

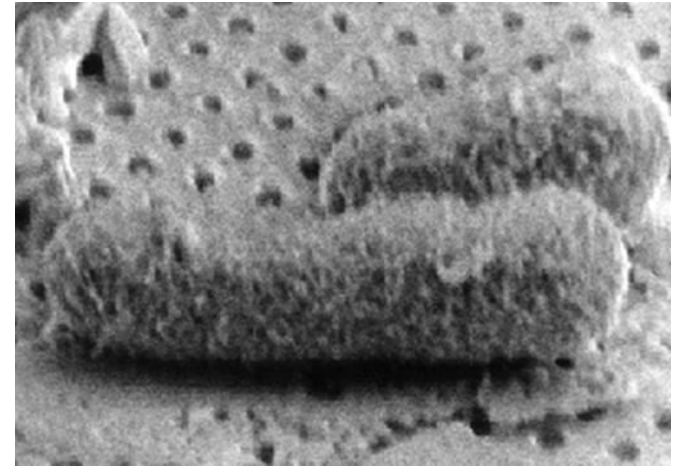
Gram Negative Rods

Escherichia Coli

Pseudomonas aeruginosa

What is *Escherichia Coli*?

- It belongs to the Family Enterobacteriaceae.
- From Greek word enterikos (intestines)
- These organisms happily inhabit the intestinal tract of most warm blooded animals.



—
←0.001 mm→

What is *Escherichia Coli*?

- Normal flora of the mouth and intestine
- There are more than 700 different serotypes of *E. coli* distinguished by different surface proteins and polysaccharides

Good *E.coli* (beneficial *E.coli*)

1. Protects the intestinal tract from bacterial infection
2. Assists in digestion
3. Produces our main source of vitamins B12 and K
4. Colonizes newborns GI tract within hours after birth
5. Lives symbiotically with us (we help them to live, and they help us to live)

Bad *E.coli*

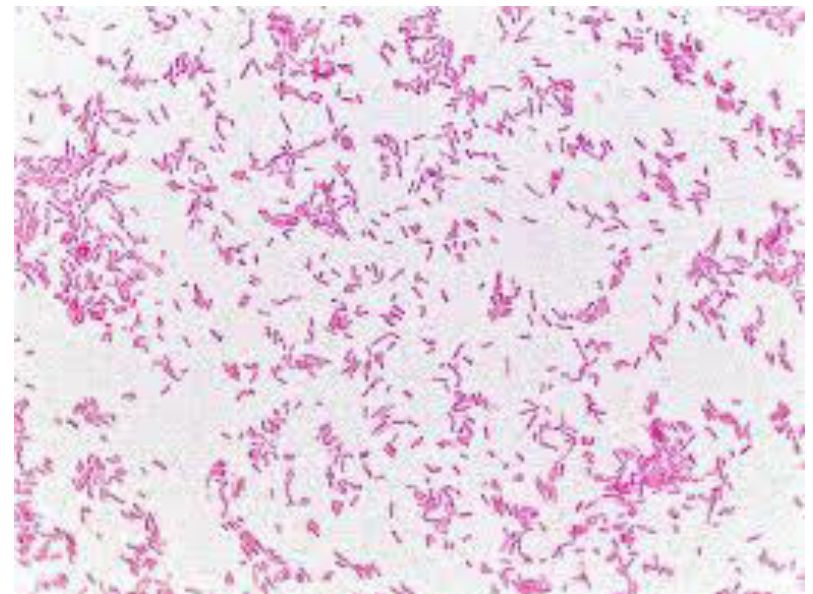
- While most *E. coli* are good for you, there are a few strains of *E. coli* that are harmful to humans.
- There are ~ 5 billion bacteria in your intestines alone, It is possible to get an individual strain of *E. coli* that is harmful to you.

***E. coli* O157:H7**

- Most notorious strain of bad *E. coli*
- Produces a powerful toxin and can cause severe illness
- Most dangerous for little kids, the elderly, and the sick
- Damages the intestines, causes bloody diarrhea, and other complications

Morphology of *E.coli*

- Gram - ve Straight rods
- Appear in singles or in pairs,
- Motile by peritrichate flagella.
- Very few strains non motile



