

Triple Sugar Iron (TSI) Agar Slant**(M1670)**

Triple Sugar Iron (TSI) Agar is recommended for use in the differentiation of Enterobacteriaceae by their ability to ferment glucose, lactose, and sucrose, and their ability to produce hydrogen sulfide.

Composition

Ingredients	g/ L
Peptone	10.000
Tryptone	10.000
Yeast extract	3.000
Beef extract	3.000
Lactose	10.000
Sucrose	10.000
Dextrose	1.000
Sodium chloride	5.000
Ferrous sulphate	0.200
Sodium thiosulphate	0.300
Phenol red	0.024
Agar	12.000

Description

Triple Sugar Iron Agar was originally proposed by Sulkin and Willett and modified by Hajna for identifying Enterobacteriaceae. Tryptone, peptone, yeast extract and Beef extract provide nitrogenous compounds, sulphur, trace elements and vitamin B complex etc. Sodium chloride maintains osmotic equilibrium. Lactose, sucrose and dextrose are the fermentable carbohydrates. Sodium thiosulphate and ferrous ions make H₂S indicator system. Phenol red is the pH indicator. Organisms that ferment glucose produce a variety of acids, turning the colour of the medium from red to yellow.

More amount of acids are liberated in butt (fermentation) than in the slant (respiration). Growing bacteria also form alkaline products from the oxidative decarboxylation of peptone and these alkaline products neutralize the large amounts of acid present in the butt. Thus the appearance of an alkaline (red) slant and an acid (yellow) butt after incubation indicates that the organism is a glucose fermenter but is unable to ferment lactose and/or sucrose. Bacteria that ferment lactose or sucrose (or both), in addition to glucose, produce large amounts of acid enables no reversion of pH in that region and thus bacteria exhibit an acid slant and acid butt. Gas production (CO₂) is detected by the presence of cracks or bubbles in the medium, when the accumulated gas escapes. Thiosulphate is reduced to hydrogen sulphide by several species of bacteria and H₂S combines with ferric ions of ferric salts to produce the insoluble black precipitate of ferrous sulphide. Reduction of thiosulphate proceeds only in an acid environment and blackening usually occurs in the butt of the tube.

The reactions can be summarized as follows:

Alkaline slant / acid butt-only glucose fermented

Acid slant / acid butt-glucose and sucrose fermented or glucose and lactose fermented or all the three sugars, glucose, lactose and sucrose fermented.

Bubbles or cracks present-gas production

Black precipitate present-H₂S gas production

Type of specimen

Isolated microorganisms

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines. After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

In Vitro diagnostic use only. For professional use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture.

Limitations

1. Some members of the Enterobacteriaceae and H₂S producing Salmonella may not be H₂S positive on TSI Agar. Some bacteria may show H₂S production on Kligler Iron Agar but not on TSI Agar. This can happen because utilization of sucrose in TSI Agar suppresses the enzymic pathway that result in H₂S production.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality control**Physical/Chemical control**

Color : Red colour Slant pH: 7.20-7.60

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Microbiological activity

Organisms	Growth	Slant	Butt	Gas	H ₂ S
Escherichia coli ATCC 25922	Good growth	acidic reaction, yellowing of the medium	acidic reaction, yellowing of the medium	positive reaction	negative, no blackening of medium
Klebsiella pneumonia ATCC 700603	Good growth	acidic reaction, yellowing of the medium	acidic reaction, yellowing of the medium	positive reaction	negative, no blackening of medium
Proteus mirabilis ATCC 12453	Good growth	alkaline reaction, red colour of the medium	acidic reaction, yellowing of the medium	negative reaction	positive, blackening of medium
Salmonella Typhimurium ATCC 14028	Good growth	alkaline reaction, red colour of the medium	acidic reaction, yellowing of the positive, blackening of mediummedium	positive reaction	positive, blackening of medium

Storage and Shelf Life ,

Store between 2-8°C. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques.

Reference

1. Finegold S. M. and Baron E. J., 1986, Bailey and Scotts Diagnostic Microbiology, 7th Ed., The C.V. Mosby Co., St.Louis.
2. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
5. Quality Assurance for Commercially Prepared Microbiological Culture Media, M22. Clinical and Laboratory Standards Institute (CLSI - formerly NCCLS)