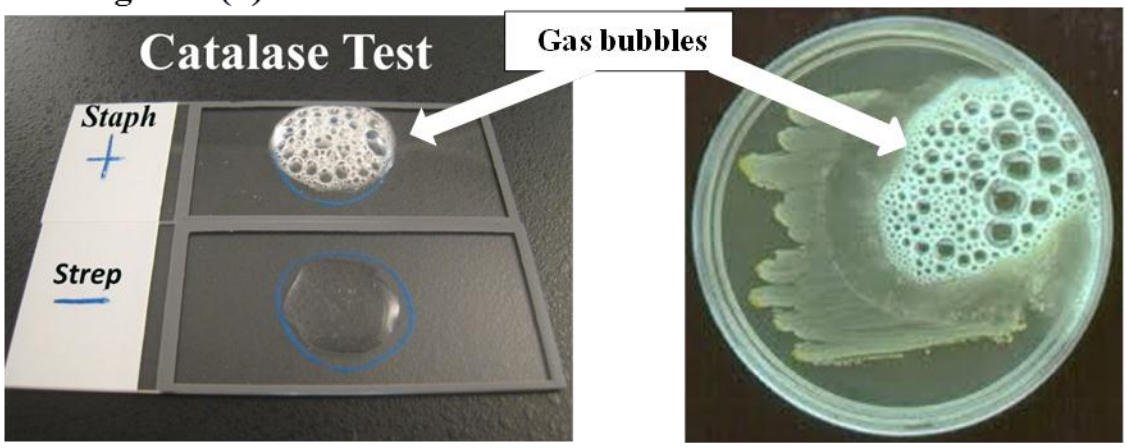
Rapid biochemical tests:

Catalase test: The enzyme catalase catalyzes the production of water and Oxygen from hydrogen peroxide (which a toxic agent for bacteria). All members of Staphylococcus genus are catalase positive, whereas all member of Streptococcus are catalase negative.

- **Catalase test: H_2O_2** —*Catalase enzyme*→ **$H_2O + O_2$** (*By bacteria*)

By mixing a drop of 3% hydrogen peroxide (H_2O_2) with a colony of the test bacteria on slide or on plate. Producing air bubbles = positive (+), without air bubbles = negative (+)



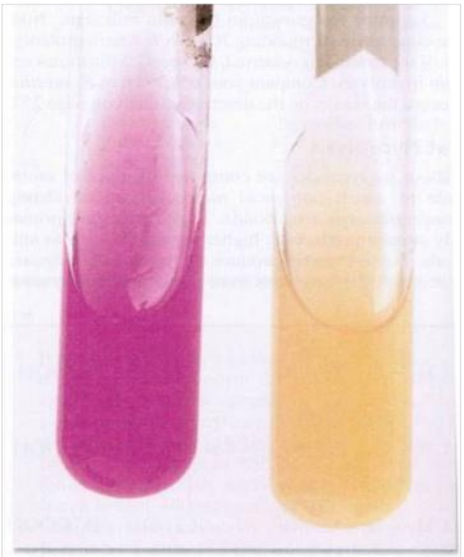
- **Coagulase**:

S. aureus produces an extracellular enzyme called coagulase which brings about clotting of **human or rabbit plasma**. **It acts along with a (coagulase reacting factor) (CRF) present in plasma**, binding to prothrombin and converting fibrinogen to fibrin. **Coagulase does not clot plasma of guinea pigs and some other species because they lack CRF**. CRF is similar to pro- thrombin but is probably not identical with it. ***Staph. aureus* strains usually secrete both coagulase and clumping factor.** Coagulase test is the standard criterion for the identification of *Staph. aureus* isolates. The role of coagulase in the pathogenesis of disease may cause the formation of a fibrin layer around a staphylococcal abscess, thus localizing the infection and protecting the organisms from phagocytosis.

[**Fibrinogen** —Coagulase→ **Fibrin (Clotting]**

- **Urease test:** Some bacteria, (such as *Proteus*, *Klebsiella*, *Citrobacter*, some *Haemophilus* species, the yeast Cryptococcus and fungi) are able to produce **Urease enzyme** which hydrolyzes Urea into ammonia, water and CO2. The alkaline end products cause the indicator phenol red to change from yellow to pink or red. This test can be used to differentiate Salmonella and Shigella which are urease negative from urease positive organisms.

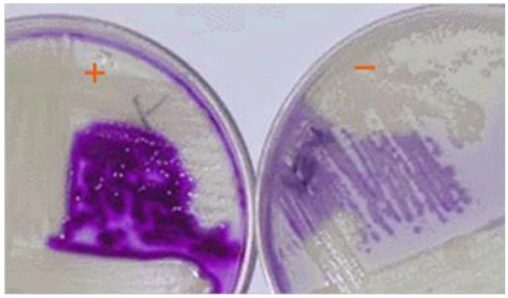
Positive



Negatuve

FIGURE 42.4 Urease test. Tube on the left is positive (*Proteus*); tube on the right is negative. © The McGraw-Hill Companies/Auburn University Photographic Service

- **Oxidase test:** Commonly used to identify ***Neisseria*** species**.** The test can be performed by flooding the bacterial colonies on surface of plate agar with **1% tetramethyl- p-phenylendiamine-dihydrochloride**, that results in deep purple color for positive result and colorless for negative result.

Purple color 

colorless

- **Spot indole test:** The organism that produces the enzyme tryptophanase is able to degrade the amino acid tryptophan in to pyruvic acid, ammonia and indole. Indole is detected by addition of the indicator **aldehyde** forming a color end product. This test can be used to differentiate ***E. coli*** and the **swarming *proteus*** species from one another.
- **Bile solubility test: *Streptococcus pneumonia*e possesses an active autolytic enzyme (Amidase) that lyse the organism's own cell wall during division. When bile salt (Sodium deoxycholate) is mixed with liquid culture of *S. pneumonia*e, it rapidly autolyzes (dissolve). Other alpha-hemolytic Streptococci do not possess such autolytic enzyme and will not dissolve in bile.**