crystal violet.

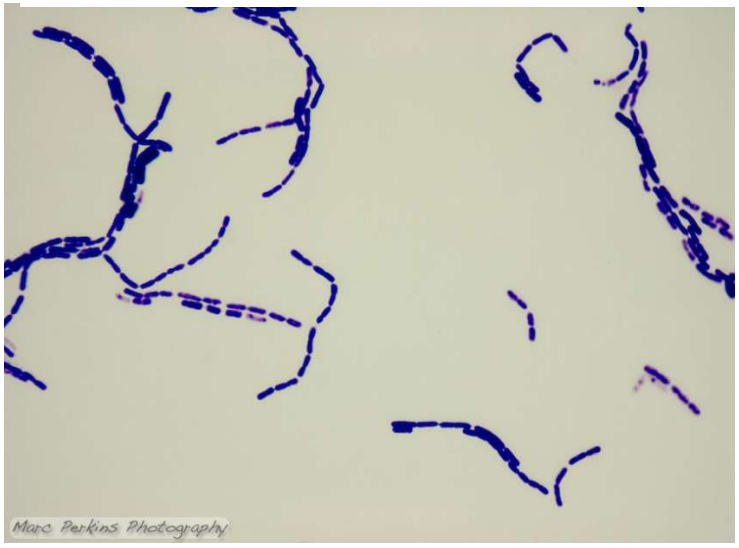
Gram negative

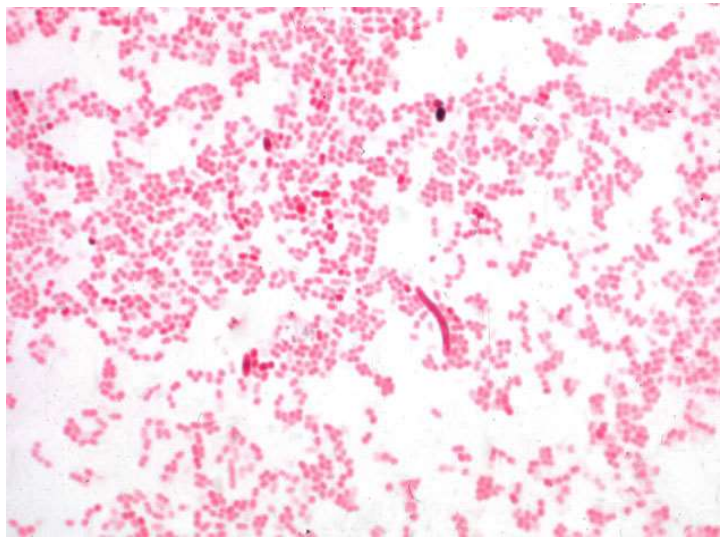


(b) Gram-negative: crystal violet is easily rinsed away, revealing red dye.

