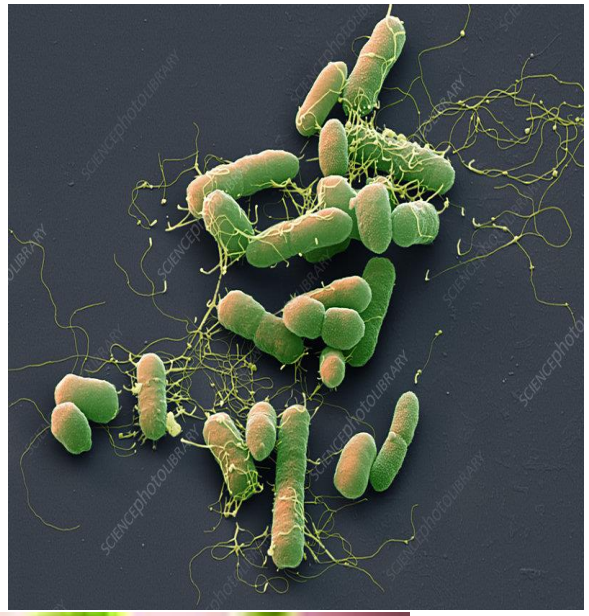
Practical Microbiology

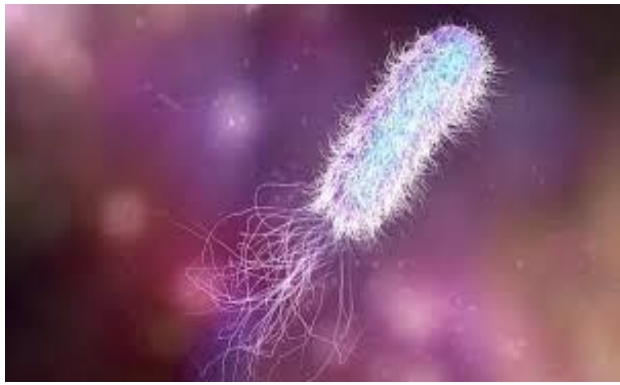
College of Dentistry
University of Babylon
"3"stage

Pseudomonas &Proteus



Lecturer
Dr . Doaa Adil Almusawi

What are Pseudomonas



- **Family Pseudomonadaceae**
  – Aerobic, non-spore forming Gram negative
- - straight or slightly curved rod
  – Motile with polar flagella
  – **Non-fermenters**
  – **Catalase and oxidase positive**
  – Pigment producing bacteria.
  – Mostly causes Hospital acquired infection.
  – Opportunistic pathogens, majorly found in soil, water

  – They are highly resistant to chemical disinfectants, salts, antibiotics.

✓ **Most important species P. aeruginosa**

- Medically important *Pseudomonas*:

P. aeruginosa.

- present in small numbers in the normal intestine flora and on the skin.
- Commonly present in moist environments in hospitals.
- It is primarily a nosocomial pathogen.

