



Triple Sugar Iron Agar

Medium for the differentiation of Enterobacteriaceae,
according to ISO 6579 and 6785.

DESCRIPTION

Triple Sugar Iron (TSI) Agar is a differential medium used for the identification of Enterobacteriaceae on the basis of four reactions, i.e. fermentation of lactose, glucose and sucrose and production of hydrogen sulphide.

TSI Agar is formulated according to ISO 6579 and 6785.

TYPICAL FORMULA	(g/l)
Meat Extract	3.0
Peptone	20.0
Yeast Extract	3.0
Sodium Chloride	5.0
Lactose	10.0
Sucrose	10.0
Glucose	1.0
Sodium Thiosulfate	0.3
Ferric Ammonium Citrate	0.3
Phenol Red	0.024
Agar	13.0
Final pH 7.4 ± 0.2 at 25°C	

METHOD PRINCIPLE

Meat extract and peptone provide amino acids, nitrogen, carbon, vitamins and minerals for organisms growth. Yeast extract is a source of vitamins, particularly of B-group. Sodium chloride maintains the osmotic balance of the medium. Lactose, sucrose and glucose are the fermentable carbohydrates. Sodium thiosulfate is reduced to hydrogen sulphide which in turn reacts with ferric ions yielding a black precipitate. Phenol red is the pH indicator. Agar is the solidifying agent.

PREPARATION

Dehydrated medium Suspend 65.6 g of the powder in 1 liter of distilled or deionized water. Mix well. Heat to boil shaking frequently until completely dissolved. Sterilize in autoclave at 121°C for 15 minutes.

Medium in bottles Melt the content of the bottle in a water bath at 100°C (loosing the cap partially removed) until completely dissolved. Then screw the cap and check the homogeneity of the dissolved medium, if it is the case turning the bottle upside down. Cool at 45-50°C, mix well avoiding foam formation and aseptically distribute into final containers.

TEST PROCEDURE

Touch an isolated colony with either a sterile needle or loop. Inoculate by streaking over the surface of the slope and stabbing in the butt of the tube. Incubate aerobically (with caps loosened) at 37°C for 24 hours. Do not incubate longer than 24 hours because the acid reaction in the slant of lactose and sucrose fermenters may revert to an alkaline reaction.

INTERPRETING RESULTS

Examine for carbohydrates fermentation, gas production and hydrogen sulfide production.

Carbohydrates fermentation results in an acidification of the medium that turns yellow:

- an alkaline slant-acid butt (red/yellow) indicates fermentation of glucose only;
- an acid slant-acid butt (yellow/yellow) indicates fermentation of glucose, lactose and/or sucrose;
- an alkaline slant-alkaline butt (red/red) indicates that the organism being tested is a non-fermenter.

Gas production from glucose is indicated by splitting and cracking of the medium.

A black precipitate in butt indicates hydrogen sulphide production.

APPEARANCE

Dehydrated medium: free-flowing, homogeneous, light beige.

Prepared medium: slightly opalescent, red.

STORAGE

The powder is very hygroscopic, store the powder at 10-30°C, in a dry environment, in its original container tightly closed. Store bottles, tubes and prepared plates at 10-25°C away from light. Do not use the product beyond its expiry date on the label or if product shows any evidence of contamination or any sign of deterioration.

SHELF LIFE

Dehydrated medium: 4 years.
 Medium in bottles: 2 years.
 Medium in tubes: 1 year.
 Ready-to-use plates: 6 months.

QUALITY CONTROL

Plates are inoculated with the microbial strains indicated in the QC table.
 Inoculum for productivity: ≤100 CFU.
 Incubation conditions: aerobically at 35 ± 2°C for 18-24 hours.

QC Table.

Microorganism		Growth	Slant	Butt	Gas	H ₂ S
<i>Escherichia coli</i>	ATCC® 25922	Good	Yellow	Yellow	+	-
<i>Proteus mirabilis</i>	ATCC® 12453	Good	Red	Yellow	-	+
<i>Pseudomonas aeruginosa</i>	ATCC® 27853	Good	Red	Red	-	-
<i>Salmonella typhimurium</i>	ATCC® 14028	Good	Red	Yellow	±	+
<i>Shigella flexneri</i>	ATCC® 12022	Good	Red	Yellow	-	-

WARNING AND PRECAUTIONS

The product does not contain hazardous substances in concentrations exceeding the limits set by current legislation and therefore is not classified as dangerous. It is nevertheless recommended to consult the safety data sheet for its correct use. The product is intended for professional use only and must be used by properly trained operators.

DISPOSAL OF WASTE









Disposal of waste must be carried out according to national and local regulations in force.

BIBLIOGRAPHY

- ISO 6579:2002. Microbiology of foods and animal feeding stuffs – Horizontal method for the detection of *Salmonella* spp.
- ISO 6785: 200. Milk and milk products – Detection of *Salmonella* spp.
- Sulkin S.E. and J.C. Willett (1940) A triple sugar-ferrous sulfate medium for use in identification of enteric organisms. J. Lab. Clin. Med. 25:649-653.

PRESENTATION		Contents	Ref.
Triple Sugar Iron Agar	90 mm ready-to-use plates	20 plates	11505
Triple Sugar Iron Agar	Slant tubes	10 x 10 ml tubes	30100
Triple Sugar Iron Agar	Slant tubes	10 x 15 ml tubes	30196
Triple Sugar Iron Agar	Bottles	6 x 200 ml bottles	413120
Triple Sugar Iron Agar	Dehydrated medium	500 g of powder	610350
Triple Sugar Iron Agar	Dehydrated medium	100 g of powder	620350

TABLE OF SYMBOLS

LOT Batch code	 Keep away from sunlight	 Manufacturer	 Use by	 Fragile, handle with care
REF Catalogue number	 Temperature limitation	 Contains sufficient for <n> tests	 Caution, consult Instruction For Use	 Do not reuse

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