Practical
Medical Bacteriology

Lab 8

Laboratory Diagnosis of Enterobacteriaceae spp:

E. coli & *Klebsiella*

2021/2022

General Characters of Enterobacteriaceae spp

- Short Gram-negative bacilli (coccobacilli or as straight).
- Their natural habitat is the intestinal tract of humans and animals.
- Many genera like (*Escherichia*, *Shigella*, *Salmonella*, *Enterobacter*, *Klebsiella*, *Serratia*, *Proteus*, and others).
- Most motile with peritrichous flagella except *Shigella*, *Klebsiella* and *Yersinia* are non-motile.
- Non–spore-forming, facultative anaerobic or aerobic microbes.
- Some are opportunistic pathogens and others regularly pathogenic for humans.
- Ferment a wide range of carbohydrates.
- Distinguishing properties associated with all enterobacteriaceae: Ferment glucose, reduce nitrate to nitrite and oxidase negative

Laboratory diagnosis

1. Specimen:

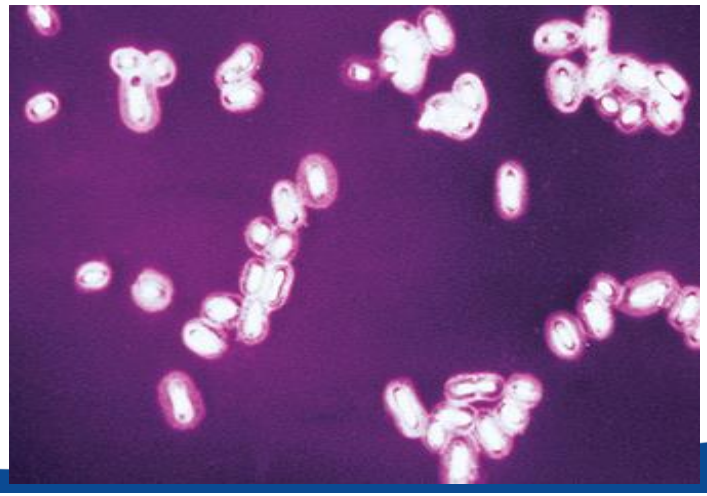
- ***Escherichia coli*:** according to disease
  - Urine, pus and stool, pus, blood, stool and CSF
- ***Klebsiella*:** according to disease
  - Sputum, urine, pus.

2. Microscopic morphology:

- ***Escherichia coli*:** Gram negative rods, coccobacilli, or straight rods with rounded ends.
- ***Klebsiella*:** Gram negative rods, with unstained halo around them due to the presence of the capsule.

A

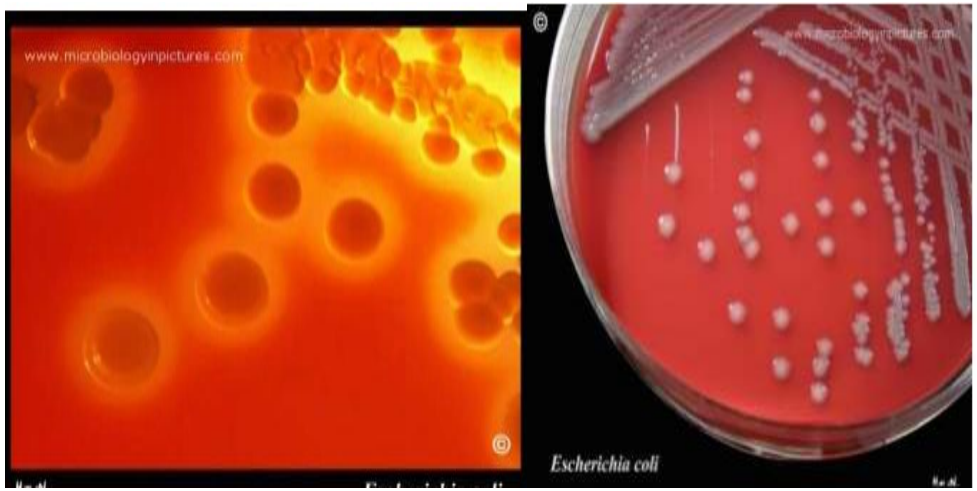
FIGURE 15-1 A: Gram stain of *Escherichia coli*. Original magnificatio



