

# TM 726 – ENTAMOEBA MEDIUM

#### **INTENDED USE**

For cultivation of Entamoeba histolytica.

#### PRODUCT SUMMARY AND EXPLANATION

Amoebiasis occurs throughout all areas of the world, the causative agent of which is Entamoeba histolytica, the only amoeba pathogenic for humans. In nature, the organism exists in the cyst form. It enters the body through contaminated food or water. The organism passes through the stomach as cyst and the trophozoite amoebas emerge in the intestine. E. histolytica destroys the tissue of the large intestine, causing lesions, deep ulcers and diarrhea.

Entamoeba Medium formulated by Cleveland and Sanders and Cleveland and Collier, is used for the cultivation of E.histolytica. This medium is highly specific for E.histolytica, which grows luxuriantly on this medium. Other intestinal amoebae do not grow readily on this medium. The technique of overlaying the medium with fresh sterile horse serumsaline mixture, as reported by Cleveland and Collier, was reported to be the best method of isolation of E.histolytica.

### **COMPOSITION**

Ingredients	Gms / Ltr	
Liver, infusion from	272.000	
Proteose peptone	5.500	
Sodium alpha-glycerophosphate	3.000	
Sodium chloride	2.700	
Agar	11.000	

### **PRINCIPLE**

The medium consists of Proteose peptone and Liver infusion from which provide amino acids and other nitrogenous substances that support growth of E.histolytica. Sodium chloride maintains the osmotic balance of the medium and sodium alpha-glycerophosphate acts as a phosphorous source.

## **INSTRUCTION FOR USE**

- Dissolve 33.0 grams in 1000 ml purified / distilled water.
- Heat to boiling to dissolve the medium completely.
- Distribute in tubes and sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes. Cool to 45-50°C.
- Allow tubes to solidify in a slanted position. Cover about half of the slant with fresh sterile horse serum-saline mixture (1:6) and add a 5 mm loopful of rice powder, which has been sterilized in an oven at 160°C for one hour.

### **QUALITY CONTROL SPECIFICATIONS**

**Appearance of Powder** : Light yellow to brownish yellow homogeneous free flowing powder. Appearance of prepared medium : Dark amber coloured, slightly opalescent gel forms in tubes as slants.

: 7.0 ± 0.2 pH (at 25°C)

## INTERPRETATION

Cultural characteristics observed under anaerobic condition after incubation.













Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Incubation Temperature	Incubation Period
Entamoeba histolytica	30190	50-100	Luxuriant	25-30°C	68-72 Hours
Entamoeba invadens	30016	50-100	Luxuriant	25-30°C	68-72 Hours
Entamoeba moskkovskii	30042	50-100	Luxuriant	25-30°C	68-72 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.

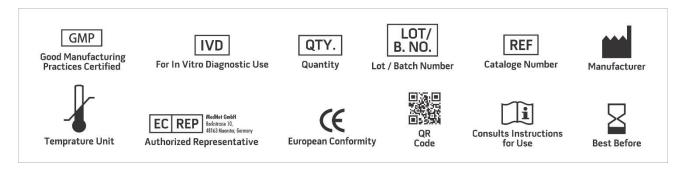
**Product Deterioration:** Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

## **DISPOSAL**

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

### **REFERENCES**

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- 3. Cleveland and Sanders, 1930, Arch. Protiskenkunde, 70:223.
- 4. Cleveland and Collier, 1930, Am. J. Hyg., 12:606.
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- 6. 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.
- 7. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
- 8. Sixth Annual Year Book 1935-36, P. 130, Suppl., Am. J. Pub. Health, 1936, 26, No. 3. 5.















**NOTE:** Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. \*For Lab Use Only **Revision: 08 Nov., 2019** 









