

TM 176 - M-ENTEROCOCCUS AGAR BASE

INTENDED USE

For isolation and enumeration of Enterococci in sewage, water and foods by membrane filter technique.

PRODUCT SUMMARY AND EXPLANATION

This medium was devised by Slanetz, Bent and Bartley for the enumeration of Enterococci by the membrane filter technique. Slanetz and Bartley modified it by the addition of Triphenyl Tetrazolium Chloride (TTC) and found that larger colonies and higher counts were obtained by placing membrane filters directly on the agar surface than on pads saturated with liquid medium. This medium is highly selective. Burkwell and Hartman used polysorbate 80 (0.5 ml/l) and sodium carbonate (2 ml of a 10% aqueous solution per litre) to increase sensitivity for direct plating of foods and increasing colony size. As per standard methods, M-Enterococcus Agar is used for the detection of faecal *Streptococcus* and *Enterococcus* groups using the membrane filtration technique.

COMPOSITION

Ingredients	Gms / Ltr
Tryptone	15.000
Soya peptone	5.000
Yeast extract	5.000
Dextrose (Glucose)	2.000
Dipotassium hydrogen phosphate	4.000
Sodium azide	0.400
2,3,5-Triphenyl tetrazolium chloride	0.100
Agar	10.000

PRINCIPLE

Tryptone and soya peptone, yeast extract, dextrose act as source of carbon, nitrogen and other essential growth nutrients. Sodium azide inhibits gram-negative organisms. TTC serves as a rapid indicator of bacterial growth. TTC is reduced to insoluble formazan inside the bacterial cells, which gives red colouration to colonies.

For filtration, choose a sample size so that 20-60 colonies will result. Transfer the filter aseptically to agar medium, avoiding air bubbles beneath the membrane. The medium can also be directly inoculated by streaking the specimen and incubating the plates at 35-37°C for 24-48 hours. Incubate the plates at 35°C for 48 hours. After incubation, count all light and dark red colonies as Enterococci.

INSTRUCTION FOR USE

- Dissolve 41.50 grams in 1000 ml purified / distilled water.
- Heat to boiling to dissolve the medium completely. Do not overheat or autoclave.
- Cool to 45-50°C.
- Add 0.5 ml polysorbate 80 and 2 ml of 10% aqueous solution of sodium carbonate, if desired.
- Dispense into sterile Petri plates.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Cream to yellow homogeneous free flowing powder
Appearance of prepared medium : Light pink colored 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	Color of the colony(on membrane filter)	Incubation Temperature	Incubation Period
<i>Escherichia coli</i>	25922	50-100	Inhibited	0%	-	35-37°C	24-48 Hours
<i>Enterococcus faecalis</i>	29212	50-100	Luxuriant	>=70%	Pink to dark red	35-37°C	24-48 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. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
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3. Eaton A. D., Clesceri L. S. and Greenberg A. W., (Eds.), 1998, Standard Methods for the Examination of Water and Wastewater, 20th Ed., APHA, Washington, D.C.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
5. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
6. Salfinger Y., and Tortorello M.L, 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
7. Slanetz, Bent and Bartley, 1955, Publ. Health. Rep., 70:67.
8. Slanetz and Bartley, 1957, J. Bact., 74:591.



GMP Good Manufacturing Practices Certified	IVD For In Vitro Diagnostic Use	QTY. Quantity	LOT/ B. NO. Lot / Batch Number	REF Cataloge Number	 Manufacturer
 Temperature Unit	EC REP MedNet GmbH Buckstrasse 10, 49163 Muenster, Germany Authorized Representative	 European Conformity	 QR Code	 Consults Instructions for Use	 Best Before

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

***For Lab Use Only**
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