PRODUCT CODE: 413762

Eosin Methylene Blue Agar (EMB) (Dehydrated Culture Media) for microbiology

Preparation

Suspend 36 grams of medium in one litre of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 121ºC for 15 minutes. Cool to 45-50ºC, mix well, avoiding the formation of bubbles and dispense carefully into Petri Dishes.

DO NOT OVEARHEAT. The prepared medium should be stored at 8-15°C. The colour is tournasol blue. Sterilization reduces the methylene blue, leaving the medium orange in colour. The normal purple may be restored by gently mixing. The reduced medium should be shaken to oxidize the methylene blue; otherwise a dark zone from the top extending downwards will gradually appear.

The dehydrated medium should be homogeneous, free flowing and purple-rose flocculent precipitate in colour. If there are any physical changes, discard the medium.

Uses

EOSIN METHYLENE BLUE AGAR is a differential medium similar to Levine EMB Agar and is used for the isolation of *Enterobacteria*. The use of Eosin Y and Methylene Blue enable differentiation between lactose-fermenting and non-fermenting organisms. It is widely used in medical bacteriology, in techniques recommended by APHA and for the detection and enumeration of coliforms, contaminants of foods and drinking water.

Peptone provide nitrogen, vitamins, minerals and amino acids essential for growth. Sucrose is added to Lactose as a fermentable carbohydrate to detect coliforms that ferment sucrose more readily than lactose. Eosin Y and Methylene blue dyes are both partial inhibitors of Gram-positive bacteria and pH indicators. Due to the lactose and sucrose, the medium can be differential in primary culture: *Salmonellae* and *Shigellae* which are lactose-negative can be differentiated from other lactose-negative and sucrose-positive organisms such *as Proteus vulgaris, Citrobacter* and *Aeromonas*.

Dipotassium phosphate acts as a buffer system and Bacteriological agar is the solidifying agent. For the isolation of enteric pathogens from clinical samples, inoculate onto a small area of one quadrant of EMB Agar and streak for isolation, allowing discrete colonies to develop. Incubate at 35 ± 2°C and observe at 24 hours and again at 48 hours. Salmonella and Shigella colonies are translucent and amber coloured or colourless. Coliforms that use lactose and/or sucrose produce blue-black colonies with dark centres and a greenish metallic sheen.

Other coliforms such as Enterobacter form mucoid, pink colonies. Strains of *Enterococcus faecalis* are partially inhibited on this medium and appear as colourless colonies. As the medium is moderately inhibitory some *staphylococci*, *streptococci* and yeast may grow. Also, some Gram-negative non-fermenting bacilli may appear as non-lactose fermenters. Further Biochemical tests are necessary for genus identification.

Composition

See in Data Sheet (TDS).

Microbiological Test

The following results were obtained in the performance of the medium from type cultures after incubation at a temperature of 35 ± 2ºC and observed after 24-48 hours.

Microorganism	Growth	Colony Colour
Enterobacter aerogenes ATCC 13048	Good	Pink
Escherichia coli ATCC 25922	Good	Green with metallic shine
Salmonella typhimurium ATCC 14028	Good	Colourless
Pseudomonas aeruginosa ATCC 10145	Good	Colourless
Staphylococcus aureus ATCC 25923	Inhibited	Colourless

Storage

Once opened keep powdered medium closed to avoid hydration.

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