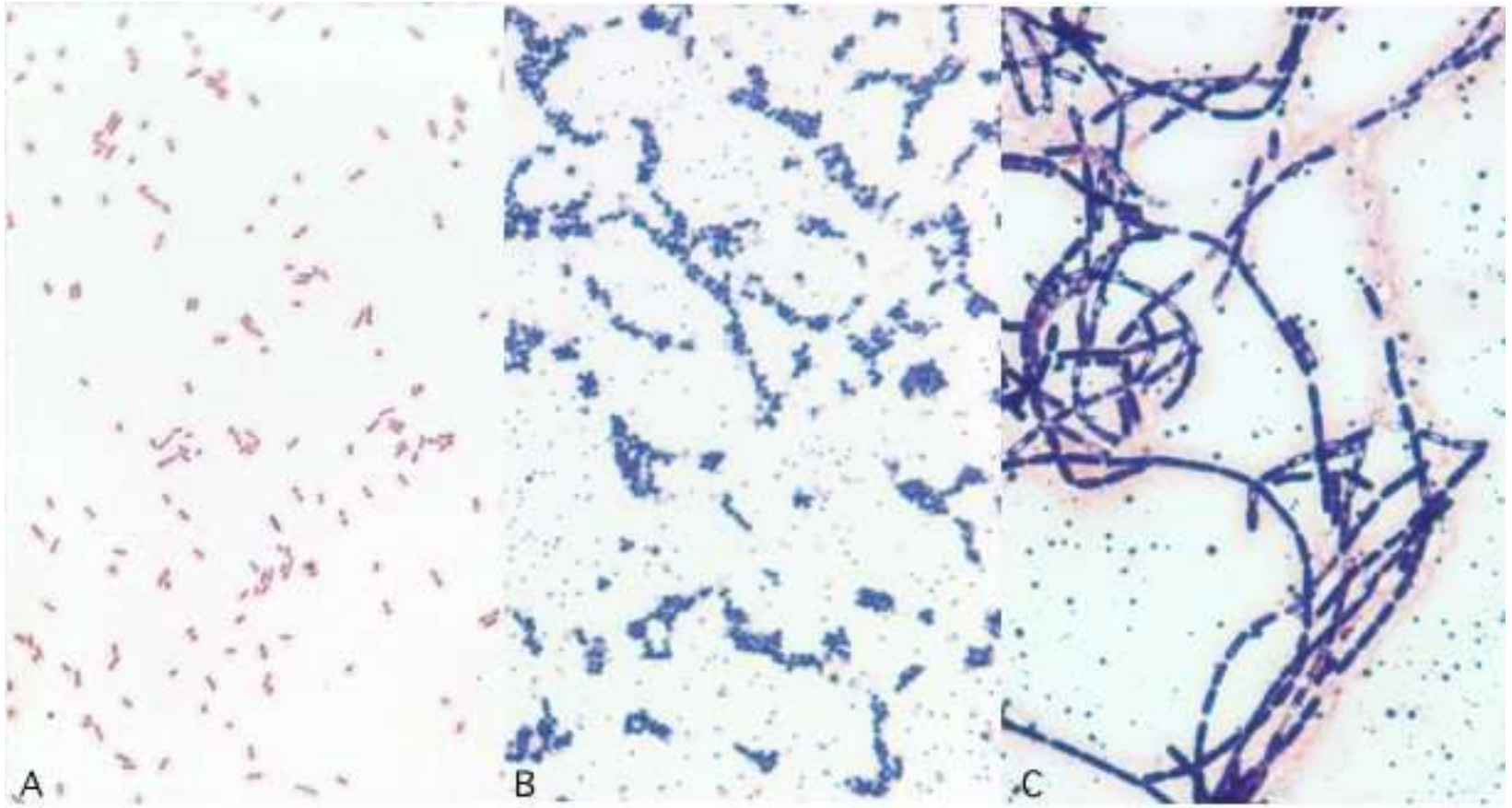
Results of Gram staining







Results of Gram staining

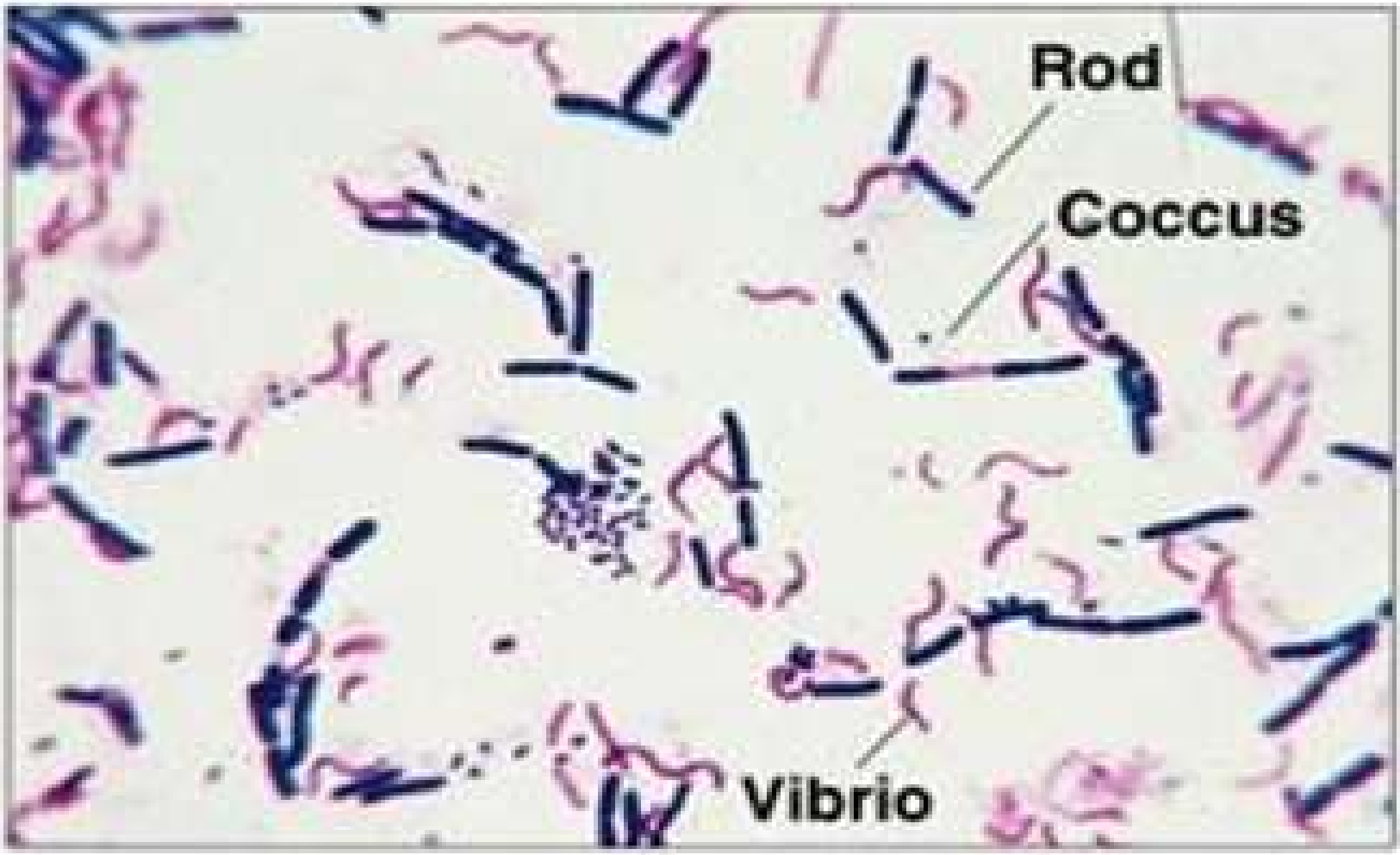


Proteus

Staphylococcus aureus (S.aureus)

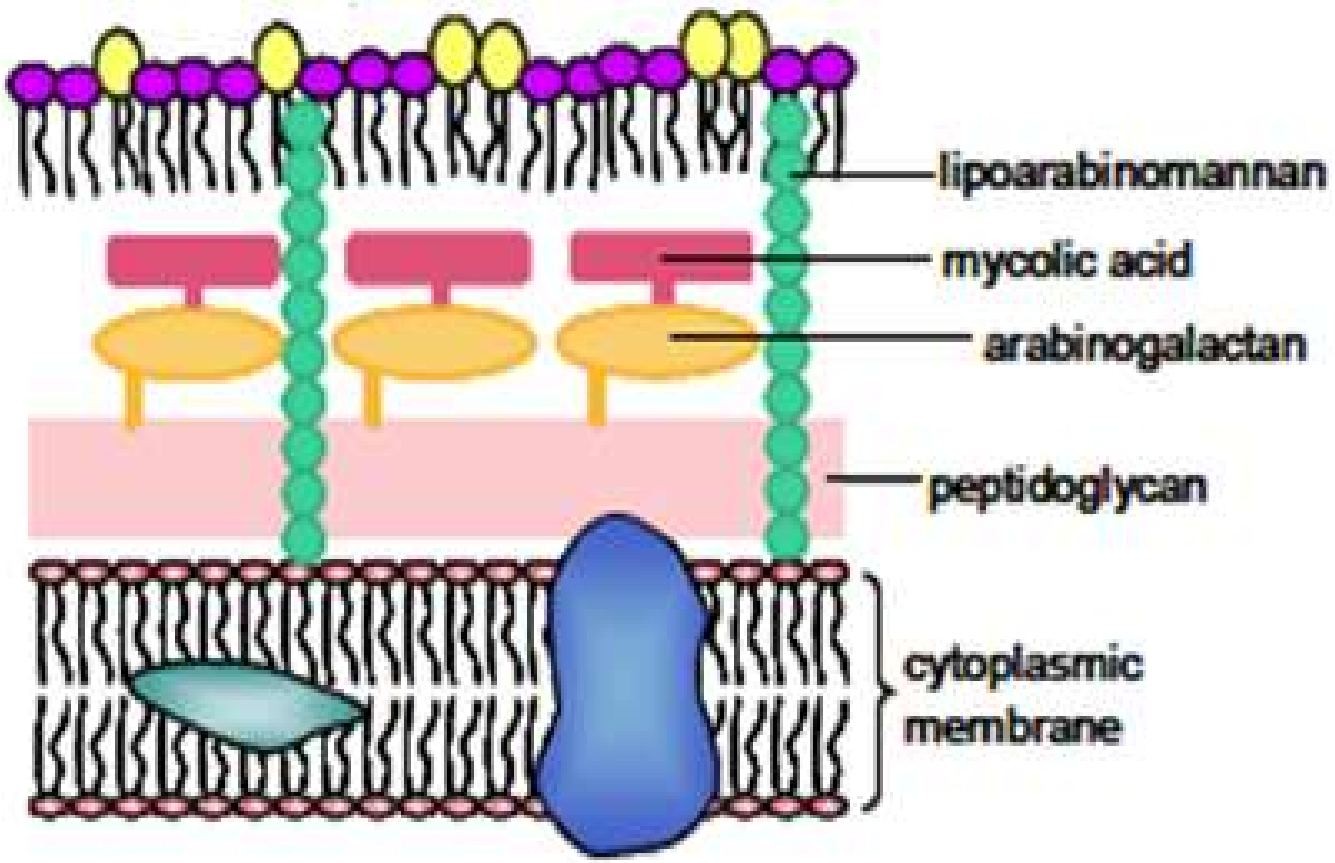
Bacillus cereus (B.cereus)