Pseudomonas aeruginosa

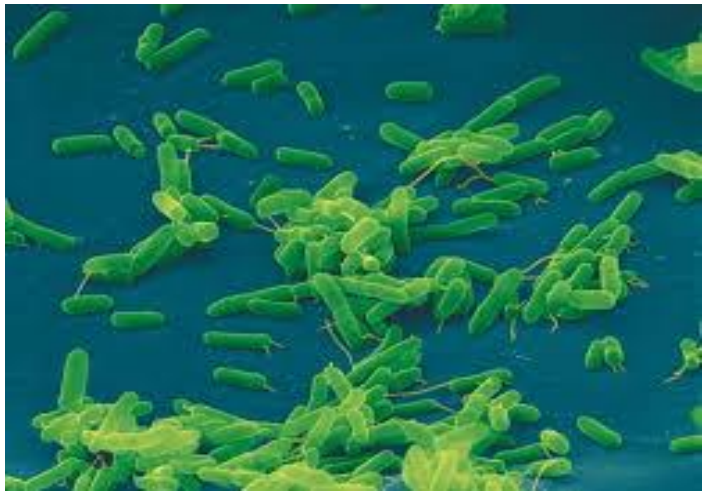
- *Morphology*

✓ They are slender gram negative bacillus

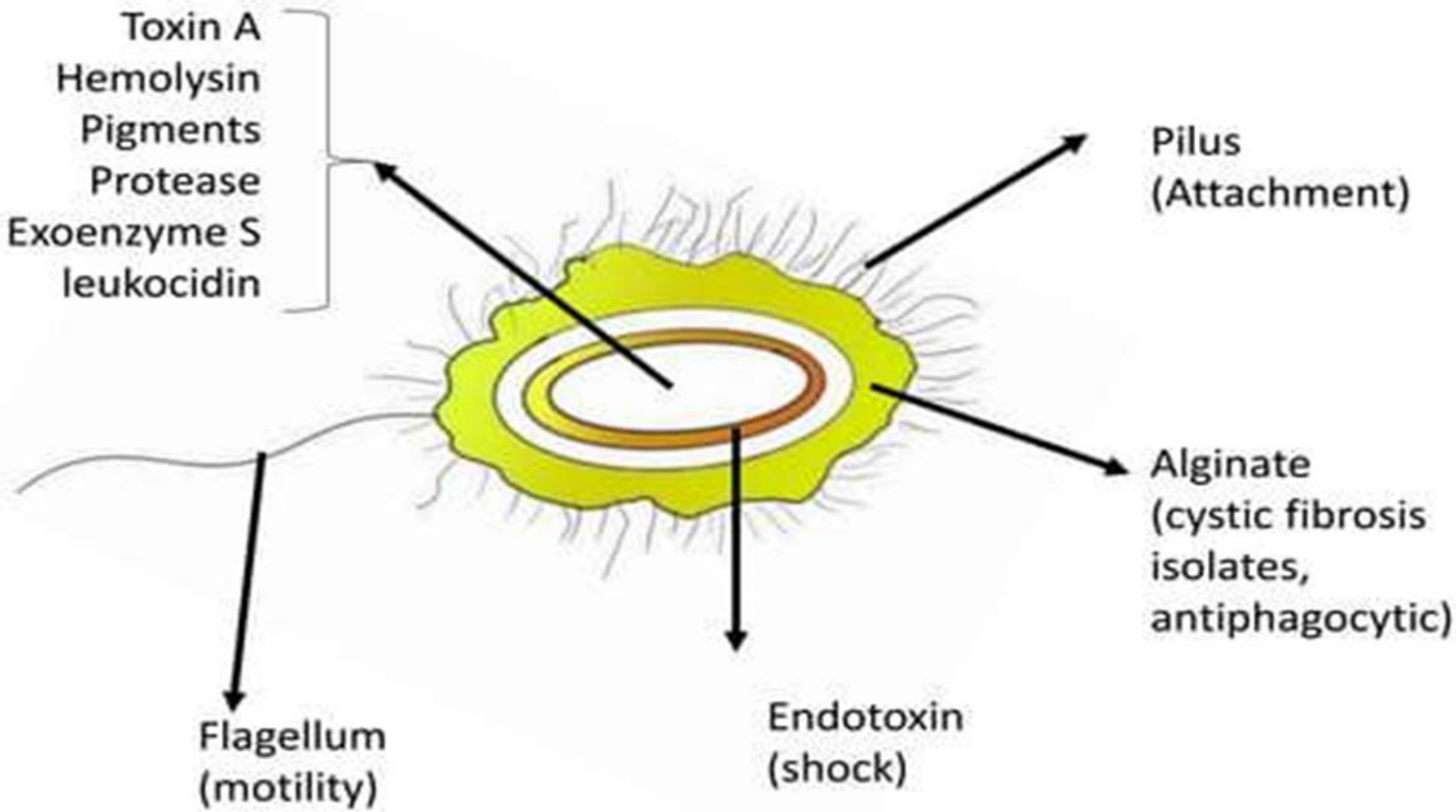
✓ Actively motile by polar flagella and some strains have Tow or three flagella .

✓ Grow well at 42C

✓ Pili aids bacterial attachment, thereby promoting Colonization



Pseudomonas aeruginosa



Structure and pathogenic mechanisms of *P aeruginosa*.

Pathogenesis

P. Aeruginosa can produce a lot of antigens some of them are

-Toxin A
-endotoxin
-Hemolysin
-pigments: fluorescein , pyocyanin
-leukocidine
-exoenzyme

- **Almost all strains of Aeruginosa are hemolytic on blood agar**
- **Leukocidine (also called cytotoxin) damage lymphocyte**
- **Toxin A may be a major virulence factor of p. Aeruginosa**
- **Most patients survive of p. aeruginosa sepsis have elevated levels of antitoxin A antibody**

Clinical manifestation

P. Aeruginosa causes various infections :

- ✓ Skin infection and Burns ,wound
- ✓ UTI
- ✓ Bacteremia
- ✓ Respiratory infection : pneumonia
- ✓ Endocarditis

Pigment production

- can produce pigments, such as:
- Pyocyanine (blue-green)
- Pyoverdin (fluorescent yellow- greenish pigment)
- Pyorubrin (red)
- Pyomelanin (brown)

Cultural Characters

Obligate aerobe

- Growth occurs at wide range of temperatures

-**42^{o}C** the optimum being 37^{o}C

- Growth on ordinary media producing large opaque irregular colonies .
- In broth forms dense turbidity with surface pellicle.

.

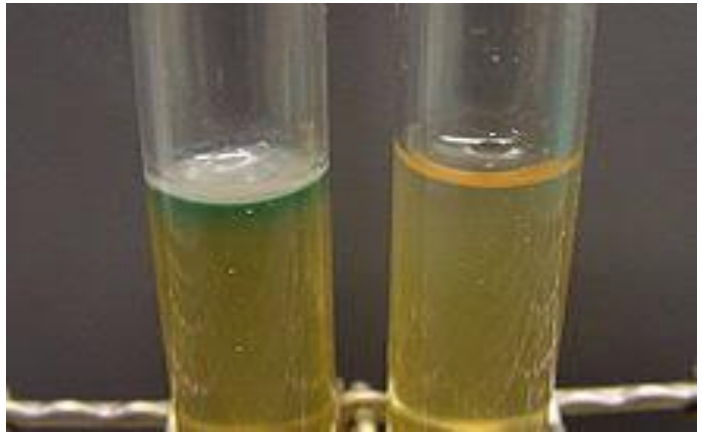
Culture character

- Form smooth and round colonies.
- **Fluorescent greenish colour.**
- production of fruity odor (grape-like) .
- Inability to ferment lactose.