Biochemical Tests

- Oxidase test – Negative
- Indole test - Positive

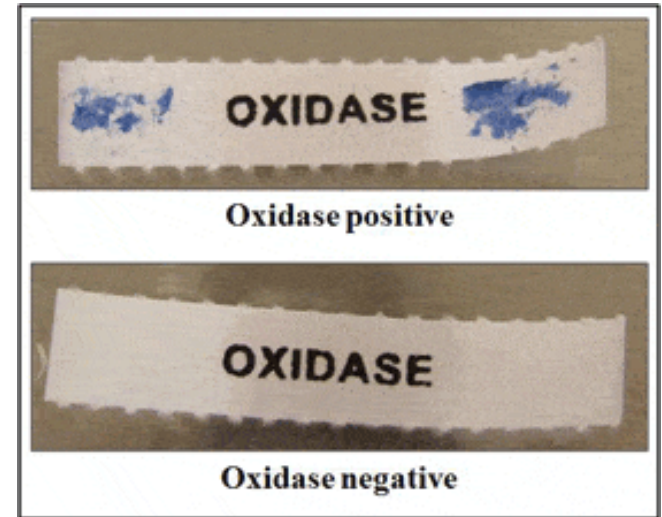
Oxidase Test

- **Aim of this test:** to identify bacteria that produce cytochrome c oxidase enzyme.
- **Reagent:** tetramethyl-p-phenylenediamine
- **Principle:**
 - ✓ If bacteria have the cytochrome c oxidase enzyme >> oxidizes the reagent to >>> indophenols (purple color) end product.
 - ✓ When the enzyme is not present >> the reagent remains reduced and is >> colorless.

Oxidase Test

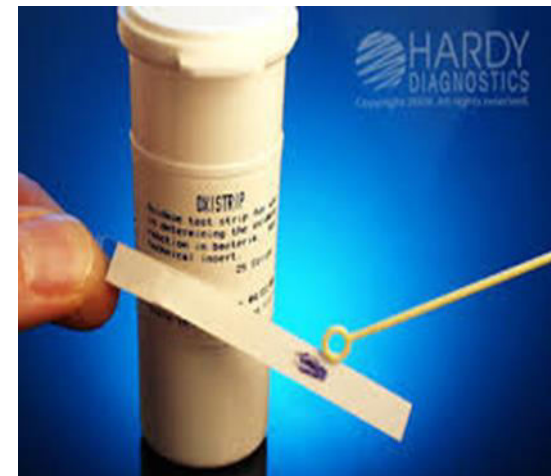
Procedure:

- Add 2-3 drops of oxidase reagent on filter paper
- Transfer bacterial colony to the filter paper



Result:

- Purple color>> oxidase positive
- Colorless>> oxidase negative

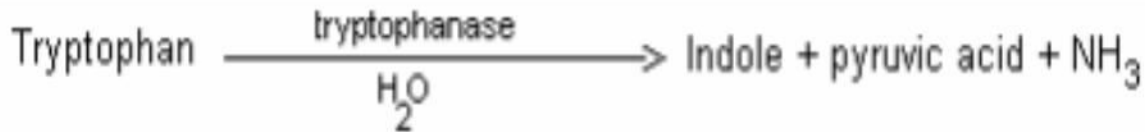


Indole Test

- **Aim of this test:** to identify bacteria that produce tryptophanase enzyme.

- **Principle:**

Tests the ability of bacteria to produce indole from the amino acid Tryptophan using the enzyme tryptophanase.



- **Reagent:** prooduction of indole is detected using Kovac's reagent whoch react with indole and give red color