Results of Gram staining

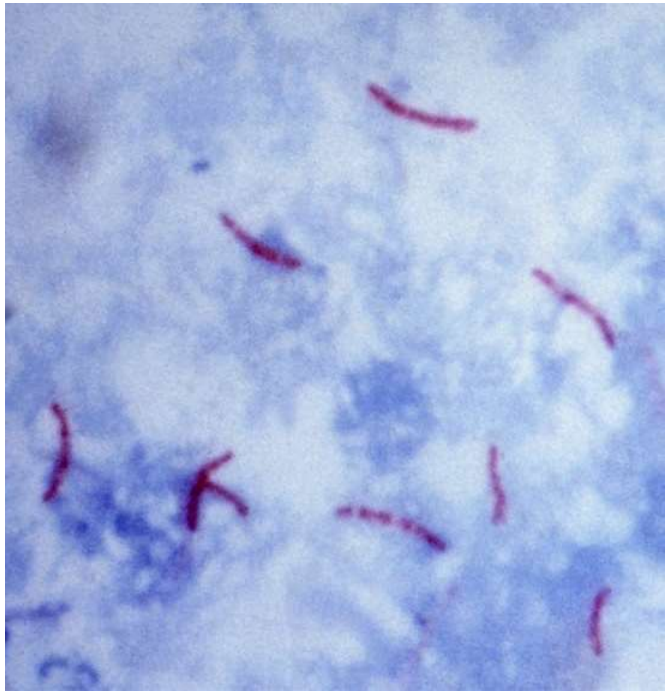


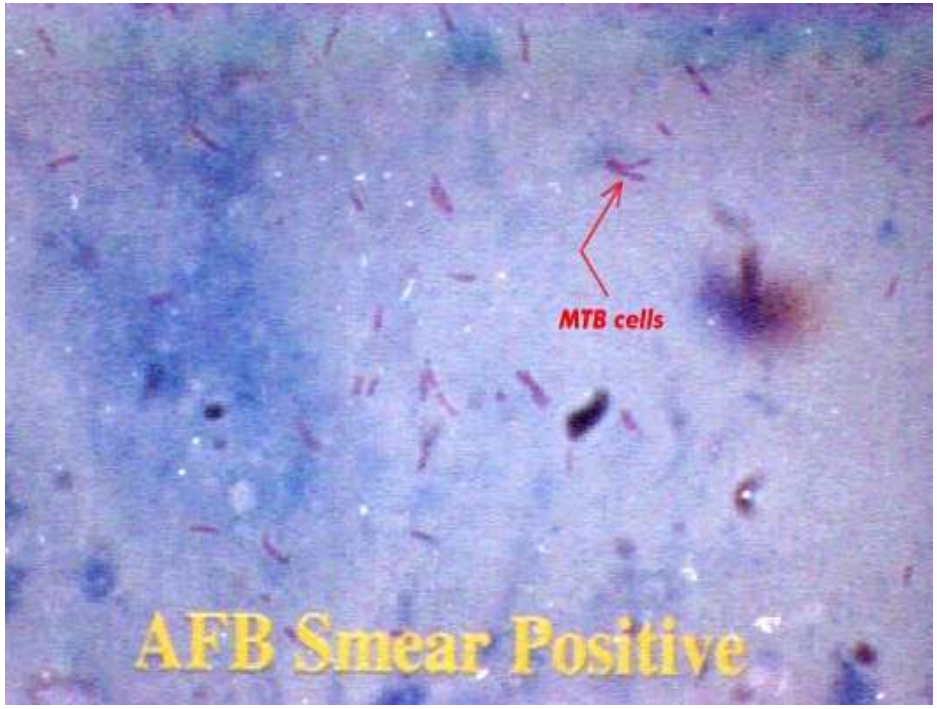
Acid fast staining

Principle



Results of acid fast staining





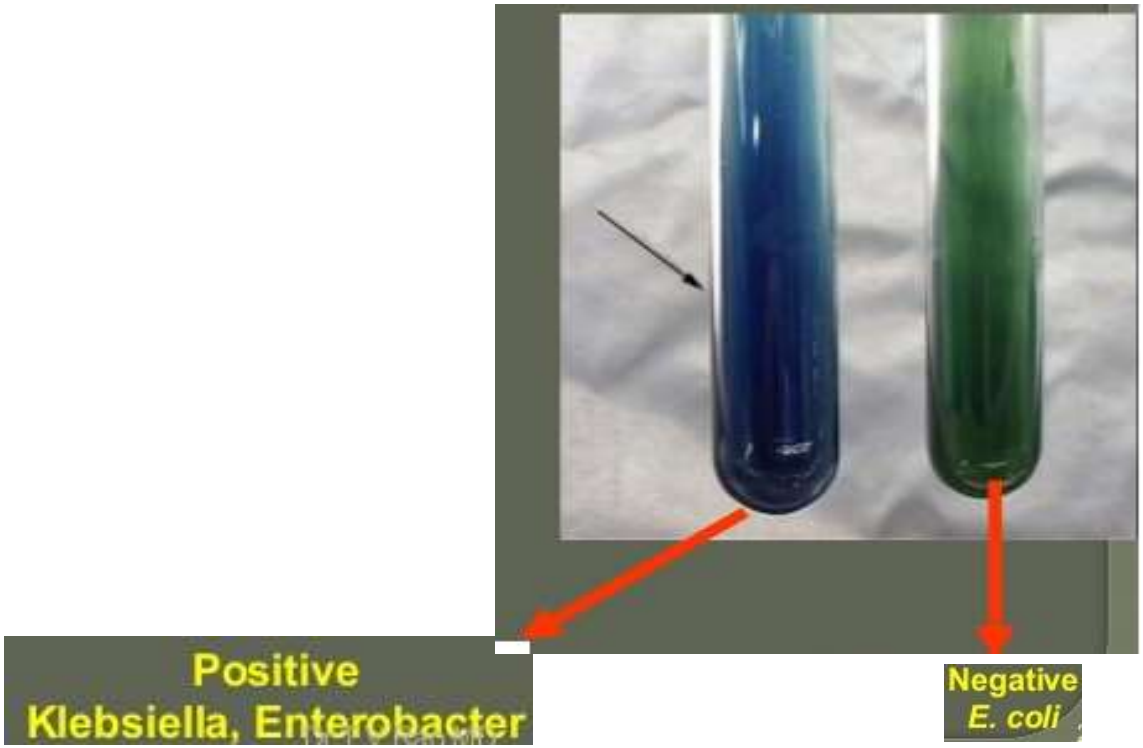
Mycobacterium tuberculosis(TB bacteria)

