Lecture 4 Part-1

Bacterial taxonomy, Classification, and laboratory diagnosis

Objectives

Def. of Taxonomy

Nomenclature

Scheme of medical bacteria

Biochemical reactions

Bacterial Taxonomy

Taxon= group , Taxa= groups= classification

The science of biological classification

Bacterial Taxonomy

Classification

Nomenclature

Identification

Bacterial Taxonomy Rank

- Kingdom or Domain
- Division or Phylum
- Class
- Order
- Family
- Genus
- Species
- Strains

Bacterial Taxonomy

Staph. aureus

(Species)

Strain

Individual member within a species

MRSA

(Strain)

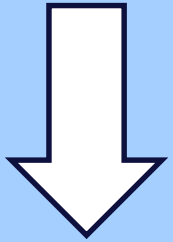
Bacterial Taxonomy

Species

A collection of strains that share many stable properties

Staph. aureus

(Species)



MRSA

(Strain)

VRSA

(Strain)

Bacterial Taxonomy

Species

***S. aureus* (Species)**

Same species

(DNA homology ≧ 70%)

(16S rRNA ＞97% identical)

Bacterial Taxonomy

Genus

Staphylococci

Species

S. epidermidis

Species

S. aureus

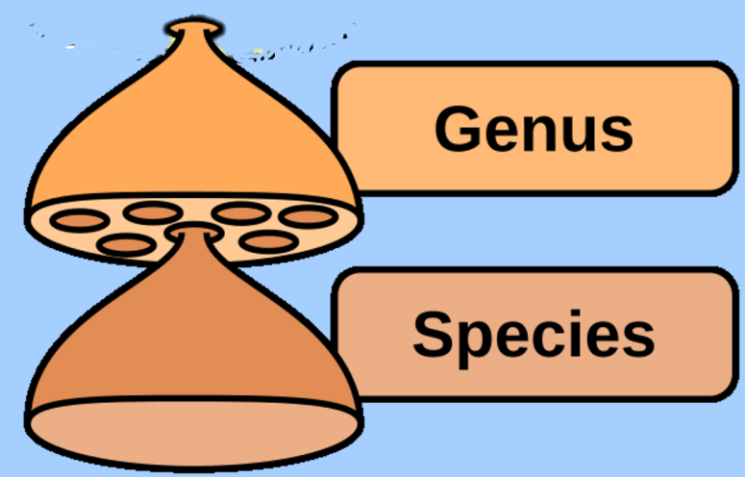
One or more species

that share common properties

DNA < 93% new genus

Nomenclature

Genus + species



Nomenclature

Escherichia coli

Italic

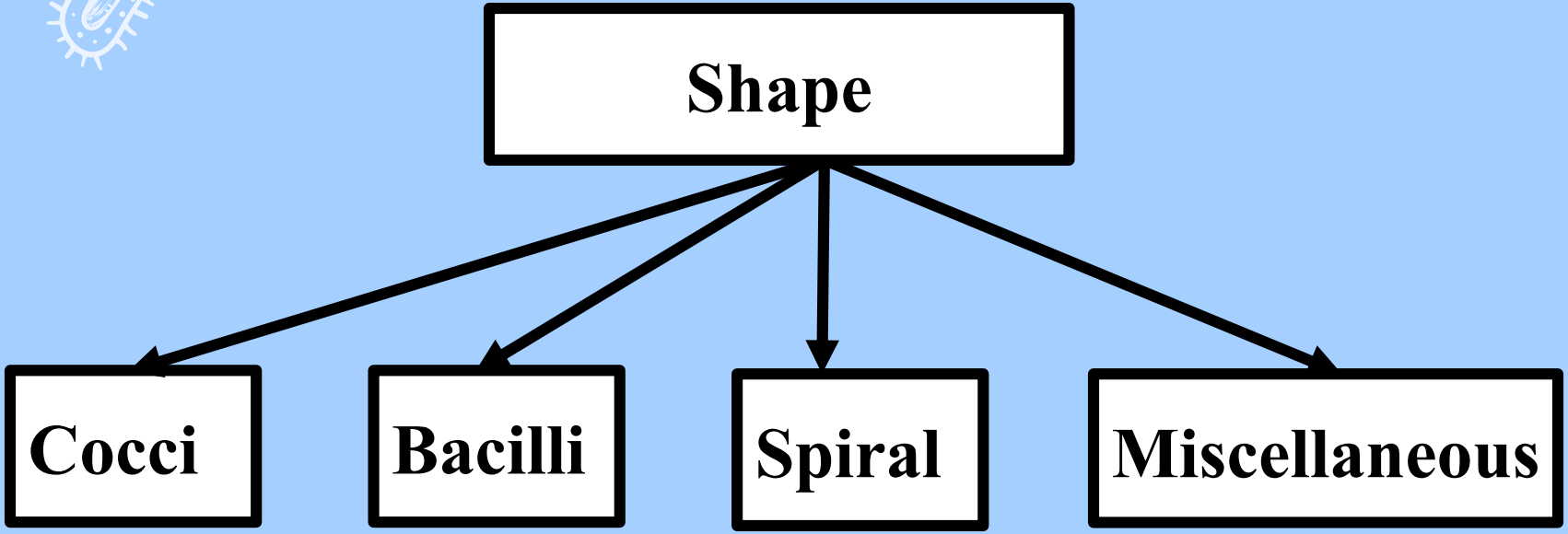
I

Nomenclature

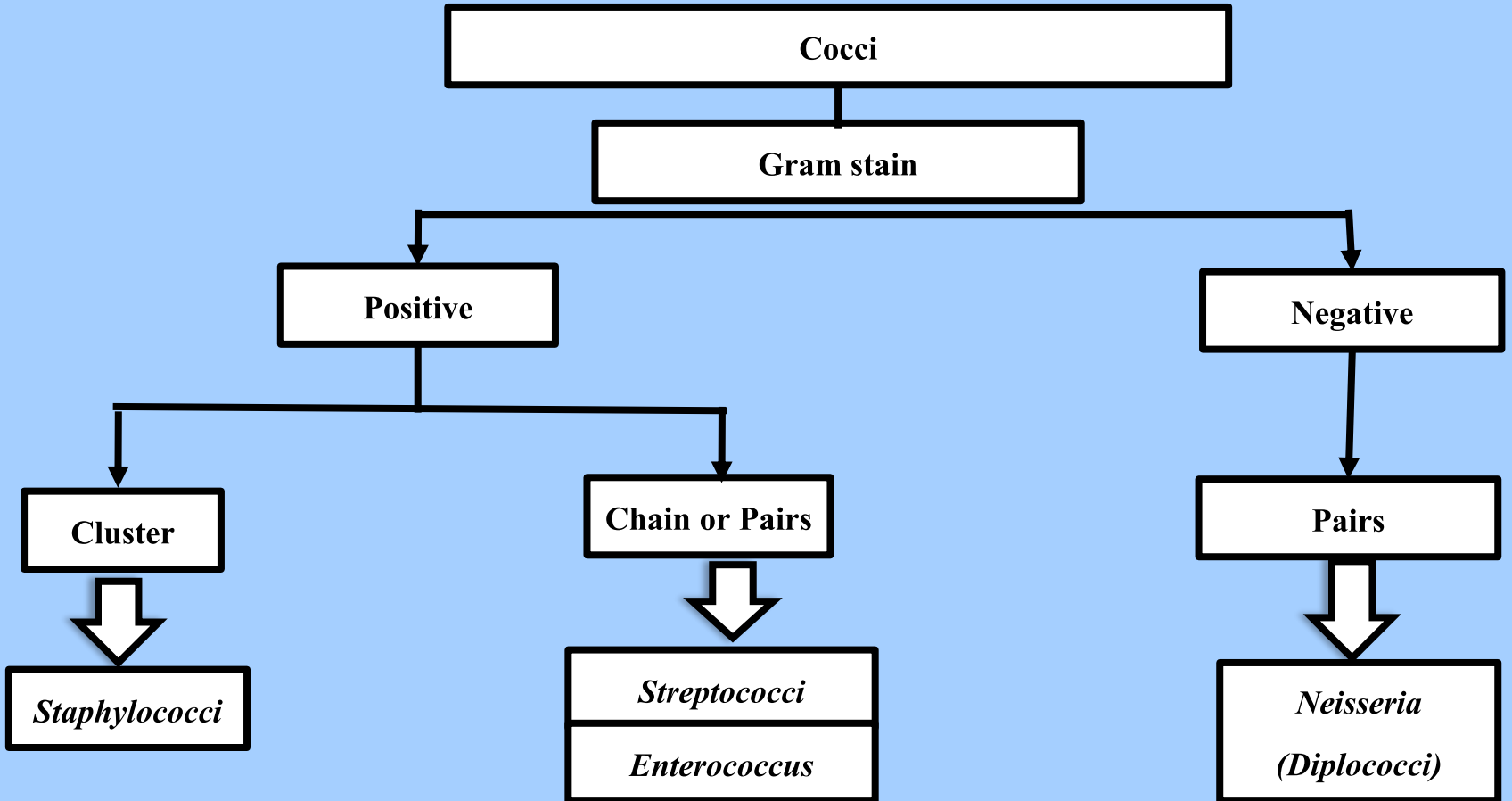
Escherichia coli

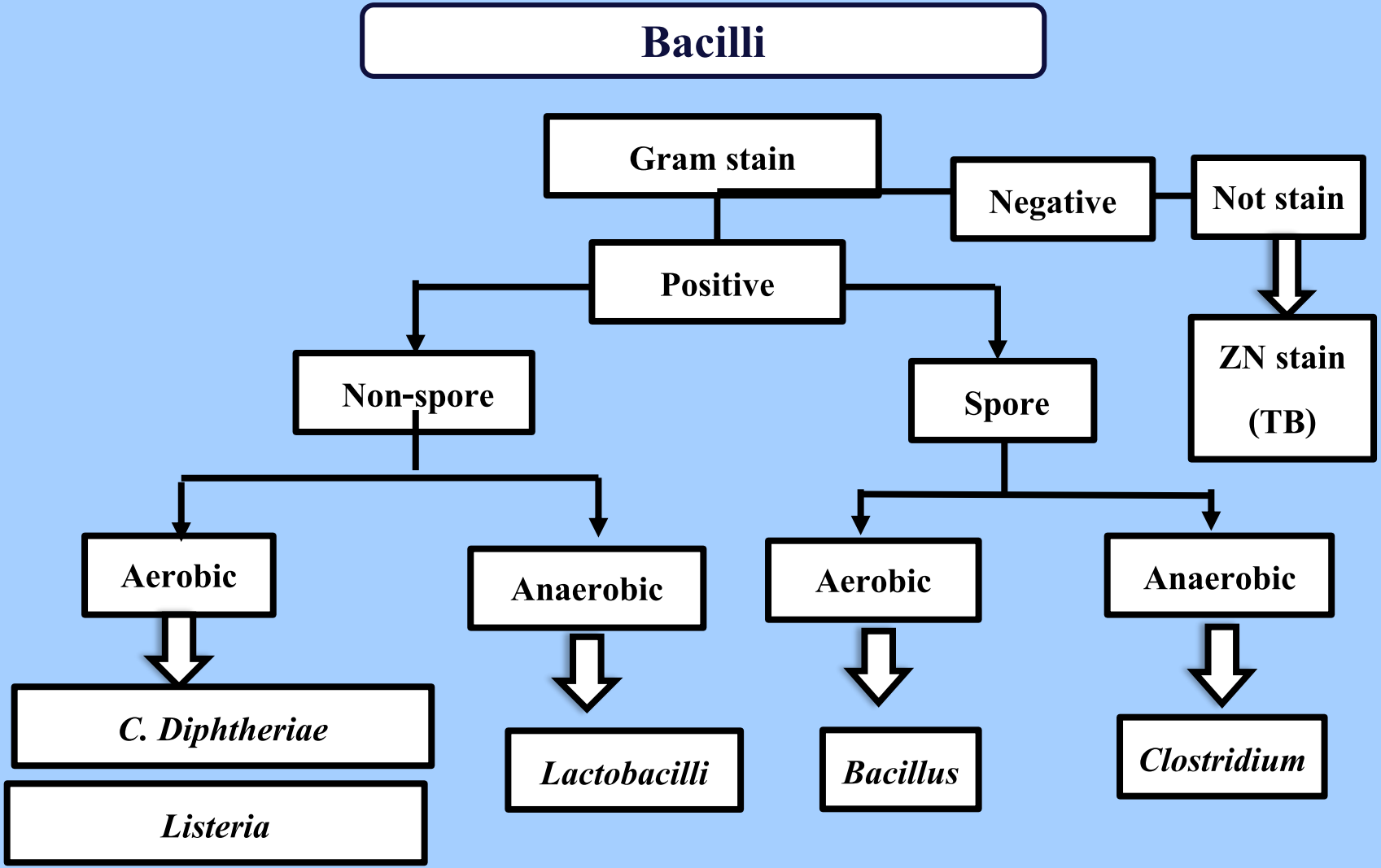
(E. coli)

