Coagulase Test

How to Perform Test: Inoculate rabbit plasma with one single colony. Break up colony and stir until blended in plasma. Incubate at 37 degrees C for 24 hours.

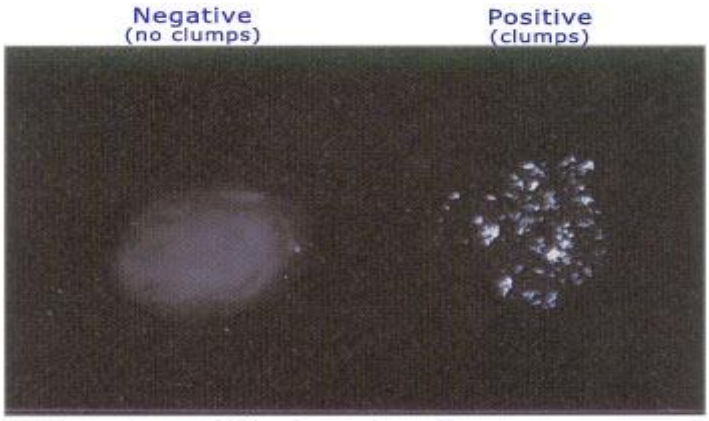
Property it tests for: This tests for the bacteria's ability to clot blood plasma using the enzyme coagulase. If the organism has coagulase it will clot rabbit plasma.

Media and Reagents: This media contains rabbit plasma dissolved in buffer.

Reading Results:

If the organism is has coagulase it will clot the plasma.

If the organism does not have coagulase it will not clot the plasma.



Slide Coagulase Test

Urea Hydrolysis

How to Perform Test: Inoculate Urea broth with inoculating loop.

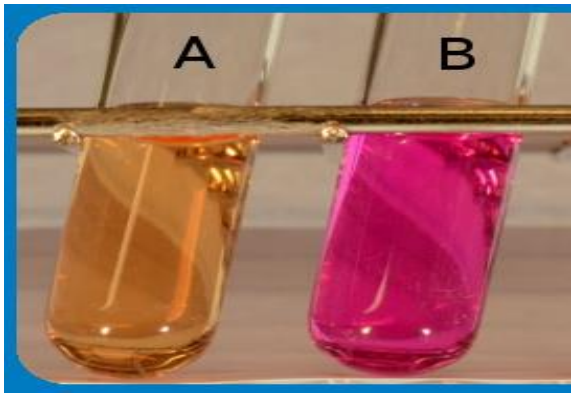
Property it tests for: This test is done to determine a bacteria's ability to hydrolyze urea to make ammonia using the enzyme urease.

Media and Reagents Used: Urea broth contains a yeast extract, monopotassium phosphate, disodium phosphate, urea, and phenol red indicator.

Reading Results:

Urea broth is a yellow-orange color. The enzyme urease will be used to hydrolyze urea to make ammonia. If ammonia is made, the broth turns a bright pink color, and is positive.

If test is negative, broth has no color change and no ammonia is made.



Triple Sugar Iron Agar Test

Major Content

- Glucose 0.1%(used first)
- Sucrose 1%(used second)
- Lactose 1%
- Proteins (used third)
- Phenol red pH indicator for acid production
- Ferric ammonium citrate H_2S indicator
- NaCl maintains osmotic pressure
- Agar solidifying agent

➲ **Purpose**

- used to determine carbohydrate fermentation and H_2S production in bacteria.
- Gas from carbohydrate metabolism can also be detected.

➲ **Principle**

- Carbohydrates pyruvate(acid) + CO_2
- Peptones NH_3 (makes medium alkaline)
- Phenol red red

Sodium thiosulfate in the medium is reduced by some bacteria to hydrogen sulfide (H_2S), a colorless gas. The hydrogen sulfide will react with ferric ions in the medium to produce iron sulfide, a black insoluble precipitate.

➲ **Theory**

- Fermentation of carbohydrates results in the production of acid which decreases the pH of the medium to change from reddish-orange to yellow.
- Utilization of peptones results alkalization of the medium due to the production of NH_3.
- The production of hydrogen sulfide is indicated by the presence of black ppt. formed by the reaction of H_2S with ferric ions.
- Slant is aerobic while butt is anaerobic.
- Gas production is indicated by the splitting of the agar of lifting of it to the top.

Glucose fermenter

- Tube reaction ---- alkaline over acid (K/A) ⟶ Red slant , yellow butt.
- With H_2S production (K/A, H_2S +ve) ⟶ Red slant, black butt.

Glucose, Lactose and/or Sucrose Fermenter

- Tube reaction --- acid over acid (A/A) ⟶ Yellow slant, yellow butt
- With H_2S production(A/A, H_2S +ve) ⟶ Yellow slant, black ppt.butt

Glucose, Lactose and Sucrose Non-fermenters

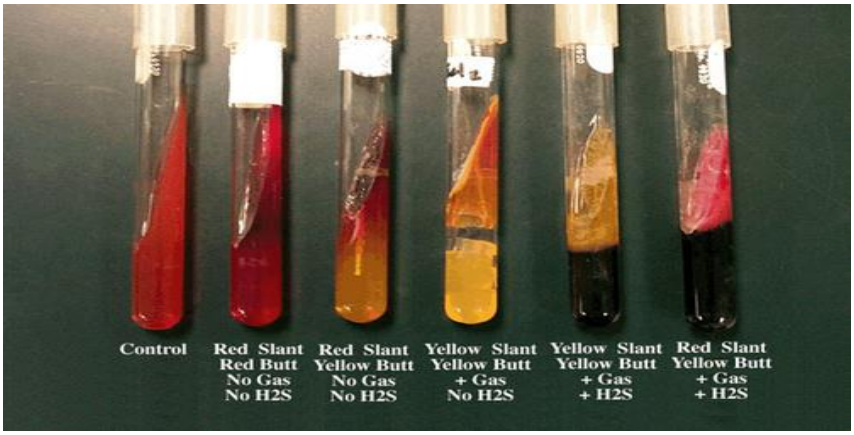
- Tube reaction:

i) alkaline over alkaline(K/K): If the bacteria can metabolize peptones both aerobically and anaerobically, both slant and butt red.

ii) alkaline over no change (K/NC): If peptones can only be metabolized aerobically slant red, butt no change.

With H_2S production

- alkaline over no change (K/NC, H_2S +ve)
- black precipitate (H_2S) in the butt.


Control
Red Slant
Red Butt
No Gas
No H2S
Red Slant
Yellow Butt
No Gas
No H2S
Yellow Slant
Yellow Butt
+ Gas
No H2S
Yellow Slant
Yellow Butt
+ Gas
+ H2S
Red Slant
Yellow Butt
+ Gas
+ H2S