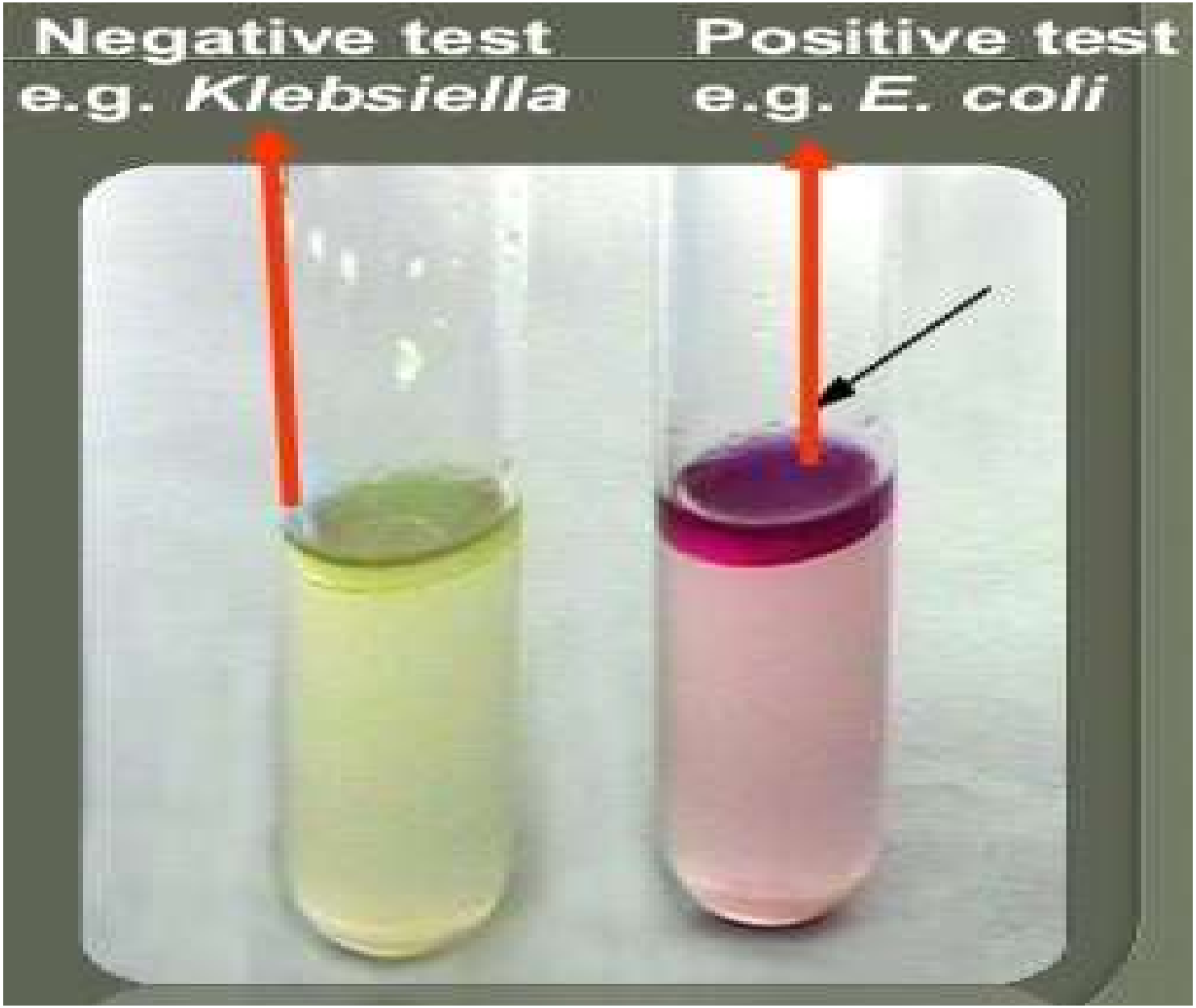
IMViC: Citrate utilization test

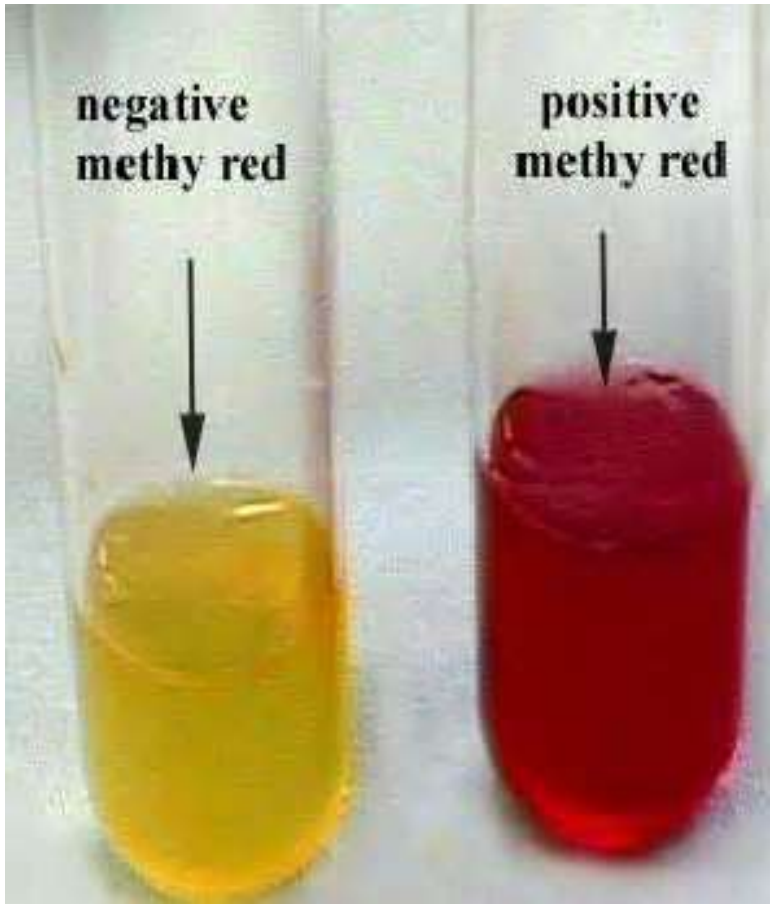
Results

Positive results: blue color (*Klebsiella*)