On Culture media

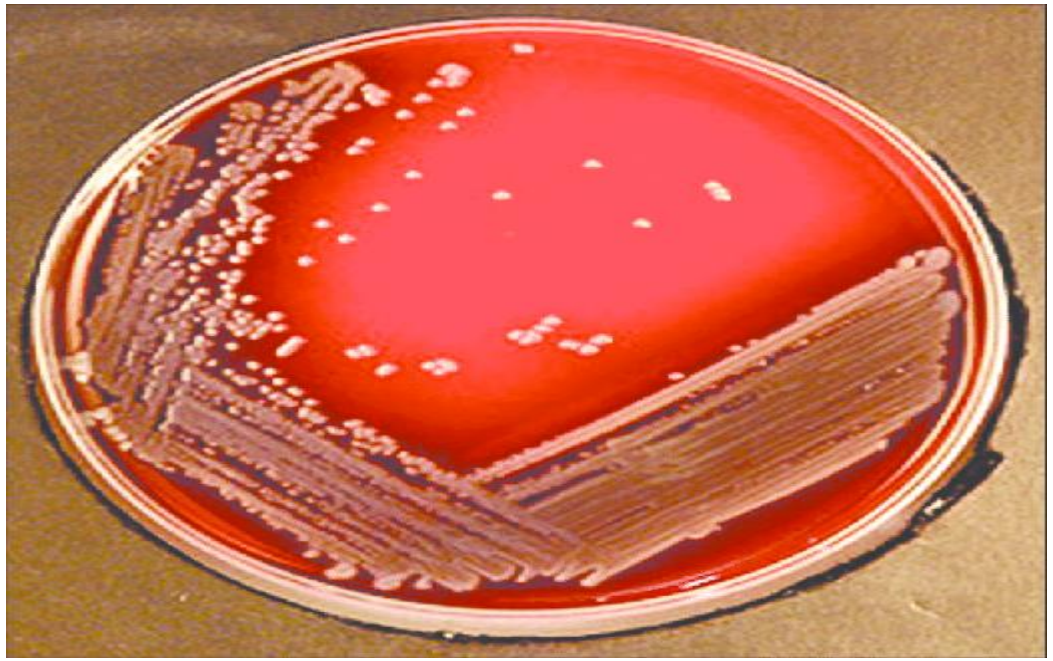
Nutrient agar

- . Colonies are smooth, large, translucent.
- Produce sweetish aromatic odor
- Greenish blue pigment diffuses



❖Growth on

- **Blood agar**
  - ➢Similar to nutrient agar
- ➢ Grayish colony
- ➢ Many are hemolytic(Beta hemolysis)

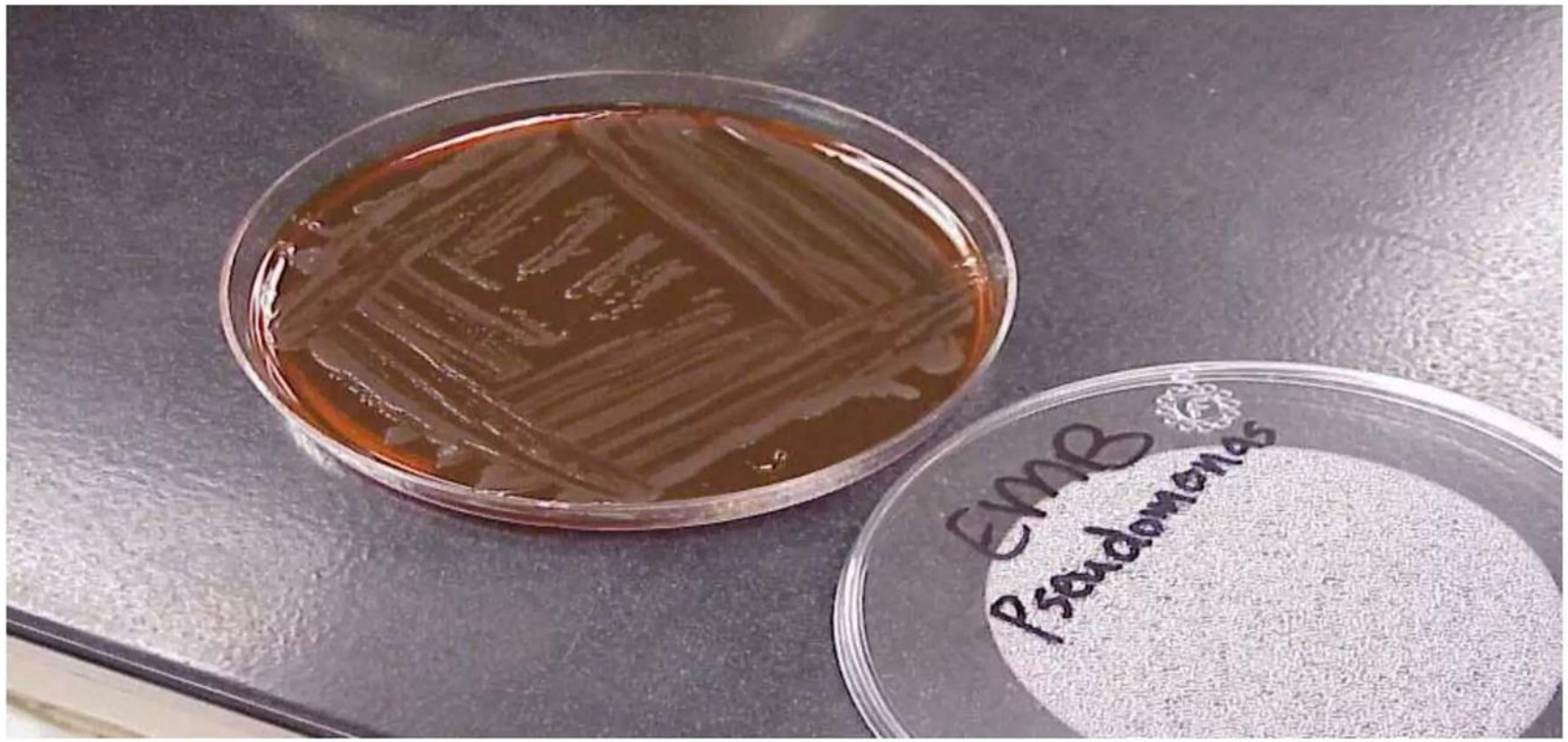


On MacConkey agar

- Non-lactose fermenting (colourless colonies)

