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93735 TSN Agar (Tryptose Sulfite Neomycin Agar; Perfringens Selective Agar acc. to Marshall) NutriSelect® Plus

Highly selective medium for the detection and enumeration of *Clostridium perfringens* in food and other material.

Composition:

Ingredients	Grams/Litre
Casein peptone	15.0
Yeast extract	10.0
Sodium sulfite	1.0
Ferric citrate	0.5
Polymyxin B sulphate	0.02
Neomycin sulfate	0.05
Agar	13.5

Final pH 7.2 +/- 0.2 at 25°C

Store dehydrated powder and the prepared medium at 2-8°C. Use before expiry period on the label.

Appearance(color): Faintly yellow to Faint beige & Faint brown, free flowing powder
Gelling: Firm, comparable with 1.35% Agar gel
Color and Clarity: Medium amber coloured clear to slightly opalescent gel forms in Petri plates.

Directions:
Dissolve 40 g in 1 litre distilled water and autoclave gently at 121°C for 10 minutes. The medium should be used on the day of preparation.

Principle and Interpretation:
Clostridium perfringens food poisoning is one of the most common types of human foodborne illness (3). The foods usually involved are cooked meat or poultry containing large numbers of viable cells. A heat labile enterotoxin produced only by sporulation cells (4) induces the major symptoms of diarrhea in perfringens poisoning (5). TSN Agar is a slecetive media which was developed by Marshall et al. as a medium that could achieve rapid enumeration of *Clostridium perfringens* (1). The formulation is a modification of Mossel's medium for the enumeration of sulfite-reducing clostridia in foods (2).

Casien peptone provides nitrogen, vitamins, minerals, and amino acids essential for growth. Yeast extract is source of vitamins, particularly the B-group. The Neomycin and polymyxin are inhibitory for gram-negative enteric bacilli. Neomycin is also lethal for *C. bifermentans* . Ferric citrate and Sodium sulphite are H_2S indicators. *C. perfringens* reduces the sulphite to sulphide which reacts with the iron and forms a black iron sulphide precipitate, seen as black colonies. Bacteriological agar is the solidifying agent.The high incubation temperature of 46°C renders the medium specific for *C. perfringens* . The presumptive black colonies of *C. perfringens* should be confirmed biochemically. The selectivity of the medium results in the inhibition of some strains of *C. perfringens* (5).

Cultural characteristics under anaerobic conditions after 18-48 hours at 46°C.

Organisms (ATCC/WDCM)	Inoculum (CFU)	Growth	Recovery	Color of Colony
Escherichia coli (25922/ -)	≥10^3	-	0%	
Clostridium perfringens (12924/-)	50-100	++/+++	≥50%	black
Staphylococcus aureus (25923/-)	≥10^3	-	0%	

References:

1. Marshall, Steenbergen and McClung. 1965. Appl. Microbiol. 13:559.
2. Mossel. 1959. J. Sci. Food Agric. 10:662.
3. Doyle M. P., (Ed.), 1989, Foodborne Practical Pathogens, Marell Dekker, New York , N. Y.
4. Dunean C. L., 1973, A. J. Bacteriol., 113: 932
5. Vanderzant C. and Splittstoesser D. F., (Eds.), 1992, Compendium of Methods for the Microbiological Examination of Foods, 3rd Ed., APHA, Washington, D.C.

Precautions and Disclaimer
This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.