

TM 609 – TRYPTOSE SULPHITE NEOMYCIN AGAR

INTENDED USE

For selective isolation and enumeration of *Clostridium perfringens* in foods or other samples.

PRODUCT SUMMARY AND EXPLANATION

Clostridium perfringens food poisoning is one of the most common types of human foodborne illness. The foods usually involved are cooked meat or poultry containing large numbers of viable cells. A heat labile enterotoxin produced only by sporulation cells induces the major symptoms of diarrhea in perfringens poisoning. Tryptose Sulphite Neomycin Agar is a modification of Mossel Medium developed by Marshall et alfor the selective isolation and enumeration of C.perfringens from food. Thioglycollate addition is recommended if the cultured medium is to be incubated anaerobically.

COMPOSITION

Ingredients	Gms / Ltr		
Tryptose	15.000		
Yeast extract	10.000		
Sodium sulphite	1.000		
Ferric citrate	0.500		
Neomycin sulphate	0.050		
Polymyxin B sulphate	0.020		
Agar	13.500		

PRINCIPLE

Tryptose and yeast extract provide nitrogenous compounds, vitamin B complex and other growth nutrients. The antibiotics neomycin and polymyxin B sulphate inhibit gram-negative enteric bacilli. Neomycin is also lethal for C.bifermentans . The colonies of C.perfringens are black due to the ferric sulphide formed after the sulphite reduction. 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.

INSTRUCTION FOR USE

- Suspend 40.07 grams in 1000 ml distilled water.
- Heat to boiling to dissolve the medium completely. Dispense in screw capped containers.
- Sterilize by autoclaving with caps loose at 118°C for 12 minutes.
- Close the caps while the medium is still hot. 5 ml of sterile buffered thioglycollate solution may be added to every 200 ml of medium if desired.
- The buffered aqueous thioglycollate solution contains 35 ml buffer mixture (5.7% dipotassium phosphate and 28% sodium carbonate) and 15 ml sodium thioglycollate solution (13.3%).

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Cream to yellow homogeneous free flowing powder.

Appearance of prepared medium : Medium amber coloured clear to slightly opalescent gel forms in Petri plates.

pH (at 25°C) : 7.2±0.2

INTERPRETATION

Cultural characteristics observed after incubation.













Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Colour of the colony	Incubation Temperature	Incubation Period
Clostridium perfringens	12924	50-100	Good- luxuriant	>=50%	Black	46°C	18-48 Hours
Escherichia coli	25922	>=10³	Inhibited	0%	-	46°C	18-48 Hours
Staphylococcus aureus	25923	>=10³	Inhibited	0%	-	46°C	18-48 Hours

PACKAGING:

In pack size of 100 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°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.

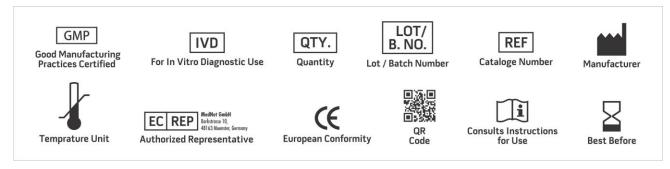
Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

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

REFERENCES

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



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. *For Lab Use Only

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