

# TSN (Tryptone Sulfite Neomycin) Agar



Medium used for the isolation and identification of *Clostridium perfringens*.

## • CONTENTS (Liter)

Pancreatic Digest of Casein	15.0 g
Yeast Extract	10.0 g
Sodium Sulfite	1.0 g
Ferric Citrate	0.5 g
Neomycin Sulfate	0.05 g
Polymyxin B Sulfate	0.02 g
Agar	15.0 g
Final pH = 7.2 ± 0.2 at 25°C	

## • PROCEDURE

Suspend 41.57 G of powder in 1 L of distilled or deionized water. Heat to boiling until completely dissolved. Sterilize by autoclave at 118°C for 12 minutes. DO NOT OVERHEAT. Cool to 45 - 50°C in water bath. Mix well. Pour into petri dishes or tubes.

## • INTERPRETATION

TSN (Tryptone Sulfite Neomycin) Agar is a medium used for the isolation and identification of *Clostridium perfringens*. Pancreatic digest of casein and yeast extract are the nitrogen and vitamin sources. Sodium sulfite and ferric citrate act as indicators of sulfite. *Clostridium perfringens* reduces sulfite to sulfide. Sulfide reacts with iron in the medium and produces iron sulfide, which forms black precipitates. When the medium becomes oxidized, colonies can turn pale and fade. Neomycin sulfate and polymyxin B sulfate inhibit Gram-negative microorganisms and *Clostridium bifementans*. Agar is the solidifying agent.

## • TECHNIC

Inoculate the specimen using a sterile loop to the medium. If necessary, overlay with 10 - 15 mL of TSN Agar or inoculate using the pour plate method to improve the blackening of colonies. Incubate at 46 ± 1°C for 18 - 24 hours under anaerobic condition. High temperature incubation increases selectivity of the medium. Refer appropriate references for recommended test procedure.

## • QUALITY CONTROL FOR USE

### Dehydrated medium

Appearance: free-flowing, homogeneous

Color: light beige

### Prepared medium

Appearance : slightly opalescent

Color: light amber

Incubation conditions: 46 ± 1°C / 18 - 24 hours under anaerobic condition

Microorganism	ATCC	Inoculum CFU	Growth	Characteristics
<i>Clostridium perfringens</i>	13124	50-100	good	black colonies
<i>Escherichia coli</i>	25922	≥ 10 <sup>3</sup>	inhibited	-
<i>Pseudomonas aeruginosa</i>	27853	≥ 10 <sup>3</sup>	inhibited	-

**• STORE**

The powder is very hygroscopic. Store the powder at room temperature, in a dry environment, in its original container tightly closed and use it before the expiry date on the label. Store prepared medium at 2 - 8°C.

**• REFERENCES**

1. Mossel, (1959). J. Sci. Agr. 662-669.
2. Marshall, Steenbergen and McClung, (1965). Appl. Microbiol. 559-563

**• PACKAGE**

Cat. No : MB-T1074 TSN (Tryptone Sulfite Neomycin) Agar	500 G
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