# Scheme of Medical bacteria

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

## Shape

### COCCi (Sperical)

Gram Stain

+ve
- Cluster
  - Staphylococci
- Chain or Pairs
  - Streptcocci
  - Enterococcus

-ve
- Pairs
  - Neisseria (Diplococci)

### Spiral (helical Shape)

- Treponema
- Borrelia
- Leptospira

### Bacilli (Rod Shape)

Not stained
- Zn Stain (TB)

Gram Stain

+ve

Non-spore
- Aerobic
  - Corina Bactrium Diphtheriae
  - Listeria
- Anaerobic
  - Lactobacilli

Spore
- Aerobic
  - Bacillus
- Anaerobic
  - Clostridium

-ve
- Enterobacteriaceae
- Vibro
- Campylobacter
- Helicobacter
- Pseudomonas
- Haemophilus
- Bordetella
- Brucella
- Legionella
- Gram-ve anaerobes

### Miscellaneous (2 or more Shapes)

- Mycoplasma
- chlamydia
- Rickettsia
- Coxiella
- Actinomycetes

# Biochemical Reactions

## Indole test

Liquid Medium has

Peptone (As nutrient)

Treptophane (to see IF the bacteria has the enzyme to break it)

↓ Tryptophanase

Indole

+

Kovac's Reagent

↓

+ve result

Red ring

## Methyl Red test

Liquid Medium has

Peptone (As nutrient)

Glucose (to see the ability of the bacteria to Ferment glucose Producing Mixture of acid

↓ incubate At 37°c For 48 h

Large amount of Mixed Acids

↓ test PH by MR indicator

| Low pH | high PH |
|---|---|
| acidic | Alkaline |
| <4 | >6 |
| Red | yellow |
| +ve | -ve |

IF MR → +ve then VP → -ve

Vice versa

IF bacteria Produce Acids it won't produce acetoin

Vice versa

## Voges-Prokauer test

Medium has

Peptone (As nutrient)

Glucose (to see the ability of the bacteria to Ferment glucose to produce Acetoin)

↓ incubate At 37° For 48 h

Acetyl Methyl Carbinol (Acetoin)

↓ test it by:
- α-naphthol
- 40% KOH

Diacetyl

↓ +ve

Red ring will Form

## Citrate utilization test

Medium has

Citrate (As only Source of carbon to See if the bacteria will metabolize Citrate) → $CO_2$

incubated at 37°c For 48h

↓ Citrate lyase + $Na^+$

$Na_2CO_3$ (sodium carbonate) increases pH

↓ test PH by bromothymol blue indicator

| + ve | -ve |
|---|---|
| Blue | Green |
| ↑PH | Neutral PH |
| >7 | |

## Urease test

Medium has

Urea (to see if the bactria can break it)

↓ Urease

Ammonia (Alkaline)

↓ test it with Phenol red indicator

| +ve | -ve |
|---|---|
| Alkaline | basic |
| Pink | Yellow |

# Biochemical Reactions

## Phenylalanine deaminase

Medium has

Bacteria from Enterobacteriaceae Family (G-ve bacilli)

(has phenylalanine to distinguse between

Proteus (has the enzyme) and

Salmonella + Shigella (doesn't has the enzyme)

↓ Phenylalanine deaminase

Phenyl pyruvic Acid + $NH_3$

↓ Add Ferric chloride As indicator

+ve

Green

## Triple Sugar Iron (TSI)

- Phenol red indicator

Medium has:

Semi-solid (gelatinous) Obliquely

- 0.1 % glucose
- 1 % lactose
- 1 % sucrose
- Ferrous sulfate

### (A/A)

inoculation

↓

Ferment glucose

↓

Acid

↓ Phenol Red

Yellow color

↓

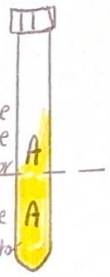
Then start to lactose or sucrose

↓ IF Fermentation takes place

Acid

↓

IF their is break in the medium (A|A) detection of gas produced by bacteria




### (K/A)

inoculation

↓

Ferment glucose

↓

Acid

↓ Phenol Red

Yellow color

↓

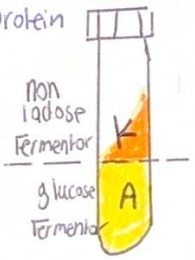
No glucose Frementer

↓

Alkaline media Utalise protein

↓ Phenol Red

Revert to Red




### $H_2S$ Production (K/K)

Bacteria - has the ability to reduce + Sulfur

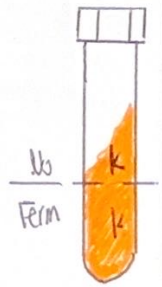
↓

Hydrogen Sulfied ($H_2S$)

↓ Iron From TSI Medium

Ferric sulfied

Black




## Ornithine decarboxylase

Medium has:

- Ornithine (to see IF the bacteria has the enzyme to break it)

(Soure of carbon) energy For growth

- glucose (to be Fermented First)

↓

Ornithine decarboxylase

↓

Removal of -coolt Putrescine

↓

Bromo-cresol Purple indicatore

+ve

Prlorganella Morgani

Purple

-ve

Providencia rettgeri

Yellow

# Biochemical Reactions

## The analytic Profile Index (API)

Commerical stripe
Contains Multiple tubes
For various biochemical reactions to identify different bacterias At once

inoculate each tube

↓

We place them in an incubator

↓ After 24 hrs

Record the color changes according to the Catalog e. This Allow us to identify the type of Bacteria

## Oxidase test

Used to differentiate

enterobacteriaceae (G-ve bacilli) (Oxidase-negtive)

From

Psedomonas (G-ve bacilli) (Oxidase-postive)

Bacteria

↓ Add Few drops of colorless Oxidase reagent Colonies

| +ve | -ve |
|---|---|
| Deep Purple | No color |
| Psedomonas | enterobacteriaceae |

## Catalase test

Used to differeniate between Staphylococci (Catalase +ve) and Streptococci (Catalase -ve)

### Procedure

- Smear a colony of the organism to a slide
- Drop $H_2O_2$ to smear
- Observe bubbling

$H_2O_2 \xrightarrow[\text{enzyme}]{\text{Catalase}} 2H_2O + O_2$ (↓ gas bubbles +ve catalase)

| Characteristic | Staphylococci | Streptococci |
|---|---|---|
| Catalse test | +ve | -ve |
| Morphology | Forms clusters | Chains |
| Gram Staining | G +ve | G -ve |
| Shape | Cocci (Sperical bacteria) | Cocci |

## Coagulase test

Used to distinguish between the different members of Staphylococci Family

Staph. aureus (Coagulase +ve)

From other Staphylococci

(Coagulase -ve)

Fibrinogen

↓ Coagulase

Fibrin (Clot Formation) using testtube

Staph. aureus

- gram – postive
- Cocci Shaped
- Forms Clusters
- Catalase +ve
- Coagulase +ve