On EMB

- Non lactose fermenting



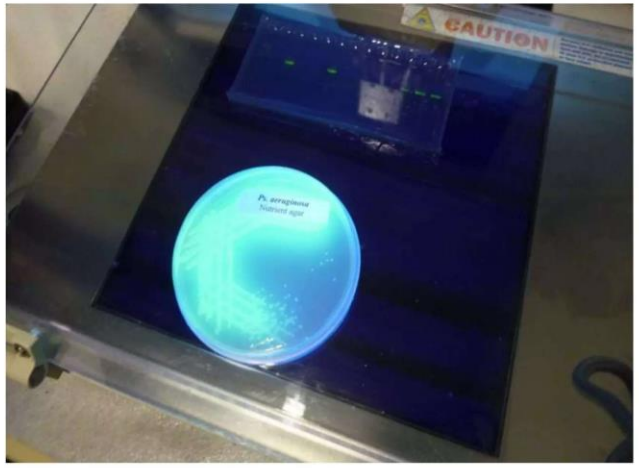
Cetrimide agar

- selective media

Is a type of agar used for the selective isolation of the gram negative bacterium pseudomonas aeruginosa.

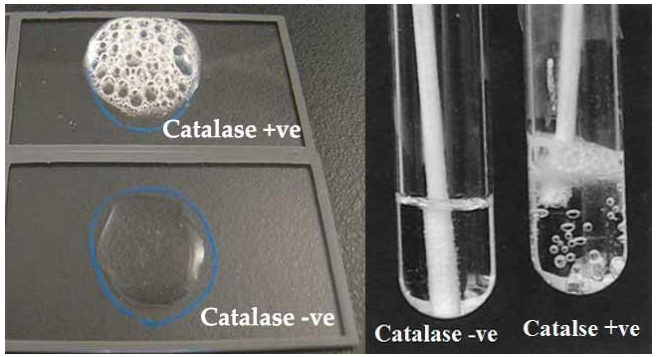
As the name suggests, it contains cetrimide, which is the selective agent against alternate microbial

cetrimide also enhance the production of pseudomonas pigments such as pyocyanin and fluorescein , which show characteristic of blue –green or yellow –green color respectively



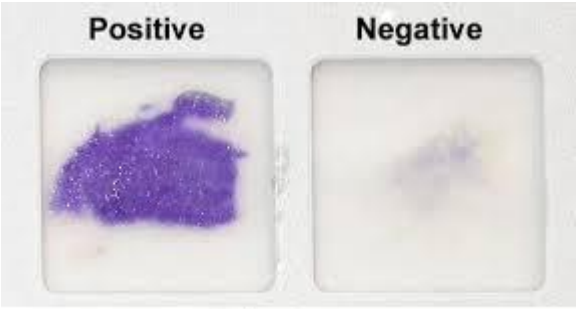
Pseudomonas aeruginosa fluorescence under UV illumination

Biochemical reactions



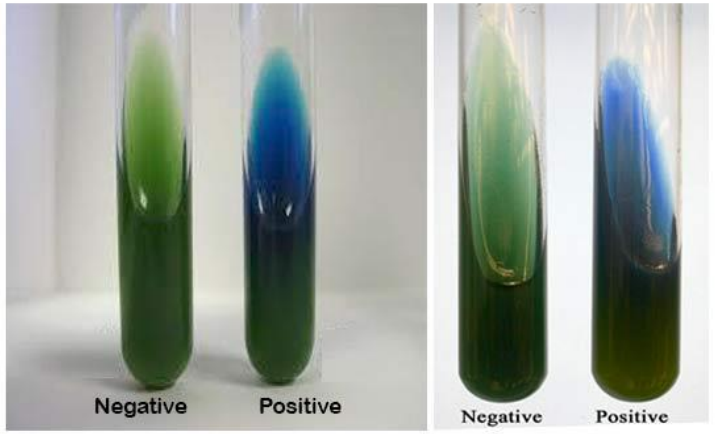
Catalase-positive

- Indole, MR and VP and H2Stests are **negative**
- **Oxidase-positive**
- **Nitrate reduction-positive**
- **Citrate test-positive**



