Cooked M-Medium

LQ080V

For cultivation of aerobes and anaerobes, especially pathogenic *Clostridia* and also for the maintenance of stock cultures.

Composition**

Ingredients	Gms / Litre
HMH Peptone B #	98.000
Proteose peptone	20.000
Dextrose (Glucose)	2.000
Sodium chloride	5.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

#- Equivalent to Beef heart, solids

Directions

Label the ready to use LQ080V bottle. Inoculate the sample and Incubate at specified temperature and time.

Principle And Interpretation

Clostridium is a large genus of gram-positive spore-bearing anaerobes. They are normally present in soil, some are responsible for human and animal diseases and others are associated with food spoilage. They may be saccharolytic, decomposing sugars to form butyric and acetic acids and alcohols. The meat in Robertsons Medium is reddened and gas is produced. Other proteolytic species attack the amino acids. Meat in Robertsons medium is blackened and decomposed by *Clostridium* species, giving the culture a foul odour. The mesophilic spore-forming anaerobes are of primary importance in the spoilage of low acid foods packed in sealed containers, because of their high heat resistance, their ability to grow in the absence of oxygen and a growth range which covers the temperature of normal storage of canned and other processed foods including the refrigerated storage of cured meats. Cooked M- Medium was originally developed by Robertson (3) for the cultivation of certain anaerobes isolated from wounds. The present formulation is a modification, also called as Chopped M Medium (2), which supports the growth of many spore forming and non-spore forming strict anaerobes. It has the ability to initiate growth of bacteria from very small inocula and to maintain the viability of cultures over long period. Mixed cultures of bacteria survive in Cooked M-Medium without displacing the slower-growing organisms. The products of growth do not rapidly destroy the inoculated organisms and therefore it is an excellent medium for the storage of aerobic and anaerobic organisms. It is used for cultivation and maintenance of Clostridia and for determining proteolytic activity of anaerobes (1,2).

FDA has recommended this medium for enumeration and identification of *Clostridium perfringens* from foods (4). Cooked M-Medium contains HMH peptone B, which provides nitrogeneous and carbonaceous compounds, long chain amino acids and other nutrients. The sulphydryl groups, which impart reducing effect, are more available in denatured protein and hence this medium is used. The addition of dextrose allows rapid and heavy growth of anaerobic bacteria in a short time and leads to more rapid identification of important anaerobes. Growth in this medium is indicated by turbidity or bubble formation by some organisms. Blackening and disintegration of the meat particles indicate proteolysis. Aerobes grow at the top whilst more anaerobic species grow deeper in the medium. For the isolation of *Clostridium* from food, use a stomacher to prepare 10% suspension of the food in Peptone Water (M028) diluent. Make dilutions and plate both suspensions and dilutions on Willis and Hobbs Medium Base (M1375) Tryptose Sulphite Cycloserine (T.C.S.) Agar Base (M837). Place a metronidazole disc on the inoculum. Incubate anaerobically at 37°C overnight. To count the clostridia, pour the plates with the dilutions on Perfringens Agar Base (O.P.S.P.) (M579). Incubate duplicate plates aerobically and anaerobically to distinguish between clostridia and other organisms. Add some of the suspension to two tubes of Cooked M-Medium. Heat one tube for 10 min at 80°C and incubate as above. Growth of clostridia is visualized as turbidity or gas bubbles. This medium can be further tested for presence of *Clostridium* (5).

Type of specimen

Clinical samples - faeces, wound exudate/ discharge, skin and tissue swab around wound, blood; Food and dairy samples; Water samples

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (4,5).
For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1,9).
For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (2).
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

In Vitro diagnostic Use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets

Limitations

1. Further biochemical tests must be carried out for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance
Sterile Clear Cooked M-Medium in bottles.
Colour
Medium amber coloured supernatant over insoluble granules.
Quantity of medium
5 ml of medium in bottles.
Reaction
7.00- 7.40
Sterility test
Passes release criteria
Cultural Response
Cultural characteristics observed after incubation at 35-37°C for 40-48 hours under anaerobic conditions.

Organism	Growth	Growth (under aerobic conditions)
Clostridium botulinum ATCC 25763	Luxuriant	-
Clostridium perfringens ATCC 12924	Luxuriant	-
Clostridium sporogenes ATCC 11437	Luxuriant	-
Enterococcus faecalis ATCC 29212 (00087)*	-	Luxuriant
Streptococcus pneumoniae ATCC 6303	-	Luxuriant

Key : (*) Corresponding WDCM numbers

Storage and Shelf Life

Store between 2-8°C. Use before expiry date on the label.
Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (4,5).

Reference

1. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
2. 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.
3. Collins C. H., Lyne P. M., Grange J. M., 1985, 7th Ed., Microbiological Methods.
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. MacFaddin J. F., 1985, Media for Isolation - Cultivation - Identification - Maintenance of Medical bacteria, Vol. I, Williams & Wilkins, Baltimore.
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. Robertson, 1916, J. Pathol. Bacteriol., 20:327.
9. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
10. U. S. Food and Drug Administration, 1984, Bacteriological Analytical Manual, 6th Ed., AOAC, Arlington, Va.

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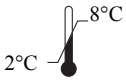
IVD — In vitro diagnostic medical device

CE Marking



Storage temperature

Do not use if package is damaged

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