Scheme of medical bacteria



Scheme of medical bacteria




Bacilli
Gram stain
Negative
Not stain
Positive
ZN stain
(TB)
Non-spore
Spore
Aerobic
Anaerobic
Aerobic
Anaerobic
C. Diphtheriae
Listeria
Lactobacilli
Bacillus
Clostridium

Gram negative bacilli

- *Enterobacteriaceae*
- *Vibrio*
- *Campylobacter*
- *Helicobacter*
- *Pseudomonas*
- *Haemophilus*
- *Bordetella*
- *Brucella*
- *Legionella*
- **Gram –ve anaerobes**

Spiral

- ***Treponema***
- ***Borrelia***
- ***Leptospira***

Miscellaneous group

- No cell wall
- Not stain by gram
- Obligate intracellular

- Mycoplasma
- Chlamydia
- Rickettsia
- Coxiella
- Actinomycetes

Systematic Bacteriology

Morphology & Culture

Virulence factor & Pathogenesis

Diseases

Lab. diagnosis

Treatment & Prevention

Biochemical reactions

1) Indole test

Bacteria

Peptone

(TRYPTOPHAN)

break down (Tryptophanase)

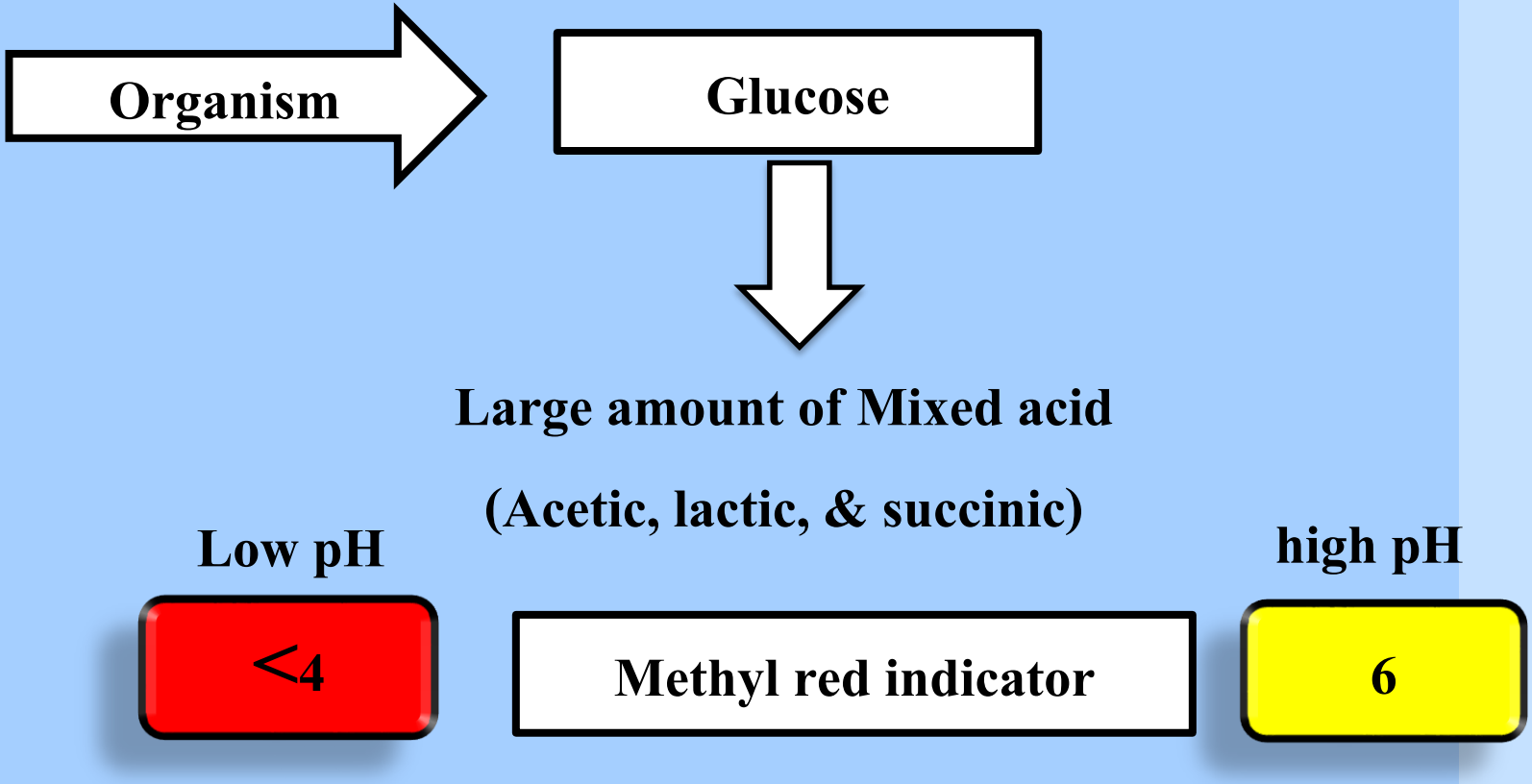
Indole

+

Kovac's R.