Oxidase Test

Citrate Utilization Test



Laboratory diagnosis

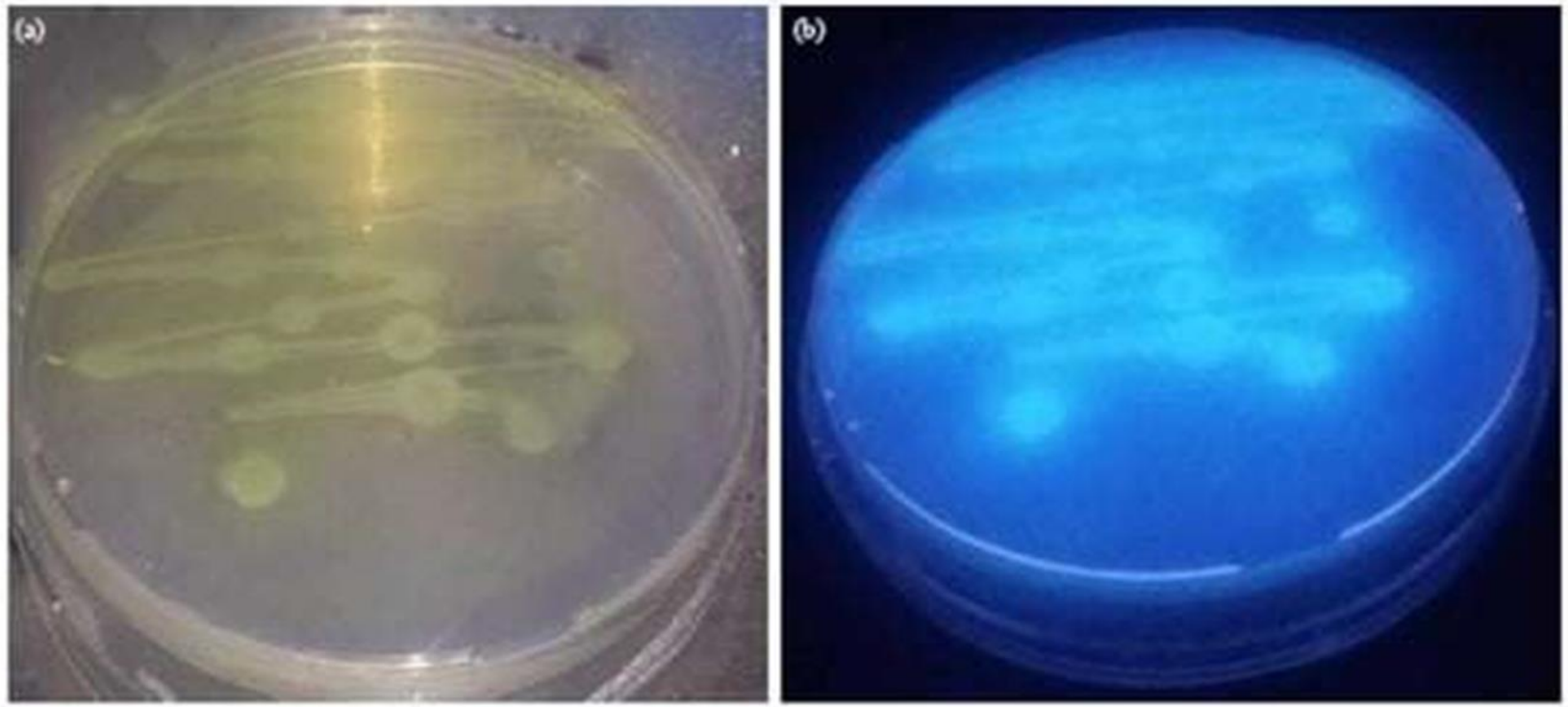
Specimen:

- Wound discharge
- sputum
- Blood
- Urine
- CSF

Diagnosis

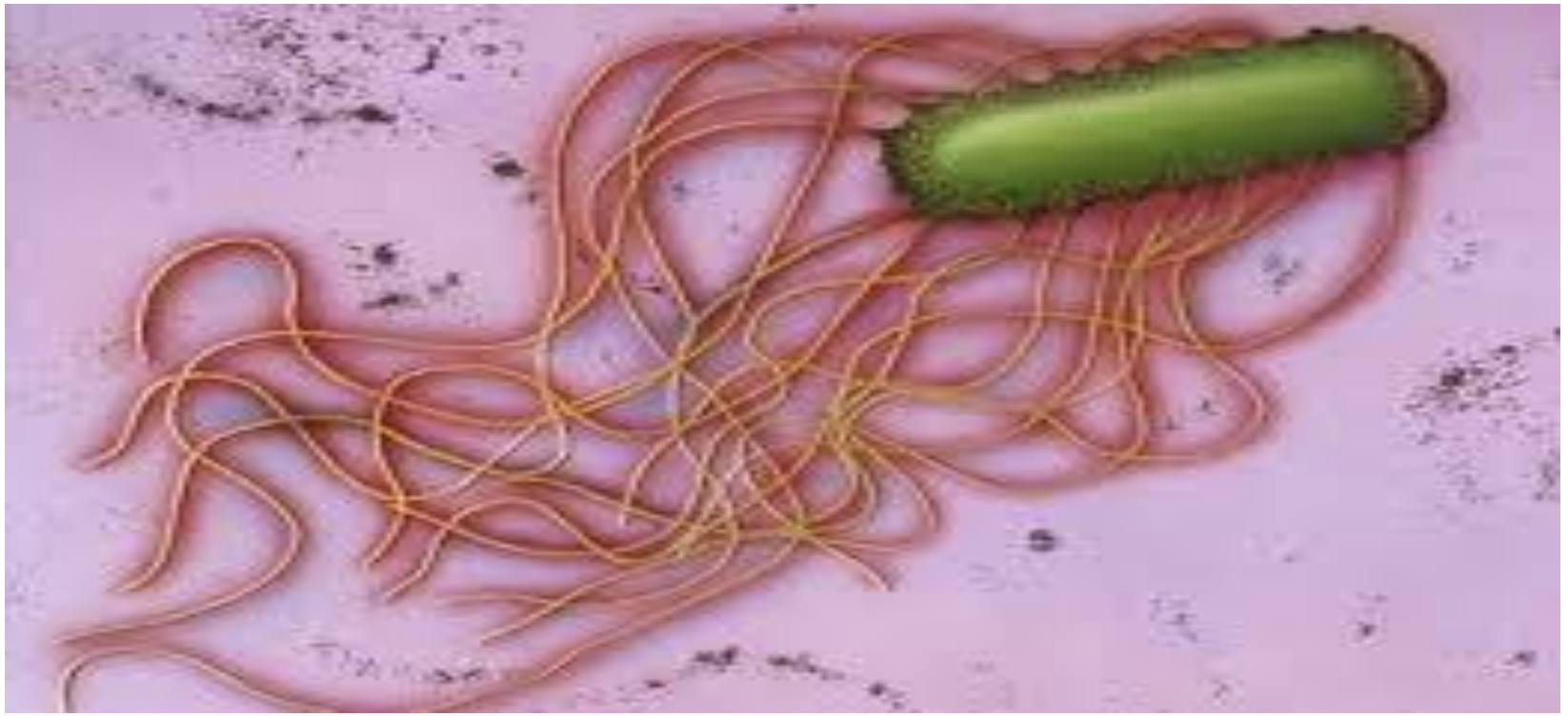
- isolation and laboratory identification
- identified on the basis of its **Gram morphology**, **inability to ferment lactose, a positive oxidase reaction, its fruity odor, and its ability to grow at 4 2° C**
- **Fluorescence under ultraviolet radiation** helps in early identification of P aeruginosa colonies and also is useful in suggesting its presence in wounds

