Instructions For Use
ENGLISH

Tryptone Sulfite Neomycin Agar

Selective medium for detection of
anaerobic sulfite-reducing bacteria.

DESCRIPTION

Tryptone Sulfite Neomycin (TSN) Agar is a medium used for the selective isolation of sulfite reducing-clostridia, especially *Clostridium perfringens*, from food and other materials.

TYPICAL FORMULA	(g/l)
Tryptone	15.0
Yeast Extract	10.0
Sodium Sulfite	1.0
Ammonium Ferric Citrate	0.5
Neomycin Sulfate	0.05
Polymyxin B Sulfate	0.02
Agar	15.0
Final pH 7.2 ± 0.2 at 25°C	

METHOD PRINCIPLE

Tryptone (Pancreatic Digest of Casein) provides amino acids, nitrogen, carbon, vitamins and minerals for organisms growth. Yeast extract is a source of vitamins, particularly of B-group. Sodium sulfite and ferric citrate are H_2S indicators. Neomycin and Polymyxin suppress largely the accompanying bacterial flora. Agar is the solidifying agent.

PREPARATION

Dehydrated medium

Suspend 41.6 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 115°C for 20 minutes. DO NOT OVERHEAT.

The culture medium should be prepared and used the same day.

TEST PROCEDURE

Inoculate tubes or plates of Tryptone Sulfite Neomycin Agar by stabbing deep tubes or streaking plates with the test sample. Incubate for 18-24 hours at 46 ± 1°C in anaerobic atmosphere.

INTERPRETING RESULTS

While the growth of other sulfite-reducing clostridia is almost completely inhibited at 46°C, *C. perfringens* form black colonies. The plates should be inspected immediately after opening, otherwise the black colonies become paler in color due to oxidation of iron sulfide.

APPEARANCE

Dehydrated medium: free-flowing, homogeneous, beige.
Prepared medium: clear, yellowish-brown.

STORAGE

The powder is very hygroscopic, store the powder at 10-30°C, in a dry environment, in its original container tightly closed. Store tubes at 2-8°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 tubes: 1 year.

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QUALITY CONTROL

The medium is inoculated with the microbial strains indicated in the QC table.
Inoculum for productivity: 10-100 CFU.
Inoculum for selectivity: 10^4-10^6 CFU.
Incubation conditions: anaerobically at 46 ± 1°C for 18-24 hours.

QC Table.

Microorganism		Growth	Black Colonies
Clostridium perfringens	ATCC® 13124	Good	+
Escherichia coli	ATCC® 25922	Inhibited	---
Pseudomonas aeruginosa	ATCC® 27853	Inhibited	---

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

1. Marshall, R.S., Steenbergen, J.F., and Mcclung, L.S.(1965) Rapid technique for enumeration of Clostridium perfringens. Appl. Microbiol., 13:559-563.
2. Mossel, D.A.A.(1959) Enumeration of sulfite reducing clostridia occurring in foods. J. Sci. Food Agr., 10:662-669.

PRESENTATION		Contents	Ref.
Tryptone Sulfite Neomycin Agar	Tubes	20 x 10 ml tubes	31201
Tryptone Sulfite Neomycin Agar	Dehydrated medium	500 g of powder	610074
Tryptone Sulfite Neomycin Agar	Dehydrated medium	100 g of powder	620074

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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