Laboratory diagnosis

3. Cultural characteristics

- **On blood agar**
  - ***Escherichia coli*:** Colonies are medium sized, smooth, round, grayish-white colonies on blood agar. Some strains produce β hemolysis.

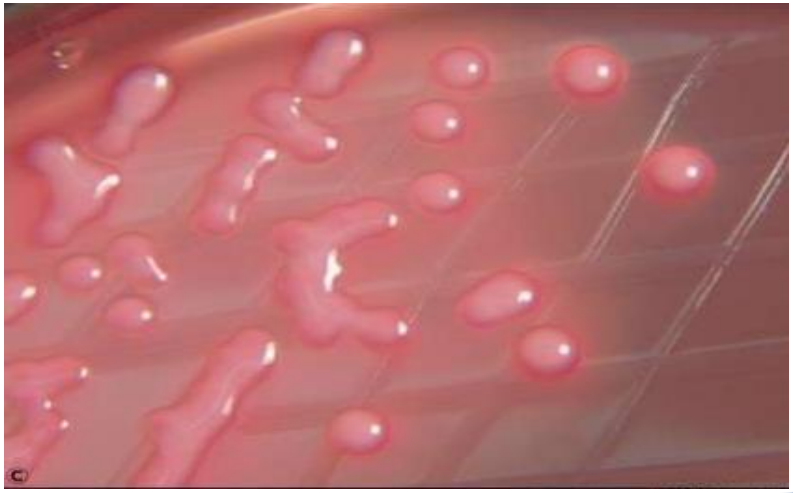


  - ***Klebsiella*:** Slimy appearance of the colonies on blood agar.

Laboratory diagnosis

3. Cultural characteristics

- **On MacConkey agar**
  - ***Escherichia coli*:** deep red colonies are produced as the organism is ferment lactose.
  - ***Klebsiella*:** it shows red/pink mucoid lactose fermenting colonies.

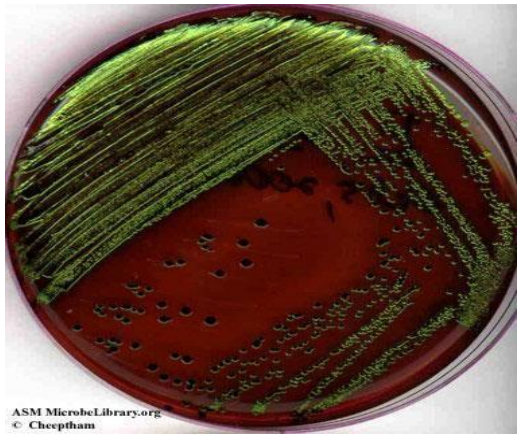


Laboratory diagnosis

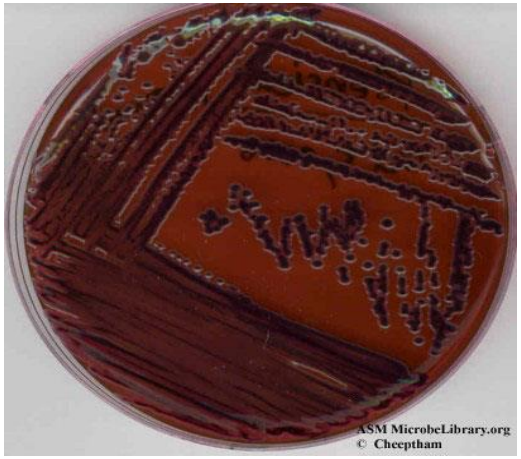
3. Cultural characteristics

❑ **On Eosin Methylene Blue agar**

- ***Escherichia coli*:** produces black colonies with greenish-black metallic sheen.