Diagnosis



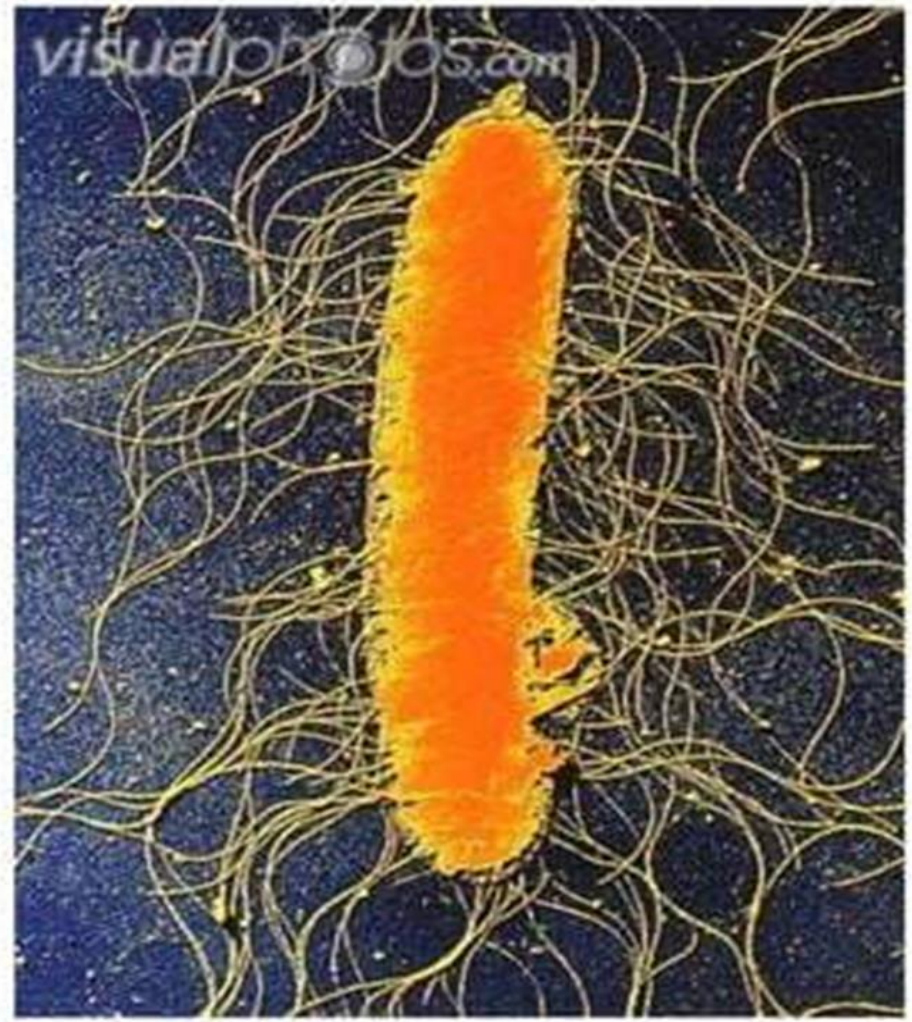
Pseudomonas aeruginosa on Pseudomonas cetrimide agar, (a) P. aeruginosa under light lamp showing green pyocyanin pigment and (b) P. aeruginosa give fluorescence under UV lamp

Proteus spp



General character:

- Rod shape.
- gram-negative.
- **motile.**
- non-capsulated .
- Possessing peritrichous flagella.
- **Non-lactose fermenting.**



- Proteus species widespread in the environment
- Are normal in habitants of the human intestine
- facultative anaerobic
- oxidative -negative
- Nitrate- positive

