

TM 699 – COLIFORM PA BROTH

INTENDED USE

For determination of presence or absence of coliform bacteria in treated water.

PRODUCT SUMMARY AND EXPLANATION

Bacteriological Examination of water samples to determine its suitability for drinking and other domestic purpose has traditionally been done by the most probable number (MPN) procedures or the membrane filter (MF) technique. Although these methods have been useful, disadvantages such as space and time requirements prompted the idea of simply testing for the presence or absence of indicator bacteria. The presence of enteric pathogens in drinking and recreational waters is of great concern to public health. Therefore, interest in presence absence methods for determining the microbiological quality of drinking water has increased. The presence of total coliforms, faecal coliforms or *Escherichia coli* is well recognized as an indication of unsafe or poor water quality for which corrective measures should be taken. Coliform Broth is recommended for the isolation and cultivation of coliform organisms from cream, yogurt and raw milk. Coliform PA Broth is used for the determination of presence or absence of coliforms during detection of pollution in treated water from treatment plants or distribution systems. Weiss and Hunter proposed a simple procedure for the bacteriological examination of treated water that should be free of pollution. Later Presence-Absence (P-A) test was developed by Clark as the simplified version of the test, which is based on the principle that coliforms and other bacterial indicators of pollution should not be found in 100ml sample of treated water. However, the common connotation of "absence" can be misleading in the case of injured bacteria that are frequently present in treated drinking water systems and fail to produce a positive test on established media. P-A test has proved to be better than MF (Membrane filter) and FT (Multiple fermentation tube) methods. Coliform P-A broth is adaptable for screening of sample for the presence of alternative indicator organisms.

The P-A analysis of drinking water for total coliforms can entail 100 ml of sample added to 50 ml of triple strength Coliform P-A Broth (M1051) in 250 ml bottles. Bottles containing aliquots of the water sample to be tested are incubated and the results observed. A distinct yellow colour results from the fermentation of lactose and gas production can be detected as bubbles with gentle shaking.

COMPOSITION

Ingredients	Gms / Ltr
Tryptone	10.000
Gelatin peptone	5.000
Beef extract	3.000
Lactose	7.500
Dipotassium hydrogen phosphate	1.375
Potassium dihydrogen phosphate	1.375
Sodium chloride	2.500
Sodium lauryl sulphate (SLS)	0.050
Bromocresol purple	0.0085

PRINCIPLE

Tryptone, Gelatin peptone, and beef extract provides nitrogenous and carbonaceous compounds, vitamin B complex and trace ingredients. Lactose is the fermentable carbohydrate. Phosphates provide buffering capacity to the medium while sodium chloride maintains osmotic equilibrium. Bromocresol purple is the pH indicator. Coliforms that ferment lactose



produce acid and gas, which is indicated by a change in colour. Sodium lauryl sulphate is inhibitory to many organisms other than coliforms.

INSTRUCTION FOR USE

- Dissolve 92.42 grams in 1000 ml purified / distilled water to prepare triple strength medium.
- Dispense 50 ml amounts in 250 ml screw capped milk dilution bottles.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 12 minutes. Cool to 45-50°C.

QUALITY CONTROL SPECIFICATIONS

- Appearance of Powder** : Cream to yellow homogeneous free flowing powder.
Appearance of prepared medium : Purple coloured, clear solution without any precipitate.
pH (at 25°C) : 6.8±0.2

INTERPRETATION

Cultural characteristics observed after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Acid	Gas	Incubation Temperature	Incubation Period
<i>Klebsiella aerogenes</i>	13048	50-100	Good-luxuriant	Positive reaction, yellow colour	Positive reaction	35-37°C	18-48 Hours
<i>Escherichia coli</i>	25922	50-100	Good-luxuriant	Positive reaction, yellow colour	Positive reaction	35-37°C	18-48 Hours
<i>Staphylococcus aureus</i>	25923	≥10 ³	Inhibited	-	-	35-37°C	18-48 Hours
<i>Salmonella Typhimurium</i>	14028	50-100	Good-luxuriant	Negative reaction, no colour change	Negative reaction	35-37°C	18-48 Hours

PACKAGING:

In pack size of 500 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. Atlas R. M., 2004, Handbook of Microbiological Media, Lawrence C. Parks (Ed.), 3rd Edition, CRC Press.
2. Clark, 1969, Can. J. Microbiol., 5:771.



- Eaton A. D., Clesceri L. S., Rice E. W. and Greenberg A. E., (Eds.), 2005, Standard Methods for the Examination of Water and Wastewater, 21st Ed., APHA, Washington, D.C.
- Weiss and Hunter, 1939, J. Am. Water Works Assoc., 31:707.

 GMP Good Manufacturing Practices Certified	 Best Before	 Quantity	 Cataloge Number	 Manufacturer
 Temprature Unit	 Lot / Batch Number	 Consults Instructions for Use	 QR Code	

NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**
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