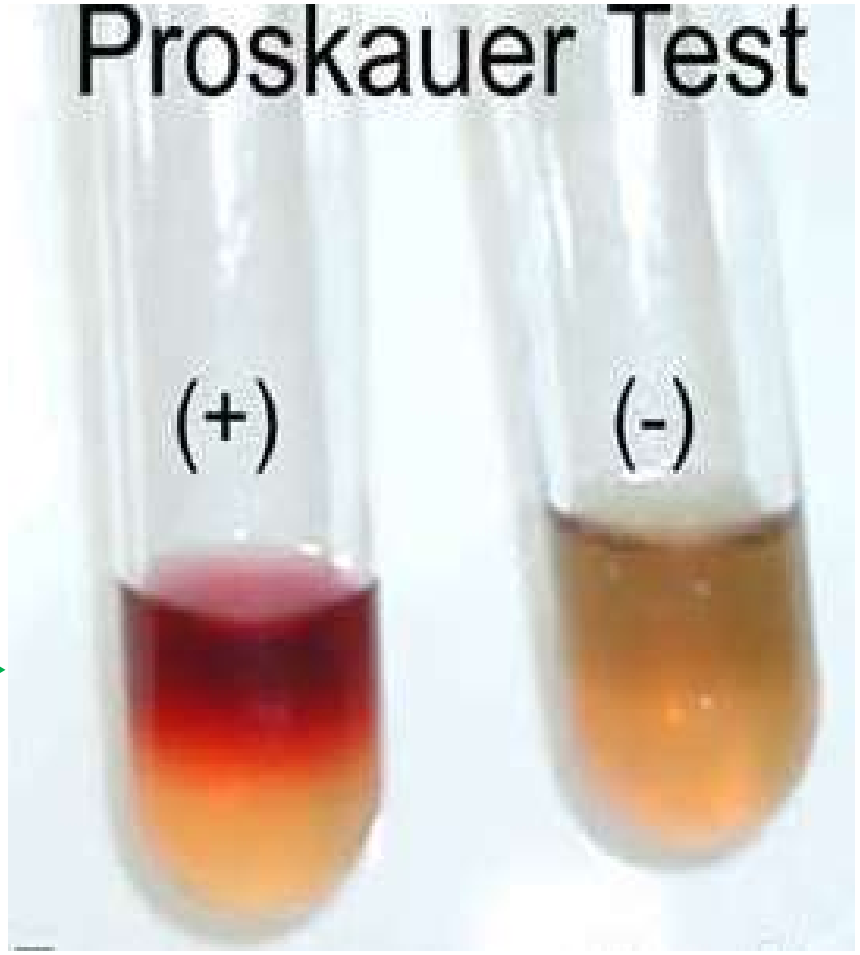
Negative results: green color (*E. coli*)




Negative test
e.g. Klebsiella
Positive test
e.g. E. coli



- ✓Red: Positive MR (*E. coli*)
- ✓Yellow or orange: Negative MR (Klebsiella)



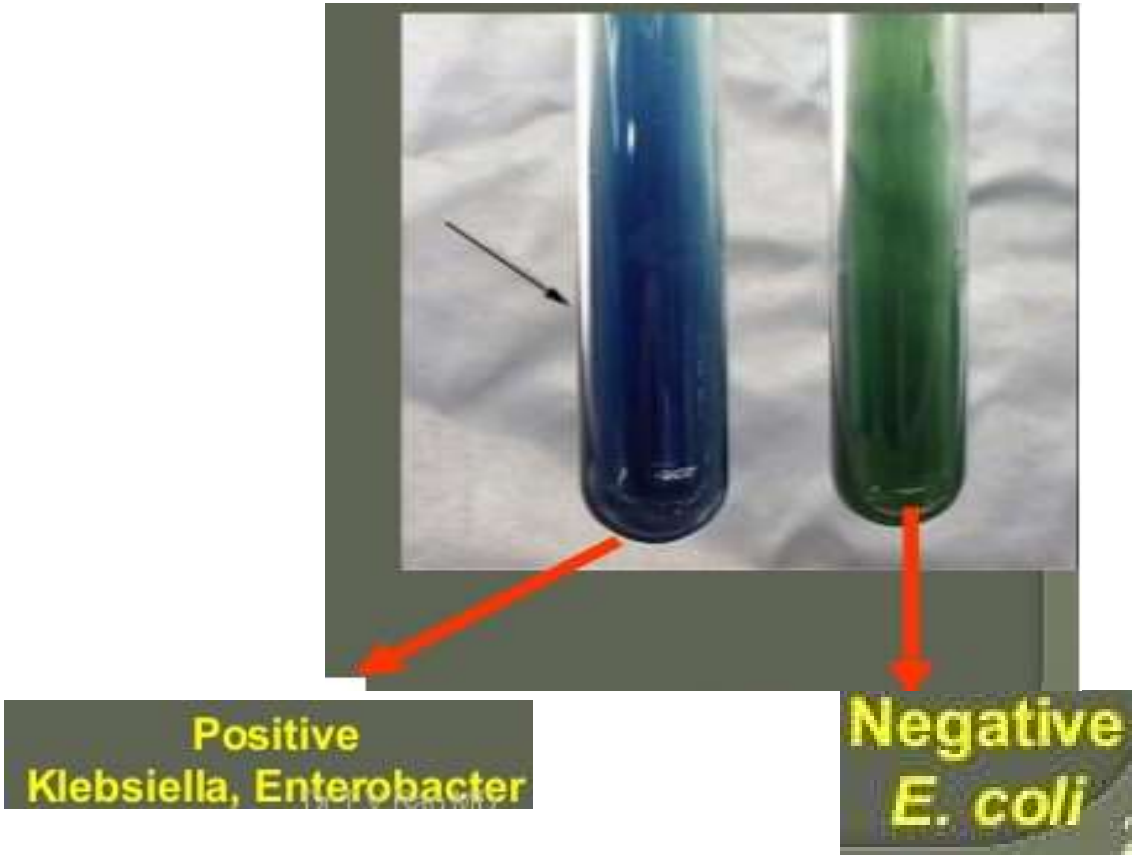
- ✓Pink: Positive VP (*Klebsiella*)
- ✓No pink: Negative VP (*E. coli*)