Red

2) Methyl red test



2) Methyl red test

Bacteria

Peptone

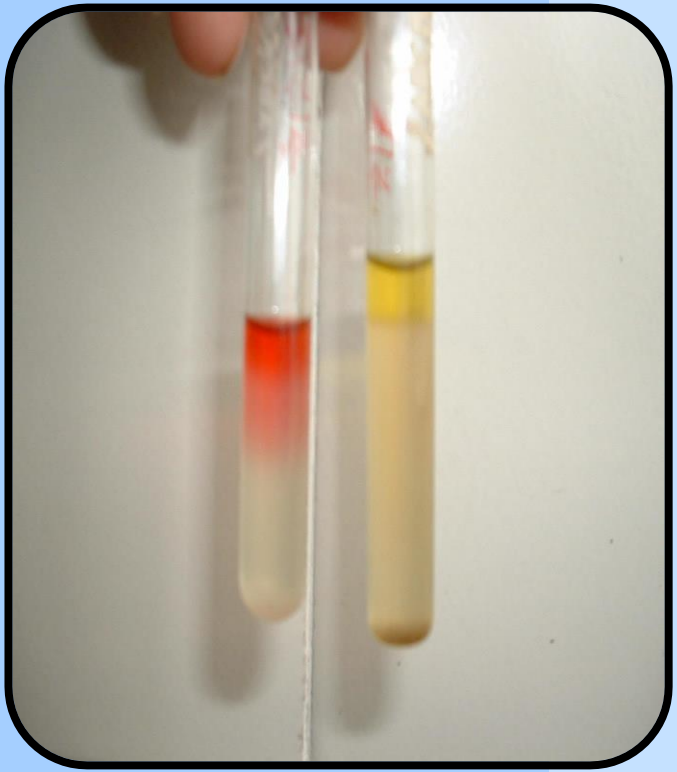
(Glucose)

Incubate at 37°C for 48h

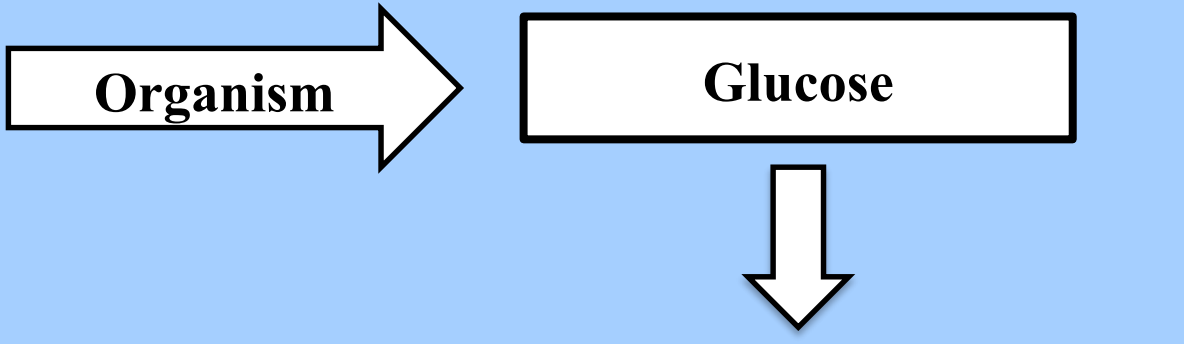
(Large Acid) Mixed

pH

MR indicator



3) Voges-Proskauer test (V.P)



3) Voges-Proskauer test (V.P)

Bacteria

Peptone

(Glucose)

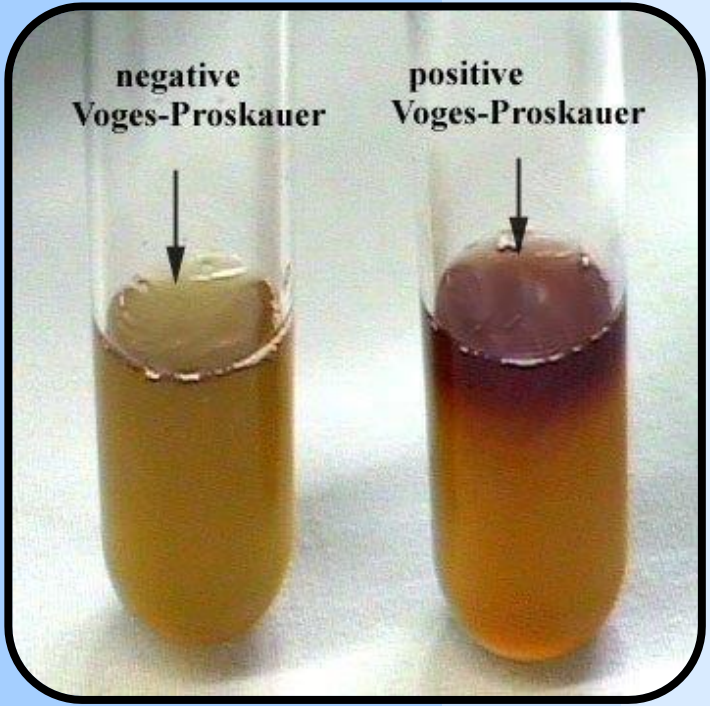
Incubate at 37°C for 48h

Ferment glucose (Acetyl methyl carbinol)

∝- naphthol + 40% KOH

Diacetyl

Red

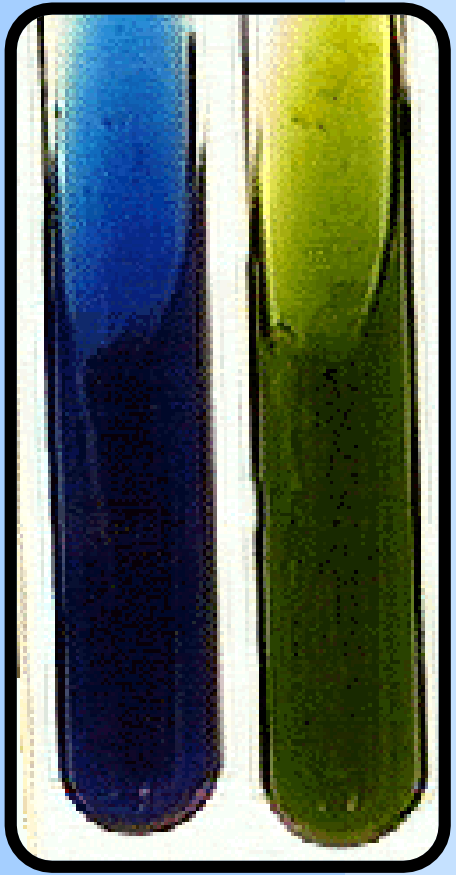


MR &VP

If methyl red is positive, the voges- proskauer should be negative and reverse is right.

