Triple Sugar Iron (TSI) Agar Slant

Principle:
To determine the ability of an organism to attack a specific carbohydrate incorporated in a basal growth medium, with or without the production of gas, along with the determination of possible hydrogen sulfide (H_2S) production.

Purpose:
Hydrogen sulfide production and/or fermentation patterns are generally characteristic for specific bacterial groups, genera, or species, especially among the Enterobacteriaceae.

Test Procedure:
1. With an inoculating needle, stab the TSI slant about 2/3 of the way into the butt, withdraw the needle and streak the slant.
2. Incubate aerobically at 35-37°C.
3. Examine after 18-24 hours of incubation.

Interpretations:

Carbohydrate utilization…

	Fermentation of only glucose	**Fermentation of glucose & lactose**	**Neither glucose nor lactose fermented**
Slant	Alkaline reaction red color	Acid reaction yellow color	Growth only No color change
Butt	Acid reaction yellow color	Acid reaction yellow color	Growth only No color change

Gas Production…
- Aerogenic Gas production (CO_2) evident by one of the following…
  - A single gas bubble or bubbles in the medium
  - Splitting of the medium
  - Complete displacement of the medium from the bottom of the tube leaving a clear area
  - Slight indentation of the medium from the side of the tube
- Anaerogenic - No gas producion

H2S production:
The presence of black precipitate is evident by…
- A black color spread throughout the entire butt masking the acidity; may even be slight evidence on the slant
- A black ring near the top of the butt area
- A black precipitate scattered throughout the butt but not entirely