IMViC: Citrate utilization test

Results

Positive results: blue color (*Klebsiella*)

Negative results: green color (*E. coli*)

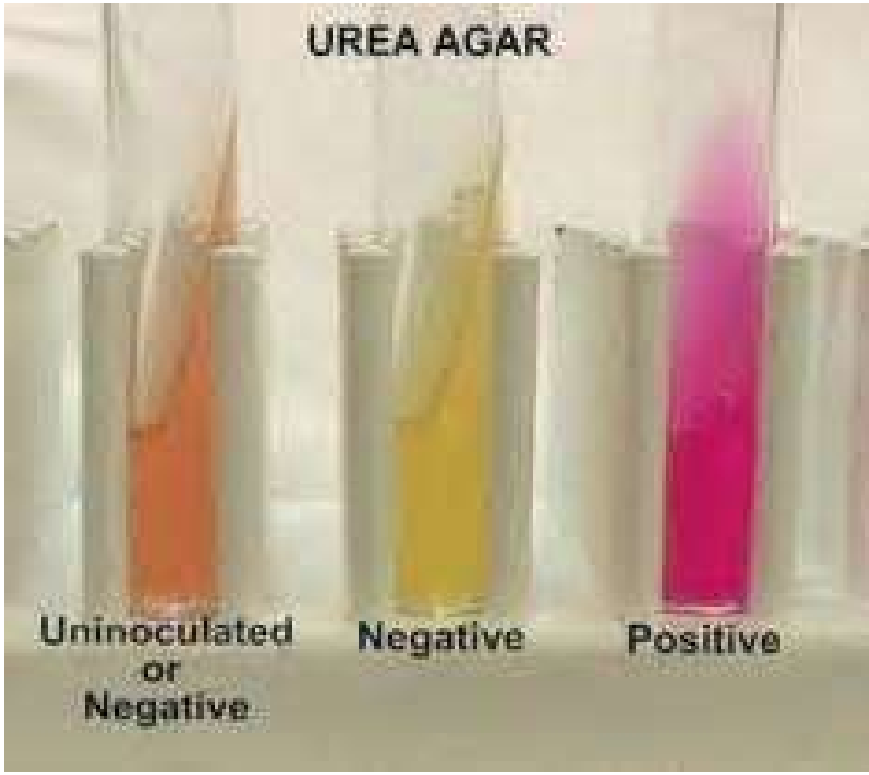


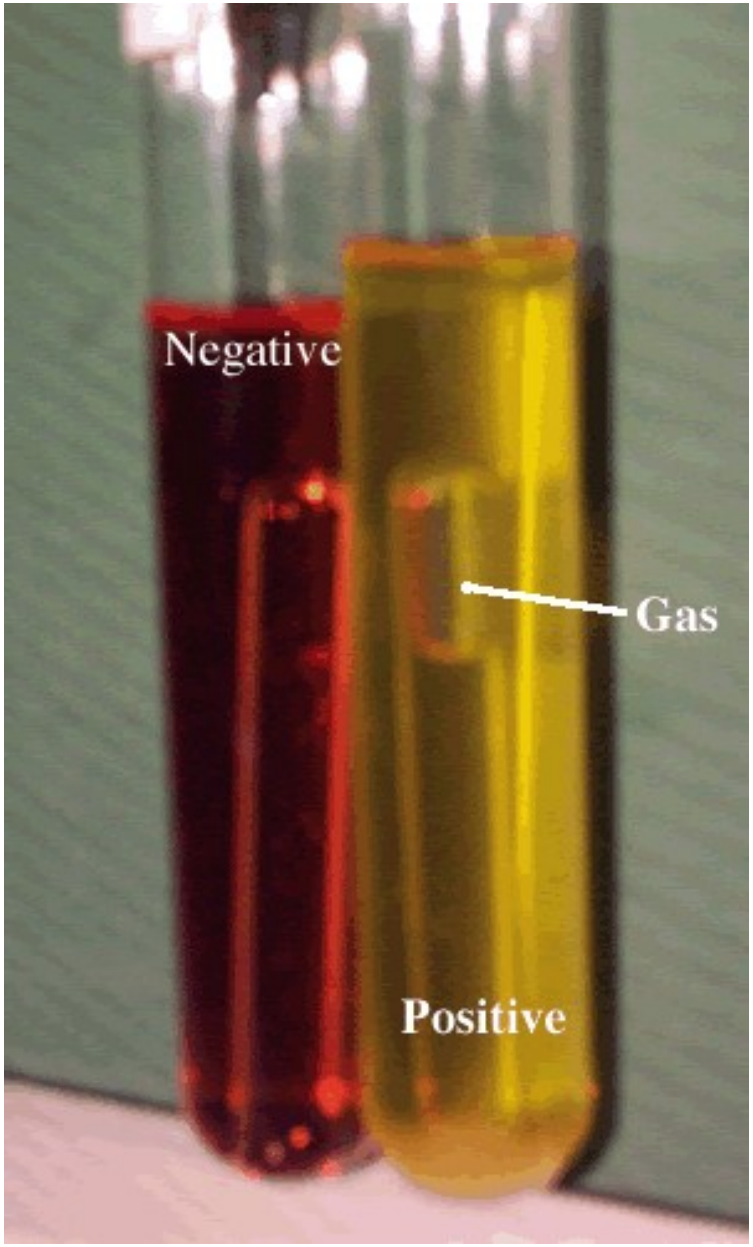
Urease test

Results

Negative = yellow color e.g., *E. coli*

Positive = **pink color e.g., klebsiella aregenes**





Sugar fermentation test

Analytical Profile Index System (API) for bacterial identification

•**API (Analytical Profile Index) 20E** is a biochemical panel for identification and differentiation of members of the family Enterobacteriaceae.