Indole Test

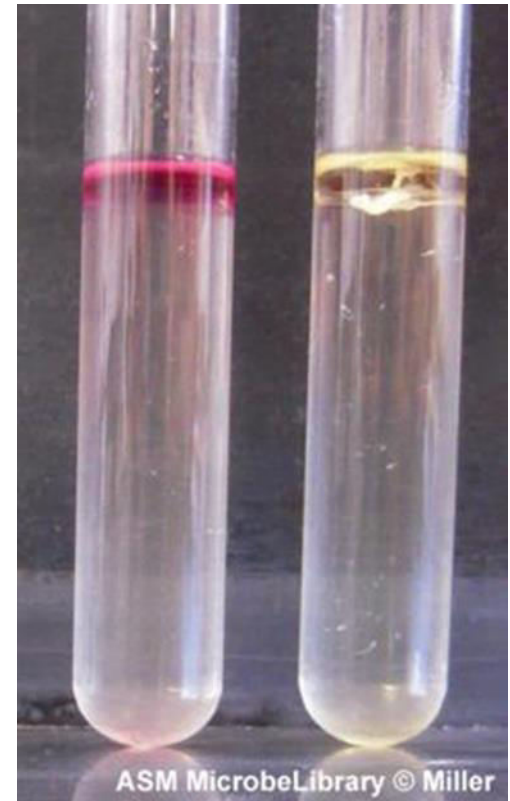
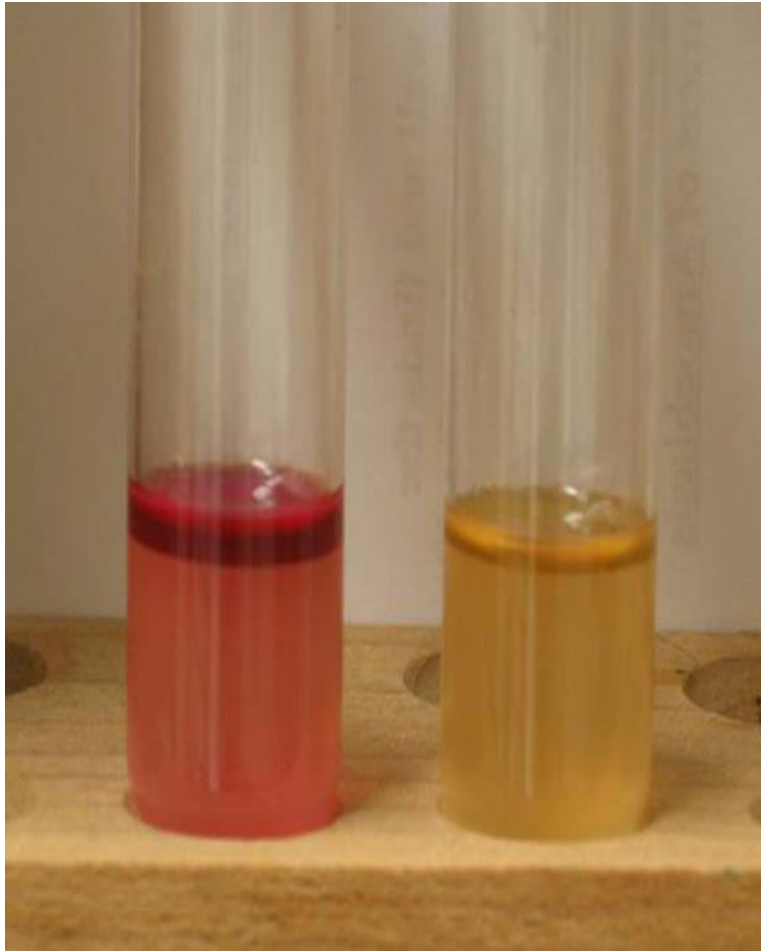
Procedure:

- An inoculum of bacteria is transferred to a sterile tube of tryptone broth using a loop or wire.
- Inoculated tube is incubated at 35-37 C for 24 hours.
- 5-10 drops of kovac's reagent are added to the tube

Result:

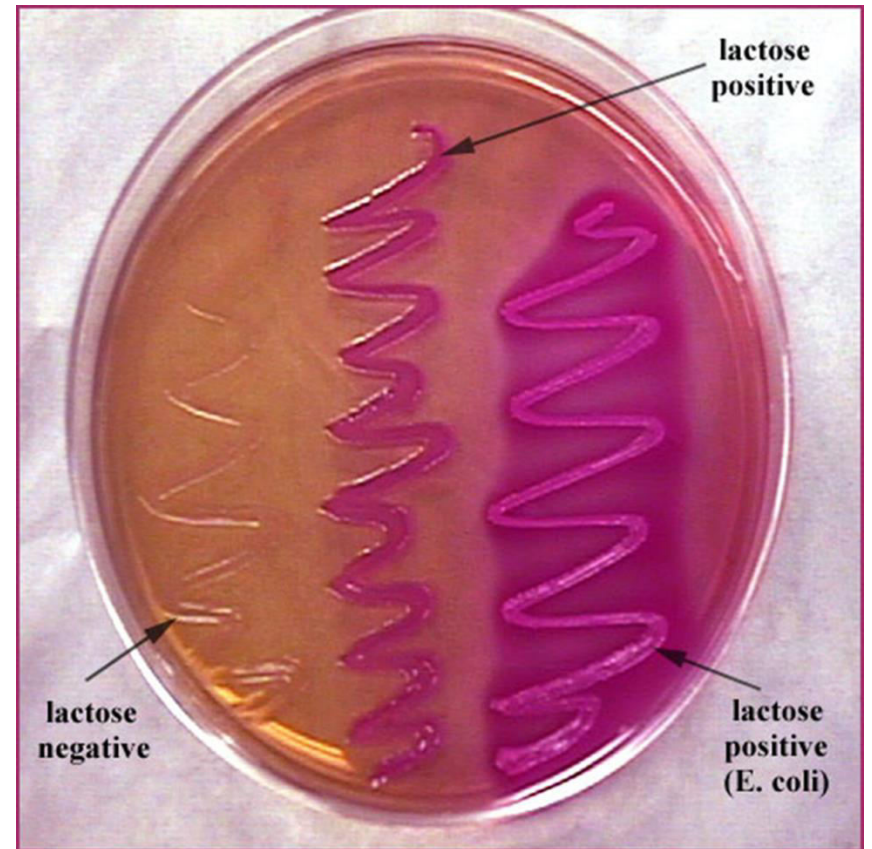
- Pos. result: red ring formation.
- Neg. result: no color change.