4) Citrate utilization test

Utilized citrate as only source of carbon



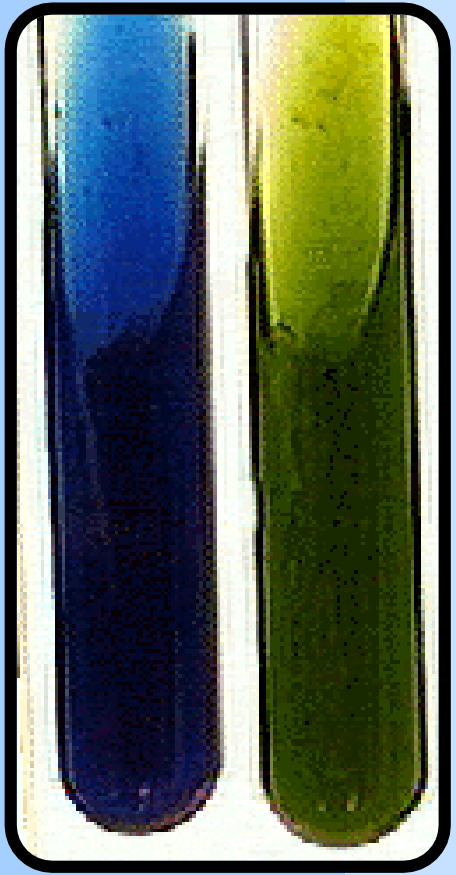
4) Citrate utilization test

The indicator is bromothymol blue.

High pH

N

7



4) Citrate utilization test

Bacteria

Citrate medium

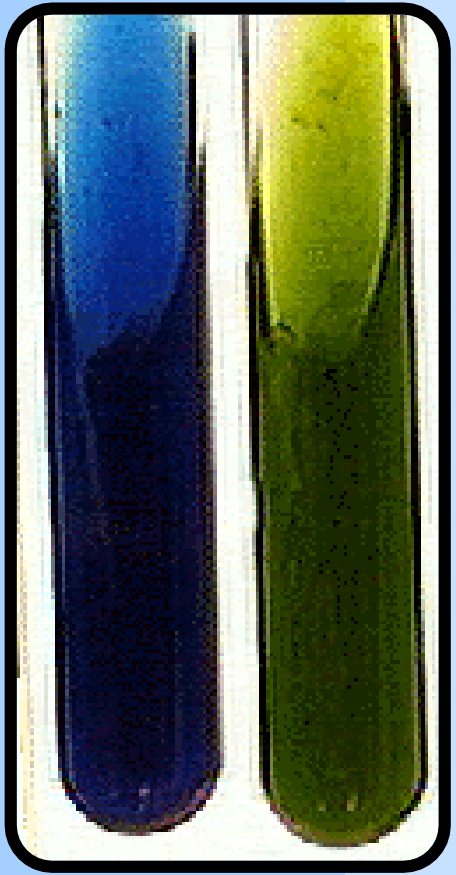
Incubate at 37°C for 48h

Liberated Co_2

Co_2+with sodium

sodium carbonate(Alkaline)

Indicator change to blue



5) Urease test

Phenol red indecator

Acid	Alkaline
6	8

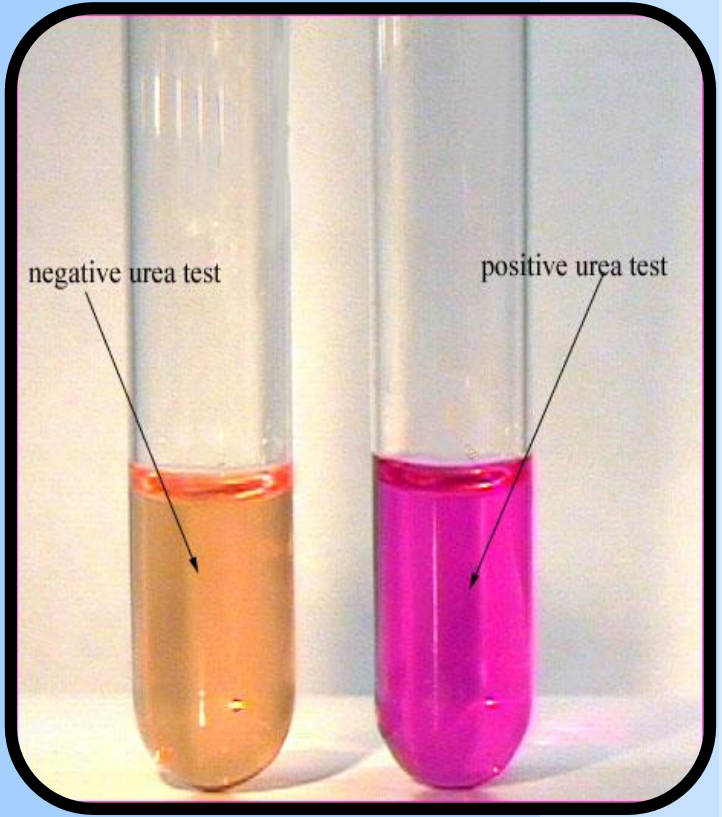
5) Urease test

Bacteria(Urease) → **Urea**

↓

Ammonia

(Pink)



