Triple Sugar Iron Agar Slant

Intended Use

Triple Sugar Iron Agar Slant is used for identification of Gram-negative enteric bacilli on the basis of dextrose, lactose and sucrose fermentation and hydrogen sulphide production.

Summary

Sulkin and Willett originally developed Triple Sugar Iron Agar, which was later modified by Hajna by adding sucrose to the double sugar (dextrose and lactose) formulation of Kligler Iron Agar. The addition of sucrose increased the sensitivity of the medium by facilitating the detection of sucrose fermenting bacilli as well as lactose and/or dextrose fermenters. Acid and gas production is an indication of carbohydrate fermentation, which gives a visible colour change from red to yellow due to change in the phenol red indicator. The production of hydrogen sulphide is indicated by the presence of a precipitate that blackens the medium in the butt of the tube. The medium complies with the recommendations of APHA for examination of food, dairy, water and wastewater and for microbial limit test in confirming the presence of *Salmonella* and in the identification of Gram-negative bacilli. Triple Sugar Iron Agar is also included in the Bacteriological Analytical Manual for food and cosmetics testing.

Principle

Tryptone, peptone, yeast extract and beef extract provide nitrogenous compounds, sulphur, trace elements, vitamin B complex, etc. while sodium chloride maintains the osmotic equilibrium. Lactose, sucrose and dextrose are the fermentable carbohydrates. Sodium thiosulphate and ferrous sulphate make the H₂S indicator system. Phenol red is the pH indicator.

| Formula* | | | | |
|---|-----------|--|--|--|
| Ingredients | g/L | | | |
| Lactose | 10.0 | | | |
| Sucrose | 10.0 | | | |
| Peptone | 10.0 | | | |
| Tryptone | 10.0 | | | |
| Sodium Chloride | 5.0 | | | |
| Yeast Extract | 3.0 | | | |
| Beef Extract | 3.0 | | | |
| Dextrose | 1.0 | | | |
| Sodium Thiosulphate | 0.3 | | | |
| Ferrous Sulphate | 0.2 | | | |
| Phenol Red | 0.024 | | | |
| Agar | 12.0 | | | |
| Final pH (at 25°C) | 7.4 ± 0.2 | | | |
| *Adjusted to suit performance parameters. | | | | |

Directions

- 1. Bring the Triple Sugar Iron Agar Slant to the room temperature 22°C-30°C.
- 2. Use Triple Sugar Iron Agar Slant as per required application.

Quality Control

Appearance: Brown-red coloured, smooth slant. **Cultural Response:** Cultural characteristics observed after an incubation of 18-24 hours at 30°C-35°C.

| Organism (ATCC) | Growth | Slant | Butt | Gas | H₂S |
|---------------------------------------|--------|-------|------|-----|-----|
| Salmonella enterica subsp. enterica | Good | Κ | А | + | + |
| serovar Typhimurium (14028) | | | | | |
| Escherichia coli (25922) | Good | А | А | + | - |
| Shigella flexneri serotype 2b (12022) | Good | K | А | - | - |
| Pseudomonas aeruginosa Strain | Good | K | Κ | - | - |
| Boston 41501 (27853) | | | | | |

Key:

A = Acidic (Colour of the media changed to yellow)

K = Alkaline (No change in colour of the media or the colour of the media changed to pink)

+ = Positive (For H_2S positive colour of the media changed to black and for gas positive bubbles / gap / disruption observed into the media)

- = No reaction (No change in colour of the media)

Storage and Stability

- 1. Store the ready to use Triple Sugar Iron Agar Slant at 15°C-25°C in a cool, dry place away from light.
- 2. Stability of the kit is as per expiry date mentioned on the label.

Precautions/Limitations

- It is important to stab the butt of the medium. Failure to stab the butt invalidates this test. Do not use an
 inoculating loop to inoculate a tube of Triple Sugar Iron Agar because while stabbing the butt, mechanical
 splitting of the medium occurs, causing a false positive result for gas production. Caps must be loosened
 during this test or erroneous results will occur.
- 2. Triple Sugar Iron Agar must be read within the 18-24 hour stated incubation period. A false positive reaction may be observed if read too early. A false-negative reaction may be observed if read later than 24 hours.
- Hydrogen sulphide production may be evident on Kligler Iron Agar but negative on Triple Sugar Iron Agar. Studies by Bulmash and Fulton showed that the utilization of sucrose could suppress the enzymatic mechanism responsible for H₂S production. Not all H₂S positive *Salmonellae* are positive on Triple Sugar Iron Agar.
- 4. Sucrose is added to Triple Sugar Iron Agar to eliminate some sucrose fermenting, lactose non-fermenters such as *Proteus* species.

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

References

- 1. Downes and Ito (ed.) 2001, Compendium Of Methods For The Microbiological Examination Of Foods, 4th edition, APHA Washington DC.
- 2. H. Wehr and J. Frank, 2004, Std. Methods for The Examination of Dairy Products, 17th Edition; APHA, Washington, DC.
- Greenberg AE; Clesceri LS and Eaton AD (Eds), 1998, Std Methods for The Examination of Water and Wastewater, 20th edition, APHA, Washington, DC.
- 4. IP, 1996, Ministry of Health and Family Welfare, Govt. of India, Vol. 2.
- 5. US Pharmacopeial Convention, Inc. 2001. The United States Pharmacopoeia 25/NF 20-2002. The US Pharmacopeial Convention, Inc; Rockville, Md.
- 6. US Food and Drug Adm; 1998, Bacteriological Analytical Manual, 8th Ed; Rev. A, AOAC, International, Gaithersburg, Md.
- 7. Data on file: Microxpress[®], A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

| Cat No. | Product description | Pack Size |
|--------------|----------------------|-----------|
| 203200440025 | Ready Prepared slant | 25 Slants |

Disclaimer

Information provided is based on our inhouse technical data on file, it is recommended that user should validate at his end for suitable use of the product.