Cooked Meat Medium

Intended Use

Recommended for cultivation of aerobes and anaerobes, especially pathogenic Clostridia from clinical, food and water samples. This can also be used as a maintenance medium for stock cultures.

Typical Composition (g/litre)

HMH peptone B 98.0 ; Proteose peptone 20.0 ; Dextrose 2.0 ; Sodium chloride 5.0

Mode of Action

Cooked M-Medium contains HMH peptone B, which provide amino acids and other nutrients. It also contains glutathione, a reducing substance that permits the growth of obligate anaerobes. The sulfhydryl groups, which impart reducing effect, are more available in denatured protein and hence cooked meat is added in the medium. 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.

Preparation

Suspend 12.5 grams in 100 ml purified / distilled water (or suspend 1.25 grams in 10 ml distilled water in test tubes). Mix thoroughly and allow to stand for 15 minutes until all the particles are thoroughly wetted.

Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Final pH (at 25°C) 7.2±0.2

Storage

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

Specimen

Clinical samples - Blood; Food and dairy samples; Water samples

Experimental Procedure and Evaluation

Growth in this medium is indicated by turbidity or bubble formation by some organisms. Blackening and disintegration of the meat particles indicate proteolysis. For best results, medium should be used on the day it is prepared, otherwise it should be boiled or steamed for a few minutes and allowed to cool without agitation and then inoculated. Inoculation should be made near the bottom of the tube in the meat particles for anaerobic cultures. 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 diluent. Make dilutions and plate, both suspensions and dilutions on Willis and Hobbs Medium Base,

(T.C.S.) Agar Base. 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 . Incubate duplicate plates aerobically and anaerobically to distinguish between clostridia and other organisms. Add some of the suspension to two tubes of Cooked Medium. Heat one tube for 10 min at 80°C and incubate as above. Growth of clostridia is visualized as turbidity or gas bubbles.

Quality Control

Organism	Inoculum	Growth
Clostridium botulinum ATCC 25763	50 - 100	Luxuriant
Clostridium perfringens ATCC 12924	50 - 100	Luxuriant
Clostridium perfringens ATCC 11437	50 - 100	Luxuriant
Streptococcus pneumonia ATCC 6303	50 - 100	Luxuriant
Enterococcus faecalis ATCC 29212	50 - 100	Luxuriant

Reference

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4. U. S. Food and Drug Administration, 1984, Bacteriological Analytical Manual, 6th Ed., AOAC, Arlington, Va
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