6) TSI

0.1% glucose

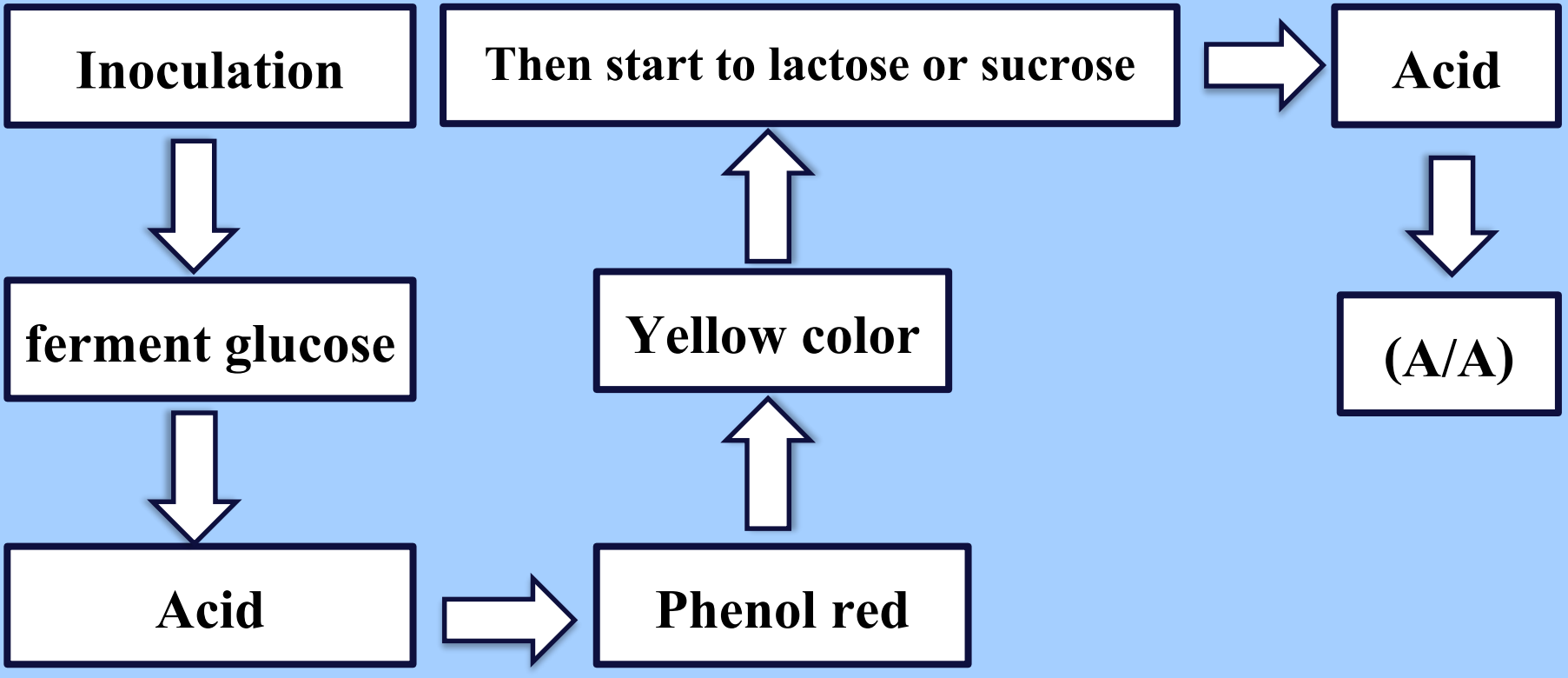
1% lactose

1% sucrose

Ferrous sulfate

pH indicator: Phenol red

6) TSI a) Acid over acid (A/A)



6) TSI (a) Acid over acid (A/A)

a) Acid over acid (A/A)

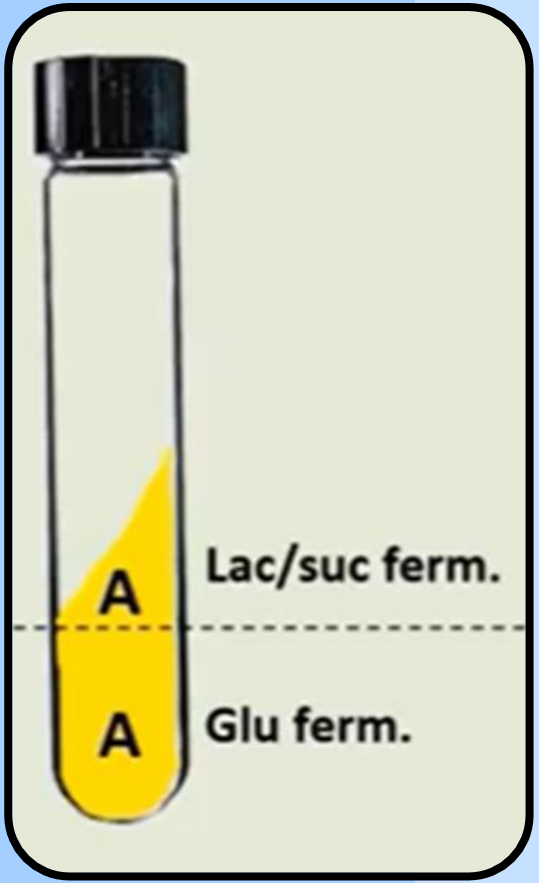
Detection of gas production by break up the medium or pushed up the tube.

a) Acid over acid (A/A)

