

Liver Broth**M928**

Liver Broth is used for the cultivation of anaerobic microorganisms.

Composition***

Ingredients	Gms / Litre
Liver, infusion from	23.000
Peptic digest of animal tissue	10.000
Liver tissues (extracted)	30.000
Dipotassium phosphate	1.000
Final pH (at 25°C)	6.8±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 64 grams in 1000 ml distilled water. Soak for 15 minutes with occasional stirring. Dispense in 18 mm diameter tubes to a depth of 50 mm so that bottom of the tube is filled with liver tissues. Sterilize by autoclaving at 115°C for 20 minutes. Cool, inoculate and seal with a layer of sterile 2% agar solution.

Principle And Interpretation

Clostridial species are one of the major causes of food poisoning / gastro-intestinal illnesses. They are gram-positive, spore-forming rods that occur naturally in soil (1). Among the family are : *Clostridium botulinum* which produces one of the most potent toxins in existence; *Clostridium tetani* , causative agent of tetanus; and *Clostridium perfringens* , commonly found in wound infections and diarrhea cases. The use of toxins to damage the host is a method deployed by many bacterial pathogens. The major virulence factor of *C.perfringens* is the CPE enterotoxin, which is secreted upon invasion of the host gut, and contributes to food poisoning and other gastrointestinal illnesses (1). Scarr recommended Liver Broth for the examination of Canners sugar for hydrogen swells caused by thermophilic anaerobes (2) and also for maintaining pure cultures of aerobes and anaerobes.

Anaerobic environment is created due to the reducing substances contained in liver tissues, which satisfy the requirements of even fastidious anaerobes.

This medium contains liver infusion from, which support luxuriant growth of saccharolytic, putrefactive, mesophilic and thermophilic anaerobes. A 20% w/v solution of the sugar steamed for 30 minutes to destroy the vegetative forms is inoculated into Liver Broth and sealed with agar. The standard proposed was a maximum of 1 positive tube in six tubes, with 20 ml inocula and incubated for 72 hours at 56°C. Some organisms like *Clostridium thermosaccharolyticum* produce gas, which often pushes the agar plug towards the top of the tube, and some organisms digest the solid liver tissues. The medium should be used on the same day of preparation as the stored medium may absorb air and then re-steaming is necessary which darkens the medium.

Quality Control**Appearance**

Brown coloured granules

Colour and Clarity of prepared medium

Medium amber coloured, clear to slightly opalescent supernatant over insoluble granules

Reaction

Reaction of 6.4% w/v aqueous suspension at 25°C. pH : 6.8±0.2

Cultural Response

M928: Cultural characteristics observed after an incubation at 55-57°C for 48-72 hours .

Organism	Inoculum (CFU)	Growth				
<i>Cl.thermosaccharolyticum</i> ATCC 7956	50-100	luxuriant				

Reference

- 1.Czeczulin J. R., Hanna P. C., Mcclane B. A., 1993, Infect. Immun., 61: 3429-3439.
- 2.Scarr M. P., 1958, DSIR, Proc. 2nd Internat. Symp. Food Microbiol., 1957, HMSO London, pp-29.

Storage and Shelf Life

Store below 30°C and prepared medium at 2-8°C. Use before expiry period on the label.