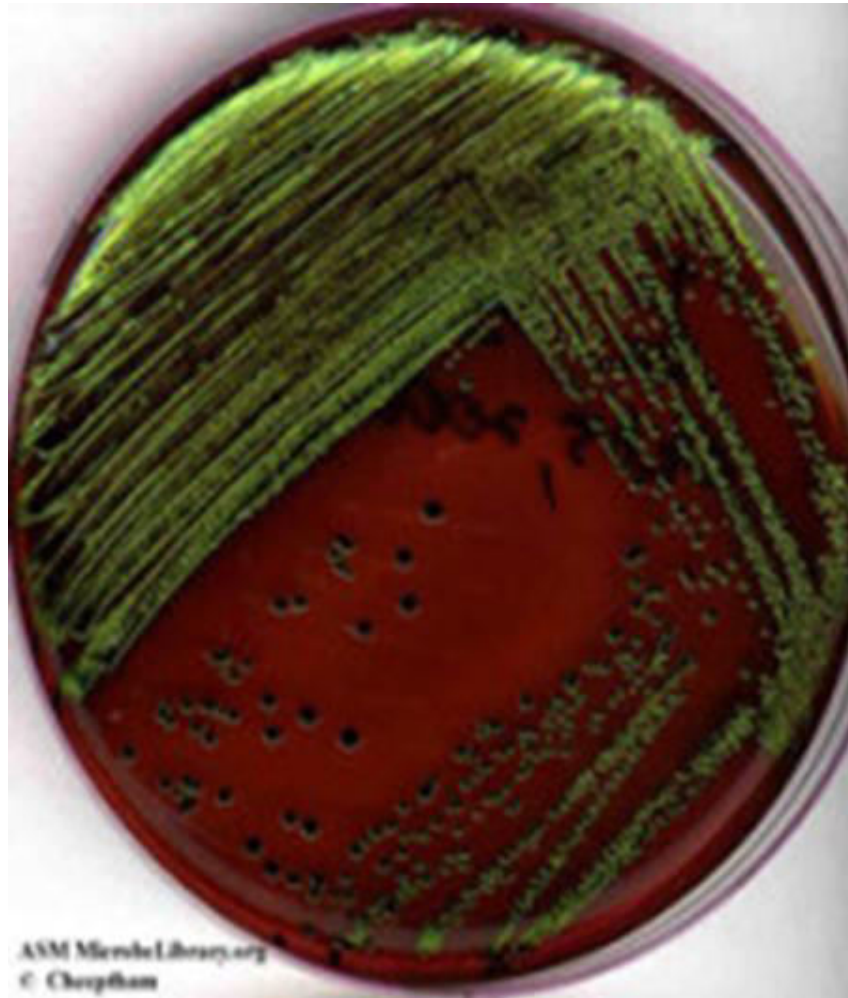
Indole Test



***E.coli* Cultural Characters**

- Facultative Anaerobic
- Grows between 10 – 40 °C, optimal at 37 °C.
- Can grown in simple medium.
- On MacConkey medium Produce Bright pink (Lactose fermenters)
- On EMB give a metallic green sheen





ASM MicroLibrary.org
© Chapman

Pseudomonas aeruginosa

- Motile (by single or multiple polar flagella)
- gram-negative rods
- Obligate (strict) aerobes
- Oxidase -- positive

CULTURAL CHARACTERS

- Growth on ordinary media (simple media) giving pigments (green colonies) and sweet odor (grape like odor).

Pseudomonas aeruginosa on nutrient agar(showing greenish exopigment)