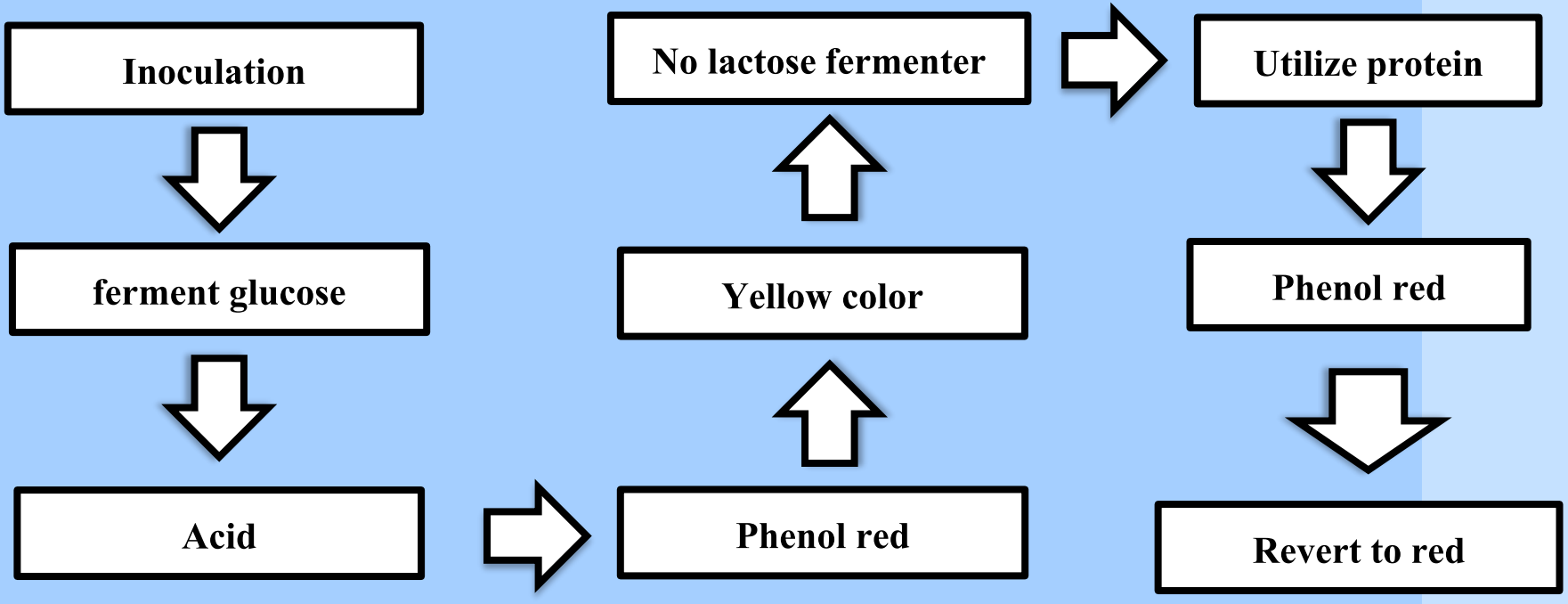
A/A

Glucose fermenter

Lactose fermenter



b) Alkaline over acid (K/A)



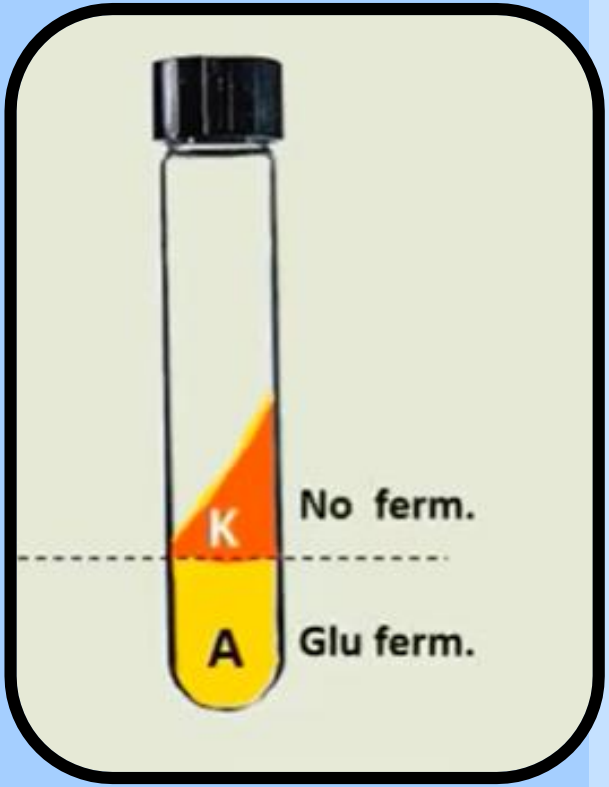
a) Alkaline over acid (K/A)

a) Alkaline over Alkaline (K/K)

K/A

Glucose fermenter

Non-lactose fermenter



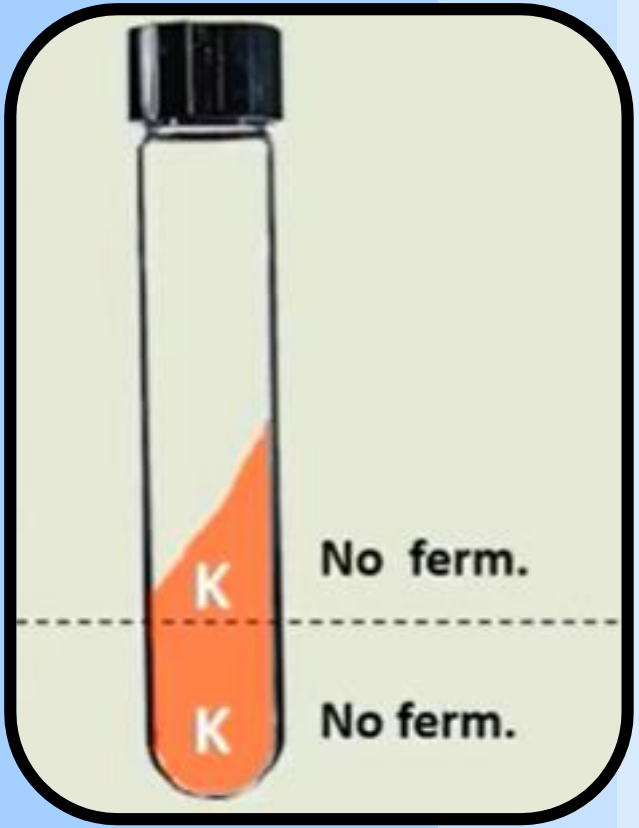
a) Alkaline over Alkaline (K/K)

If the organism can not use the glucose in the medium. The color of the medium remains red.

a) Alkaline over Alkaline (K/K)

K/K

No sugar fermenter



H2S production

Bacteria (Reduce) → Sulfur

↓

Hydrogen sulfide

(H_2S) + iron

↓

Ferric sulfide

(Black)