- ***Klebsiella*:** produces large, mucoid, pink to purple colonies with no metallic green sheen on EMB agar.

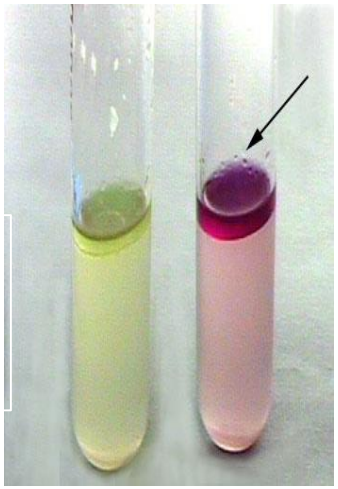


Laboratory diagnosis

4. Biochemical reactions A. IMViC

Escherichia coli

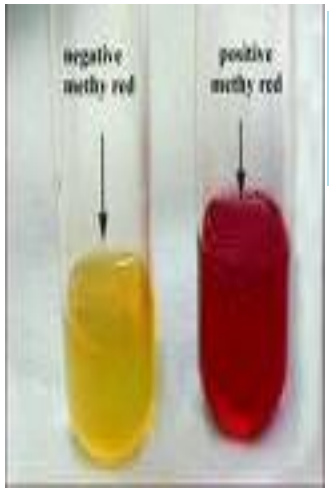
+ +- -



Indole test

Positive result *E. coli*

Negative result *Klebsiella*



Klebsiella species

- -+ +

Methyl Red test

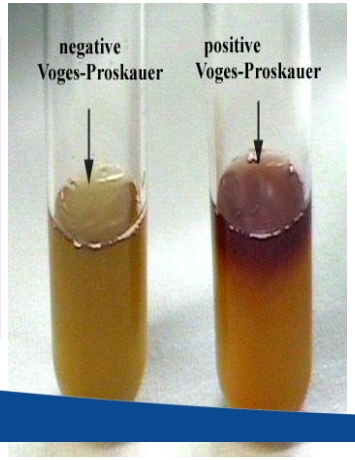
Red: Positive MR (*E. coli*)

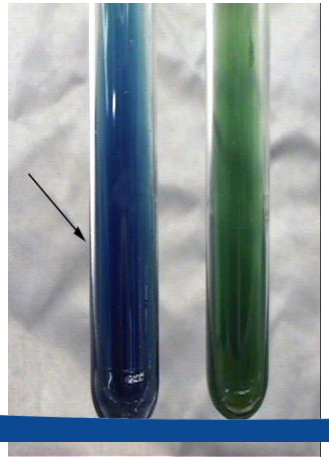
Yellow or orange: Negative MR (*Klebsiella*)

Voges-Proskauer test

Pink: Positive VP (*Klebsiella*)

No change: Negative VP (*E. coli*)





