

## TM 1874 -TRIPLE SUGAR IRON AGAR (IS : 5887 (Part I, III and V) 1976, reaffirmed 2005)

### INTENDED USE

For confirmation of gram-negative enteric bacilli on basis of dextrose, lactose and sucrose fermentation and H<sub>2</sub>S production.

### PRODUCT SUMMARY AND EXPLANATION

Triple Sugar Iron Agar was originally proposed by Sulkin and Willett and modified by Hajna for identifying Enterobacteriaceae. This medium is a modification of the Kligler Agar where sucrose was added to differentiate *Proteus* and *Hafnia* (sucrose positive) from *Salmonella* and *Shigella* (sucrose negative). It complies with the specifications given by IS: 5887(Part I, III and V)

### COMPOSITION

Ingredients	Gms / Ltr
Agar	12.000
Lactose	10.000
Peptic digest of animal tissue	10.000
Casein enzymatic hydrolysate	10.000
Sucrose	10.000
Sodium chloride	5.000
Yeast extract	3.000
Beef extract	3.000
Dextrose	1.000
Ferrous sulphate	0.300
Sodium thiosulphate	0.300
Phenol red	0.024

### PRINCIPLE

The Medium is composed of Peptic digest of animal tissue, Beef extract and Yeast extract which provides nitrogen, carbon and vitamins required for bacterial growth. Triple Sugar Iron Agar consists of three carbohydrates; Dextrose, Lactose and Sucrose. When carbohydrates are fermented, acid production is detected by Phenol red pH indicator. Sodium thiosulphate is reduced to Hydrogen sulphide and Hydrogen sulphide reacts with iron salt yielding typical black iron sulphide. Ferrous sulphate is a Hydrogen sulphide (H<sub>2</sub>S) indicator and gives a typical black precipitate. Sodium chloride maintains osmotic balance of the medium. Agar is used as a solidifying agent.

### INSTRUCTION FOR USE

- Dissolve 64.62 grams in 1000ml distilled water.
- Gently heat to boiling with gentle swirling and dissolve the medium completely.
- Mix well and dispense into tubes as desired.
- Sterilize by autoclaving at 15 psi (121°C) for 15 minutes
- Allow the medium to set in sloped form with a butt about 1 inch long.

### QUALITY CONTROL SPECIFICATIONS

- Appearance of Dehydrated powder** : Light yellow to pink, homogeneous free flowing powder  
**Appearance of Prepared medium** : Pinkish red coloured, clear to slightly opalescent gel  
**pH (at 25°C)** : 7.4± 0.2

### INTERPRETATION

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

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Slant	Butt	Gas	H <sub>2</sub> S
<i>Citrobacter freundii</i>	8090	50-100	Luxuriant	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	Positive reaction	Positive reaction, blackening of medium
<i>Enterobacter aerogenes</i>	13048	50-100	Luxuriant	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	Positive reaction	Negative reaction, no blackening of medium
<i>Escherichia coli</i>	25922	50-100	Luxuriant	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	Positive reaction	Negative reaction, no blackening of medium
<i>Klebsiella pneumoniae</i>	13883	50-100	Luxuriant	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	Positive reaction	Negative reaction, no blackening of medium
<i>Proteus vulgaris</i>	13315	50-100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Negative reaction	Positive reaction, blackening of medium
<i>Salmonella Paratyphi A</i>	9150	50-100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Positive reaction	Negative reaction, no blackening of medium
<i>Salmonella Typhi</i>	6539	50-100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Negative reaction	Positive reaction, blackening of medium
<i>Salmonella Typhimurium</i>	14028	50-100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Positive reaction	Positive reaction, blackening of medium
<i>Shigella flexneri</i>	12022	50-100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Negative reaction	Negative reaction, no blackening of medium
<i>Vibrio cholerae</i>	15748	50-100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Negative reaction	Negative reaction, no blackening of medium

### PACKAGING:

In 100 & 500 gm packaging size.

### STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers below 25°C and protect from direct Sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the



first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

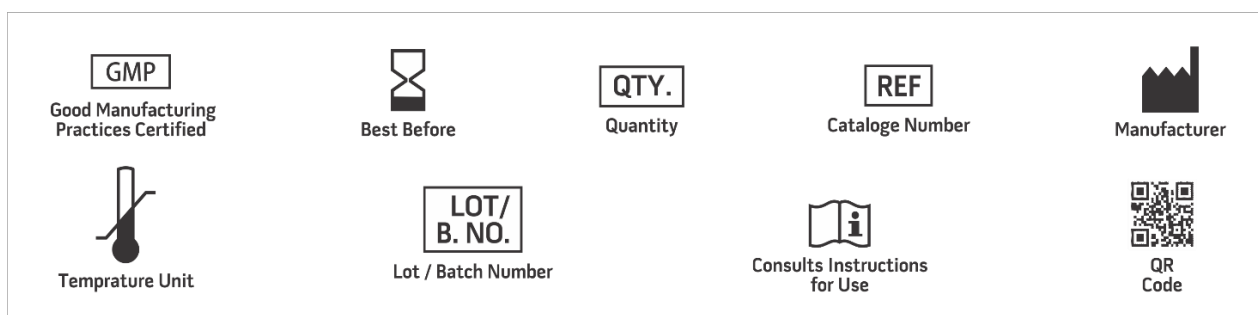
**Product Deterioration:** Do not use powder if they show evidence of microbial contamination, discoloration, drying, or other signs of deterioration.

### DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

### REFERENCES

1. Sulkin E.S. and Willett J.C., 1940, J. Lab. Clin. Med.,
2. Hajna A.A., 1945, J. Bacteriol, 49:516.
3. Vanderzant C. and Splittstoesser D., (Eds.), 1992, Compendium of Methods for the Microbiological Examination of Foods, 3rd ed. APHA, Washington D.C.
4. Marshall R. (Ed.), 1992, Standard Methods for the Examination of Dairy Products, 16th ed., APHA, Washington., D.C.
5. Finegold and Baron, 1986, Bailey and Scotts Diagnostic Microbiology, 7th ed., The C.V. Mosby Co., St. Louis.
6. Greenberg A. E., Trussell R. R. and Clesceri L. S. (Eds.), 1985, Standard Methods for the Examination of Water and Wastewater, 16th ed., APHA, Washington, D.C.
7. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.
8. International Organization for Standardization (ISO), 1993, Draft ISO/DIS 6579.
9. Bureau of Indian Standards IS : 5887 (Part I) 1976, reaffirmed 1986.
10. Bureau of Indian Standards IS : 5887 (Part V) 1976, reaffirmed 1996.
11. Bureau of Indian Standards IS : 5887 (Part III) 1999.



**NOTE:** Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

**\*For Lab Use Only**  
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