- The two species to most commonly produce infections in humans are

-proteus mirabilis

-proteus vulgaris

Pathology

P. Mirabilis

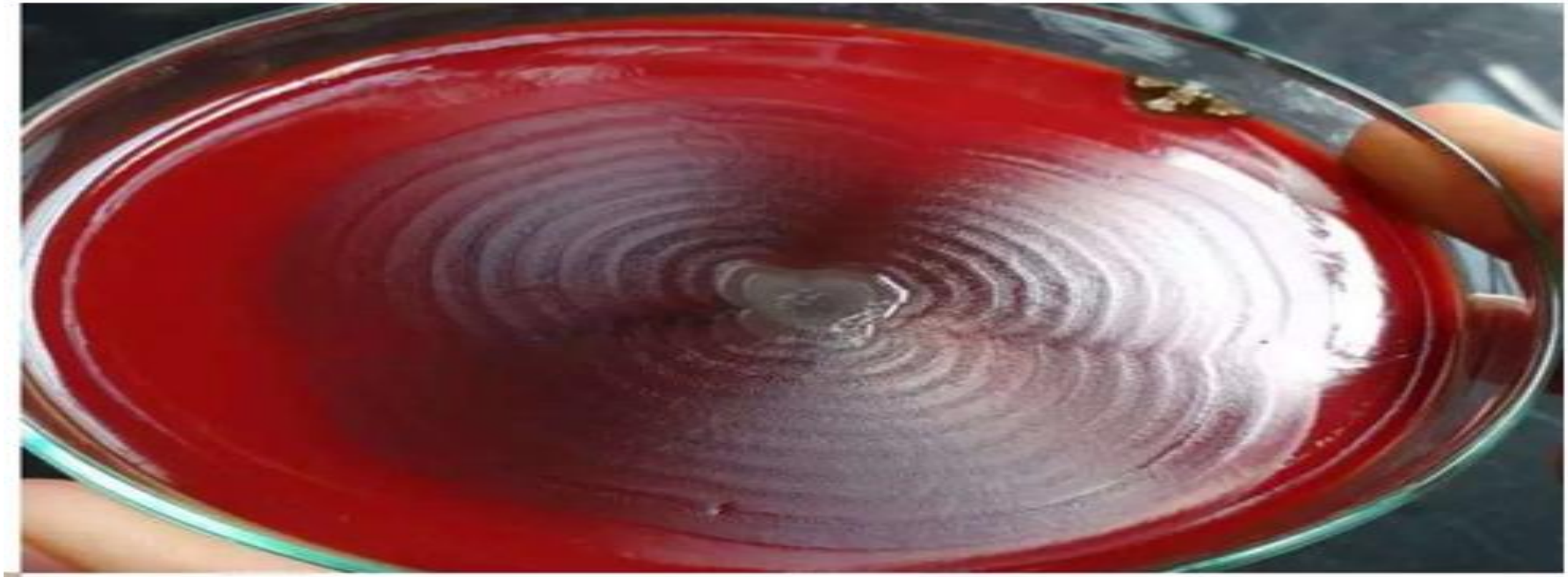
P. Mirabilis causes 90% of all proteus infection in human

P. Mirabilis cause urinary tract infections (make stones)

And occasionally others infections such as : bloodstream infection and respiratory tract infections .

P. Vulgaris

P. Vulgaris is probably more frequently implicate in wound and soft tissue infection than UTI



- They have ability to swarm over the surface of media. (motile)
- H2S positive
- Non Lactose ferment.
- **Urease positive.**
- Oxidase negative
- Catalase positive.

Culture characteristic

On blood agar

Proteus mirabilis dose not form distinctive colonies on blood agar, instead the bacteria swarm across the surface of agar.

Proteus produce very distinct fishy odour

- Discontinuous swarming produces concentric circles around the point of inoculation.