Citrate utilization test

Positive *Klebsiella*

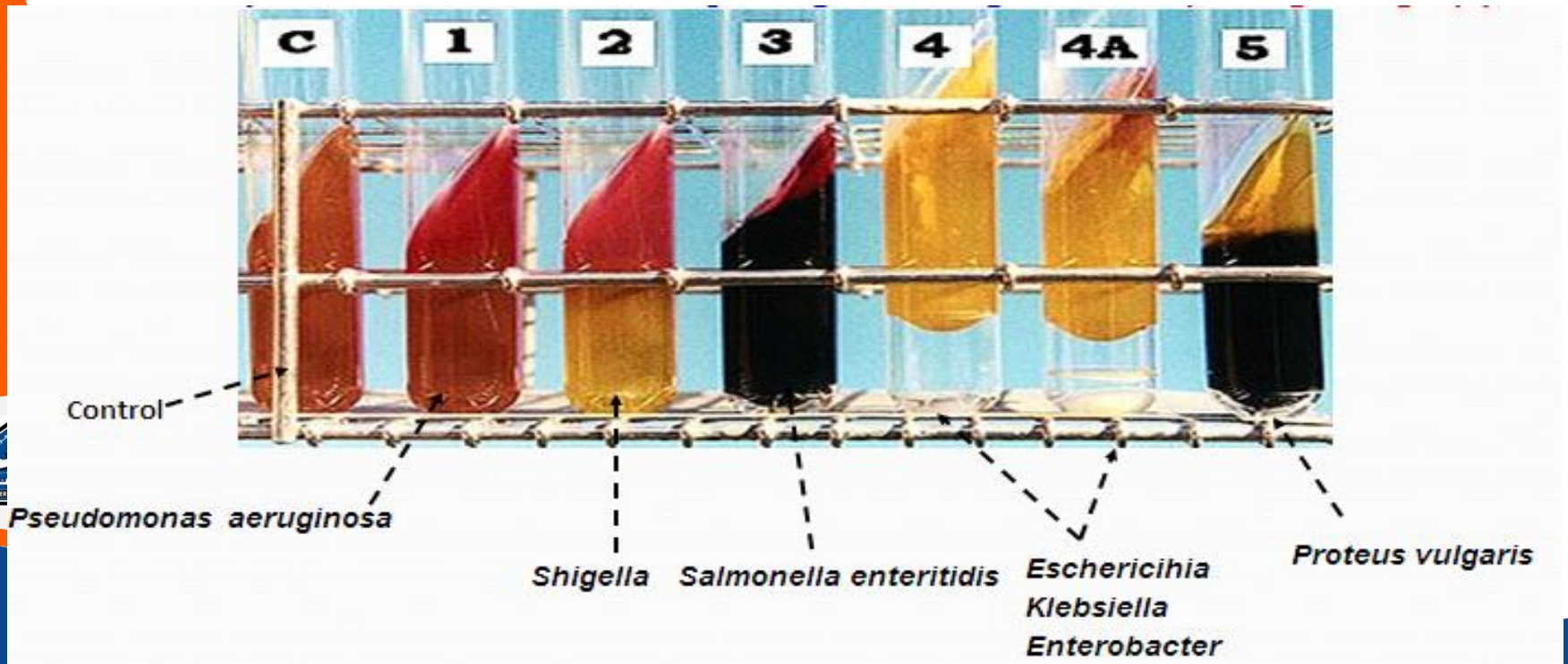
Negative *E. coli*

Laboratory diagnosis

4. Biochemical reactions

B. Carbohydrates fermentation in triple sugar iron agar

- ***Escherichia coli*: A/A /g+ / H_2S –**
- ***Klebsiella species*: A/A /g++ / H_2S -**



Laboratory diagnosis

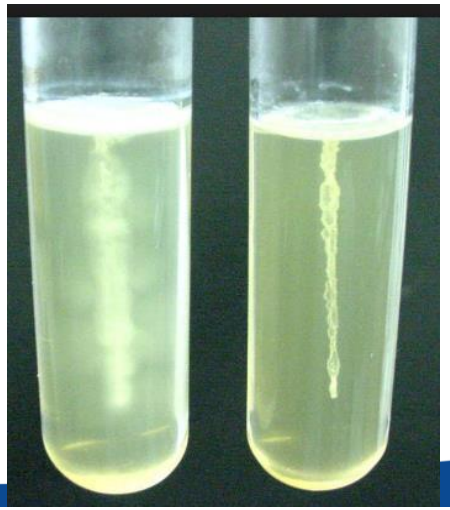
4. Biochemical reactions

C. Urease test

- ***Escherichia coli*: Negative**
- ***Klebsiella species*: Positive**

Motility test:

- ***Escherichia coli*: Motile**
- ***Klebsiella species*: Non-motile**



Laboratory diagnosis

Identification by API20E