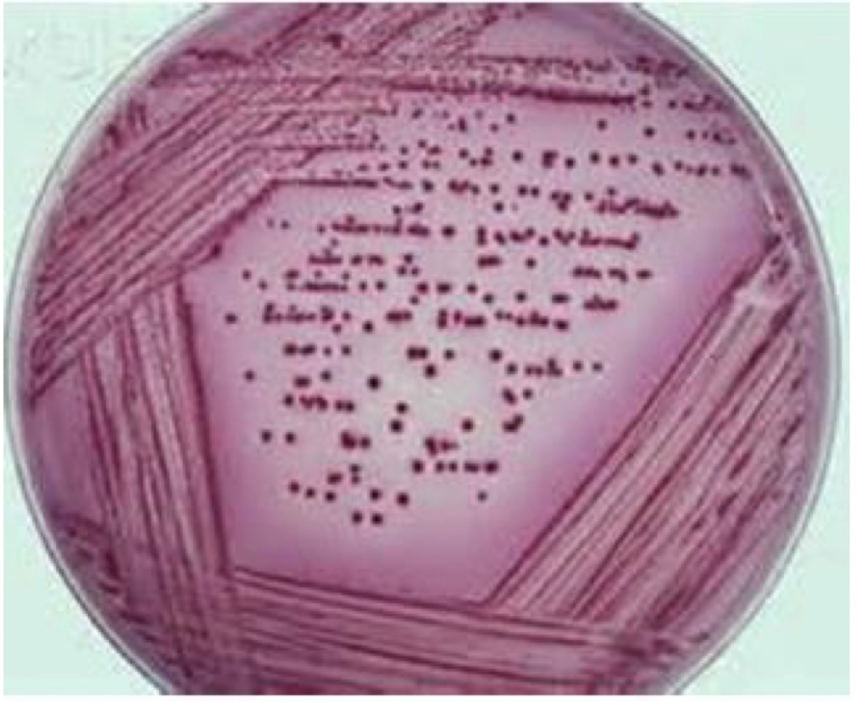
Proteus vulgaris

Proteus mirabilis

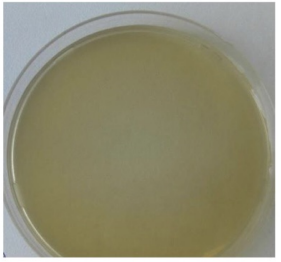
Escherichia coli

Providencia alcalifaciens

Gram negative bacilli
Lactose fermenter



Principle Standardized filter-paper disc-agar diffusion



Mueller Hinton agar

→ Confluent growth → Applying antibiotic disks

↓

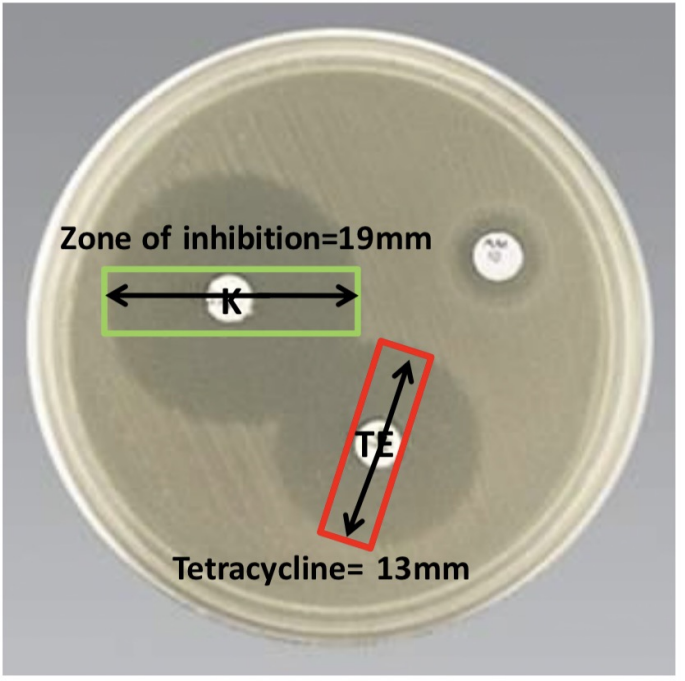
Incubation 24h at 37°C

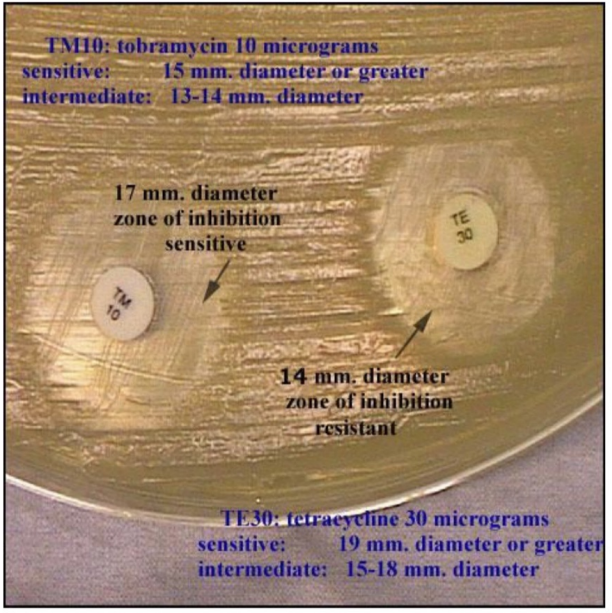
←

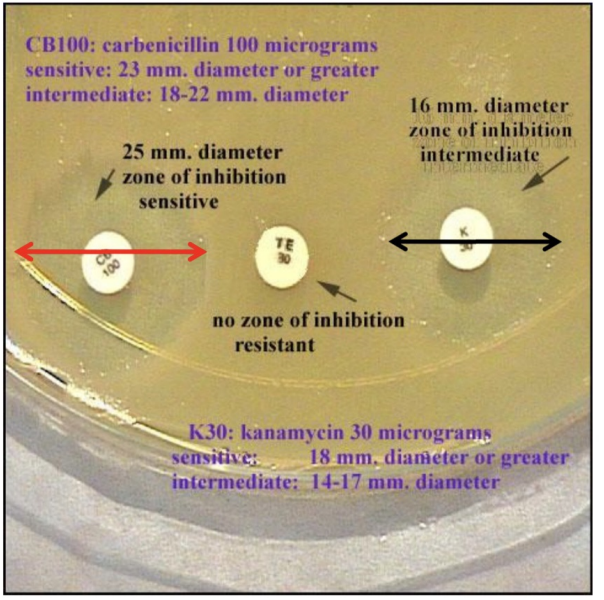
Read the diameter of the inhibition zone

Standardized filter-paper disc-agar diffusion

Tetracycline




TM10: tobramycin 10 micrograms
sensitive: 15 mm. diameter or greater
intermediate: 13-14 mm. diameter
17 mm. diameter
zone of inhibition
sensitive
TM 10
TE 30
14 mm. diameter
zone of inhibition
resistant
TE30: tetracycline 30 micrograms
sensitive: 19 mm. diameter or greater
intermediate: 15-18 mm. diameter


CB100: carbenicillin 100 micrograms
sensitive: 23 mm. diameter or greater
intermediate: 18-22 mm. diameter
16 mm. diameter
zone of inhibition
intermediate
25 mm. diameter
zone of inhibition
sensitive
CB 100
TE 30
K 30
no zone of inhibition
resistant
K30: kanamycin 30 micrograms
sensitive: 18 mm. diameter or greater
intermediate: 14-17 mm. diameter