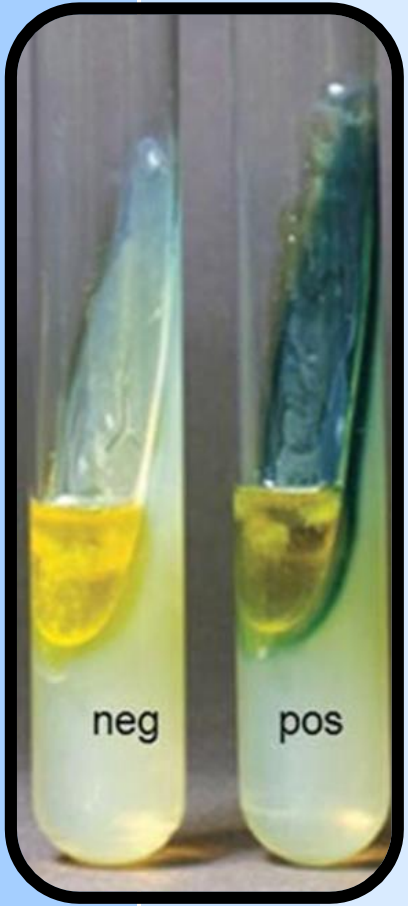
7) Phenylalanine deaminase

Phenylalanine deaminase

Phenylalanine → Phenyl pyruvic acid + NH_3

Add Ferric chloride

Green color



Distinguishes *Proteus* from *Salmonella & Shigella*

8) Ornithine decarboxylase

Ornithine decarboxylase

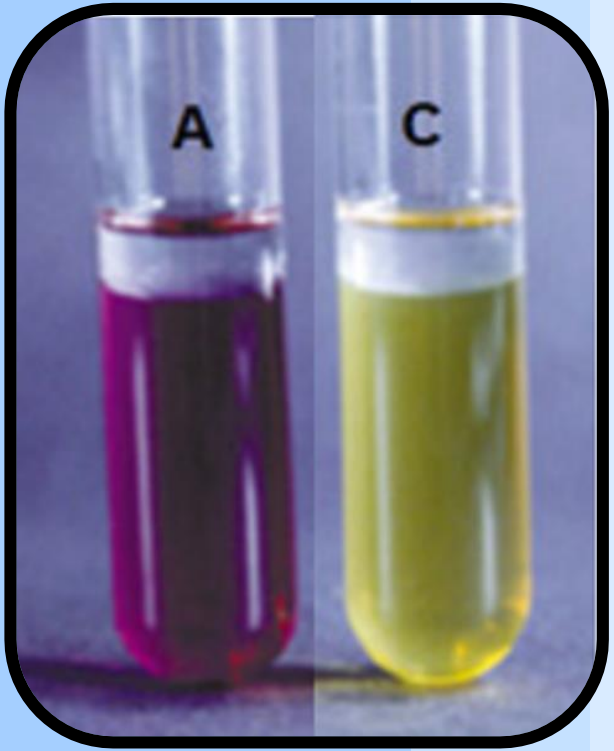
⇩

Ornithine

Source of carbon (energy for growth)

Providencia rettgeri & Morganella morgani (+ve)

P. rettgeri & M. morganii



8) Ornithine decarboxylase

Organism

ferment glucose

Ornithine

Bromocresol purple (Indicator)

Yellow

Purple

Glucose

Ornithine

9) The analytical profile index (API)

The analytical profile index (API)

(Biochemical tests for identification)

9) The analytical profile index (API)

Several API systems for different groups of organism

API 20E &API 20NE (Enterobacteria)

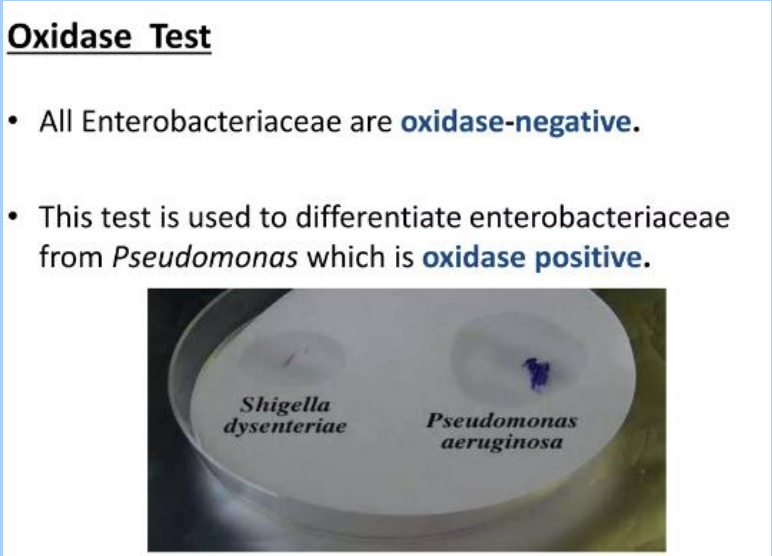
API 20 STREP (Streptococci) etc.

9) The analytical profile index (API)

Color change

Oxidase test

Some bacteria produce Oxidase enzyme Detection by adding few drops of colorless Oxidase reagent Colonies turn deep purple in color (positive)



Catalase test

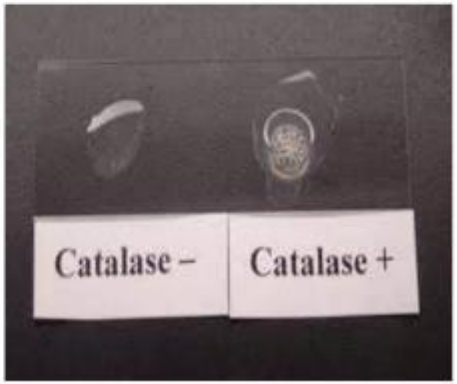
Some bacteria produce catalase enzyme

Addition of H2O2 lead to production of gas bubbles (O2 production)

- **Catalase test:**
  - Is used to differentiate between staphylococci(catalase +ve) and streptococci(catalase –ve).
  - Principle:

$$2\,H_2O_2 \xrightarrow{\text{Catalase enzyme}} 2\,H_2O + O_2$$

  - Procedure
    - Smear a colony of the organism to a slide
    - Drop H_2O_2 onto smear
    - Observe



Coagulase test

Coagulase test:

Some bacteria produce coagulase enzyme

Coagulase enzyme converts fibrinogen to fibrin (plasma clot)

Detected by slide or test tube method

Coagulase test

is used to differentiate *Staphylococcus aureus* from coagulase-negative staphylococci.

fibrinogen —(coagulase)→ fibrin (clot formation)