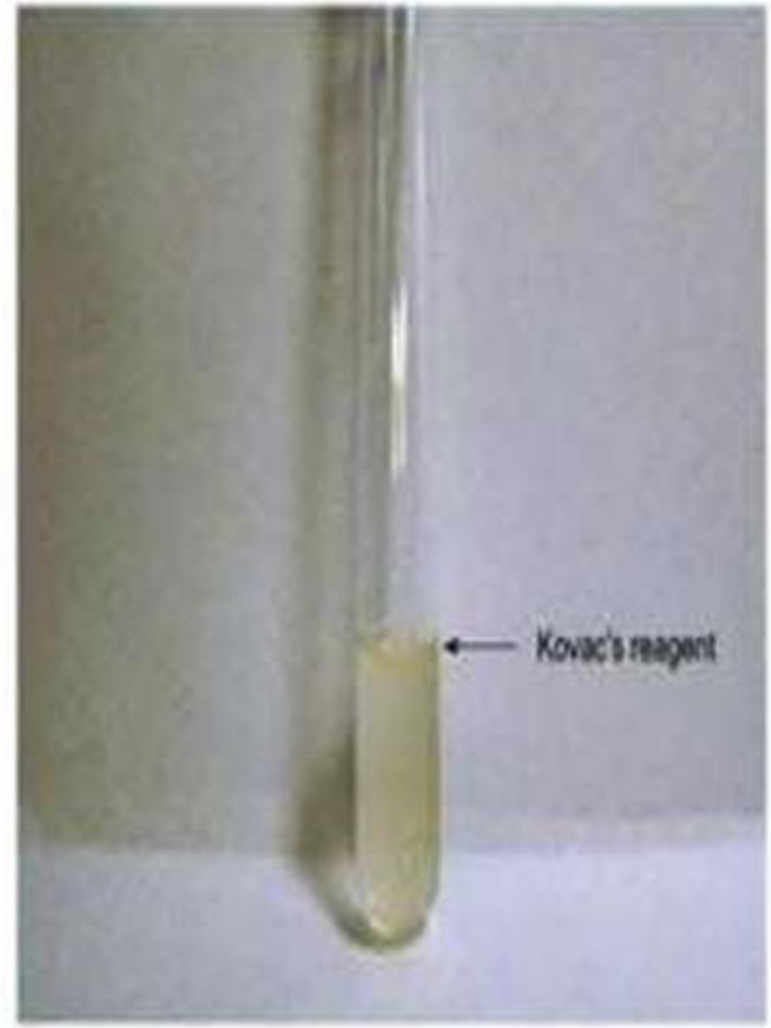
Biochemical test

- **Indole test**

  is used to differentiate

P.mirabilis (-ve) from

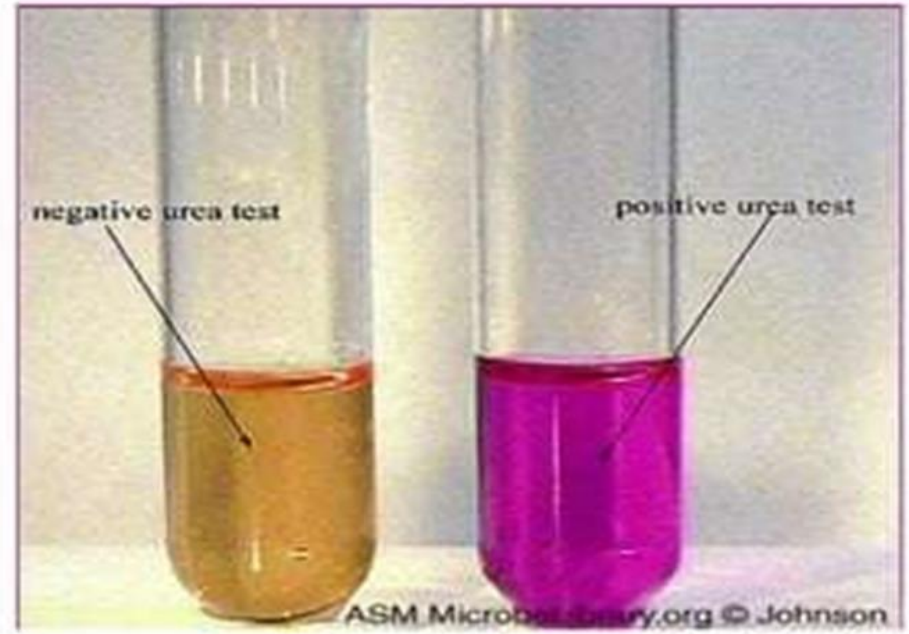
P. vulgares(+ve)



Urease test

- **positive urease** (which is the fundamental test to differentiate *Proteus* from *Salmonella*).

- Most strains produce a **powerful urease enzyme**, which rapidly hydrolyzes urea to ammonia and carbon monoxide.



- oxidase-negative
- Catalase-positive.

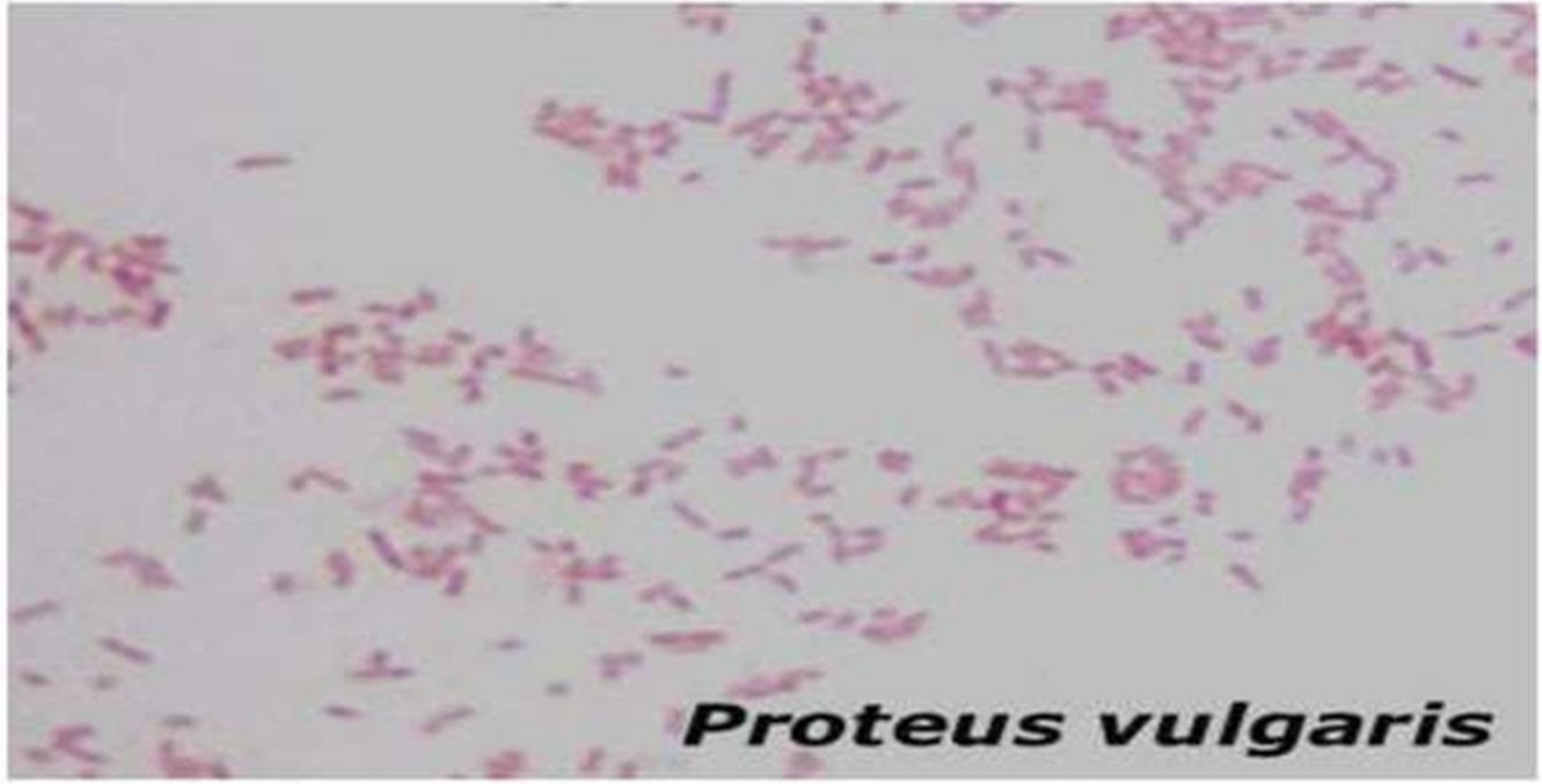
Laboratory diagnosis

Specimen

- Urine.
- Pus.
- blood.
- ear discharge

Gram stain

- Gram-negative rods

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