# CM 22159 -TRIPLE SUGAR IRON AGAR (IS : 5887 (Part I, III and V) 1976, reaffirmed 2005)

## INTENDED USE

Forconfirmationof gram-negative enteric bacilli on basis of dextrose, lactose and sucrose fermentation and $H_2S$ production.

## PRODUCT SUMMARYA NDEXPLANATION

TripleSugarIronAgarwas originallyproposed 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 composedofPepticdigest of animal tissue, Beef extract and Yeast extract which provides nitrogen, carbon and vitamins required forbacterialgrowth. Triple Sugar Iron Agar consists of three carbohydrates; Dextrose, Lactose and Sucrose. When carbohydratesarefermented, acid production is detected by Phenol red pH indicator. Sodium thiosulphate is reducedtoHydrogensulphide and Hydrogen sulphide reacts with iron salt yielding typical black iron sulphide. Ferrous sulphateisaHydrogen sulphide ($H_2S$) indicator and gives a typical black precipitate. Sodium chloride maintains osmotic balanceofthemedium. Agar is used as a solidifying agent.

## INSTRUCTION FOR USE

Dissolve 64.62 gramsin1000ml distilled water.
Gently heat to boilingwithgentle swirling and dissolve the medium completely.
Mix well and dispenseintotubes as desired.
Sterilize by autoclavingat15psi (121°C) for 15 minutes
Allow the mediumtosetinsloped 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

Culturalcharacteristics observed after anincubation at 35-37°C for 18-24 hours.

| Microorganism | ATCC | Inoculum (CFU/ml) | Growth | Slant | Butt | Gas | $H_2S$ |
|---|---|---|---|---|---|---|---|
| Citrobacter freundi | 8090 | 50-100 | Luxuriant | Acidic reaction, yellowing of the medium | Acidic reaction, yellowing of the medium | Positive reaction | Positive reaction, blackening of medium |
| Enterobacter aerogenes | 13048 | 50-100 | Luxuriant | Acidic reaction, yellowing of the medium | Acidic reaction, yellowing of the medium | Positive reaction | Negative reaction, no blackening of medium |
| Escherichia coli | 25922 | 50-100 | Luxuriant | Acidic reaction, yellowing of the medium | Acidic reaction, yellowing of the medium | Positive reaction | Negative reaction, no blackening of medium |
| Klebsiella pneumoniae | 13883 | 50-100 | Luxuriant | Acidic reaction, yellowing of the medium | Acidic reaction, yellowing of the medium | Positive reaction | Negative reaction, no blackening of medium |
| Proteus vulgaris | 13315 | 50-100 | Luxuriant | Alkaline reaction, red colour of the medium | Acidic reaction, yellowing of the medium | Negative reaction | Positive reaction, blackening of medium |
| Salmonella Paratyphi A | 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 |
| Salmonella Typhi | 6539 | 50-100 | Luxuriant | Alkaline reaction, red colour of the medium | Acidic reaction, yellowing of the medium | Negative reaction | Positive reaction, blackening of medium |
| Salmonella Typhimurium | 14028 | 50-100 | Luxuriant | Alkaline reaction, red colour of the medium | Acidic reaction, yellowing of the medium | Positive reaction | Positive reaction, blackening of medium |
| Shigella flexneri | 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 |
| Vibrio cholerae | 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:

In100&500gm packaging size.

## STORAGE

Dehydratedpowder, 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

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

## REFERENCES

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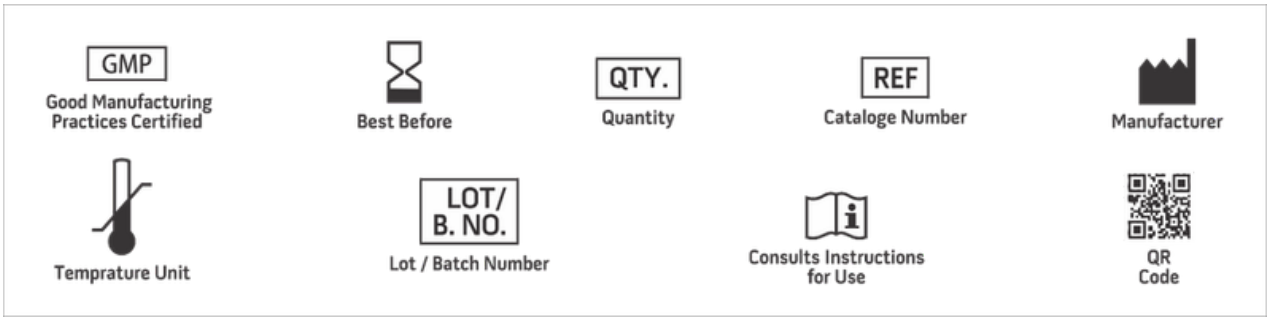
4. Marshall R. (Ed.), 1992, Standard Methods for the Examination of Dairy Products, 16th ed., APHA, Washington., D.C.
5. Finegold and Baron, 1986, Bailey and Scotts Diagnostic Microbiology, 7th ed., The C.V. Mosby Co., St. Louis.
6. Greenberg A. E., Trussell R. R. and Clesceri L. S. (Eds.), 1985, Standard 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.