McFarland standard

Different McFarland standards

0.5

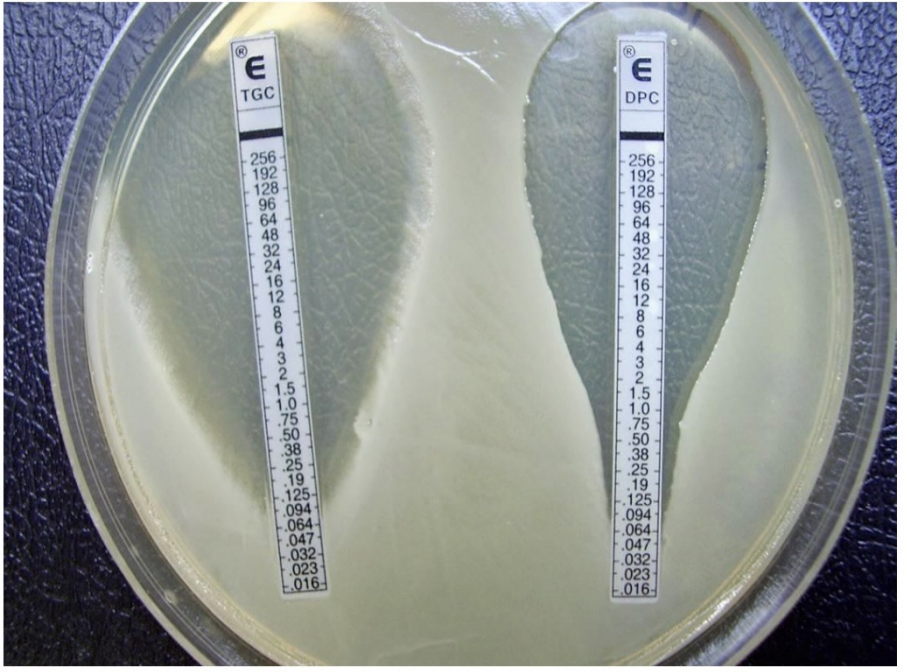
Absorbance at 600 nm
(0. 08 to 0.1)

The broth used to Inoculate the Hinton Muller agar

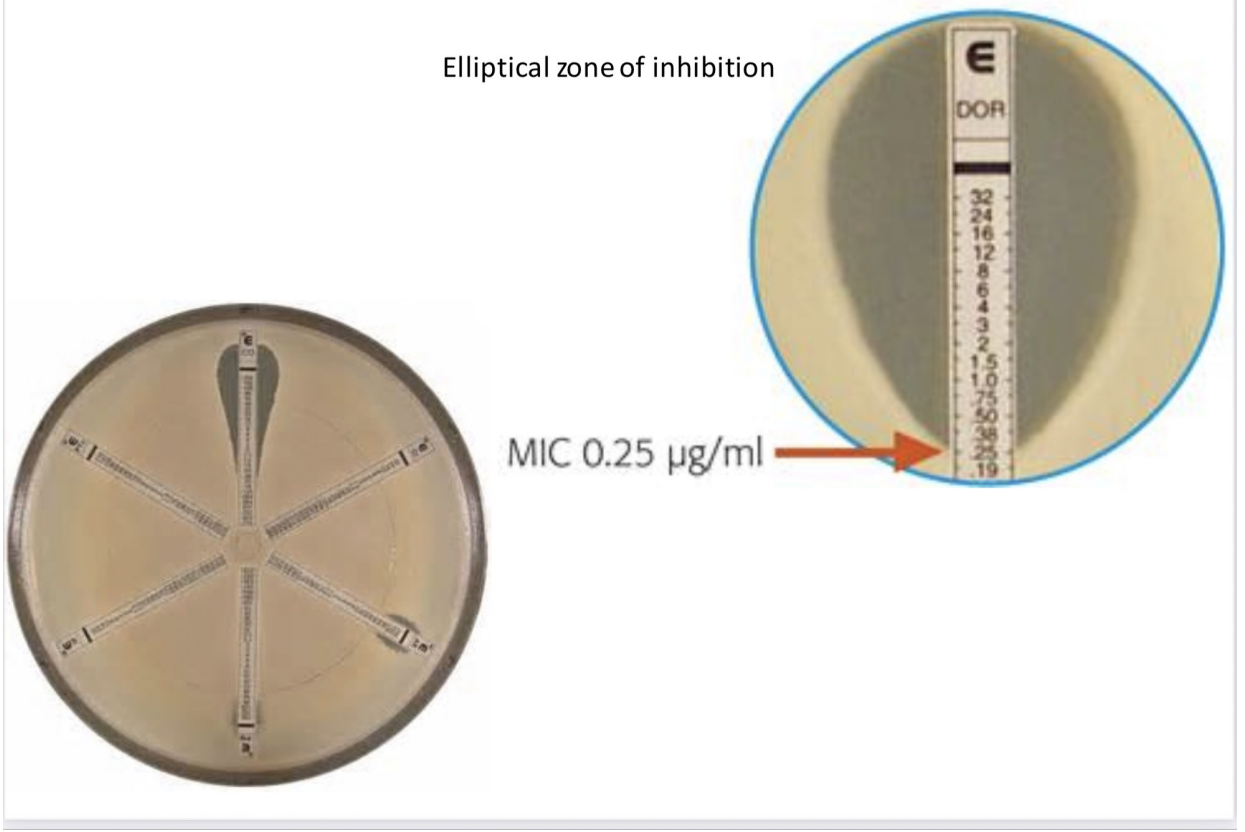
When equal turbidity=
150,000,000 CFU/ml

Epsilometer test (E-test)

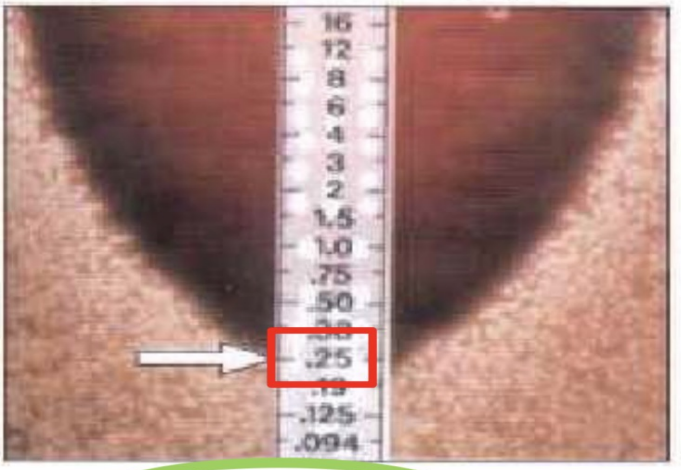
Elliptical zone



Epsilometer test (E-test)



Reading E-tests



Ciprofloxacin