Technical Information

Anaerobic Egg Agar Base

Product Code: DM 1902

Application: - Anaerobic Egg Agar Base when supplemented with egg yolk emulsion is recommended for detection of *Clostridium perfringens* in foods.

Composition**

Ingredients	Gms / Litre
Proteose peptone	20.000
Casein enzymic hydrolysate	5.000
Yeast extract	5.000
Sodium chloride	5.000
Agar	20.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Besides Salmonella species and *Staphylococcus* aureus, *Clostridium perfringens* has been the third most common etiological agent *responsible for* food-borne disease [(1)]. *Clostridium* species are spore forming, gram-positive rods found naturally in soil [(2)]. C. perfringens food poisoning results from eating contaminated food. The major virulence factor of *C. perfringens* is the CPE enterotoxin, which is secreted upon invasion of bacteria in the host gut, and resulting in food poisoning and other gastrointestinal illnesses [(2)]. C. perfringens cells may lose viability if the suspected food samples are refrigerated, thereby making it difficult to incriminate the organisms in food poisoning outbreaks [(3)]. Anaerobic Egg Agar is one of the media recommended by APHA [(4)] for detecting *C. perfringens* in foods.

Casein enzymic hydrolysate and proteose peptone supply amino acids and other complex nitrogenous nutrients. Yeast extract provides essential B-complex vitamins. Egg yolk emulsion is added to the medium by which the lipase and lecithinase activity can be observed. Lecithinase of *C. perfringens* degrades lecithin of egg yolk, forming an insoluble opaque precipitate [(5)]. Lipase breaks down free fats present in the egg yolk causing irridescent sheen to form on the colony surface. For the lipase reaction, plates may be kept upto a week for incubation [(5)]. Proteolysis is indicated by clear zones in the medium surrounding the growth [(6)].

Methodology

Suspend 55 grams of powder media in 1000 ml distilled water. Shake well and heat to boil to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and aseptically add 80 ml sterile Egg Yolk Emulsion (MS 2045). Mix the contents thoroughly before pouring into sterile Petri plates.

Quality Control

Physical Appearance
Cream to yellow homogenesous free flowing powder
Gelling
Firm comparable with 2.0% Agar gel
Colour and Clarity of prepared medium
Basal medium -Light yellow coloured, clear to very slightly opalescent gel. After addition of Egg Yolk Emulsion -Light yellow coloured, opaque gel forms in Petri plates
Reaction
Reaction of 5.5% w/v aqueous solution at 25^0 C pH :7.2±0.2

pH Range:- 6.80-7.20

Cultural Response/Characteristics

DM 1902: Cultural characteristics observed with addded Egg Yolk Emulsion (FD045) when incubated anaerobically,at 35-37°C for 8-24 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Lecithinase	Lipase
Clostridium perfringens ATCC 12924	50-100	good-luxuriant	>=50%	positive	Negative reaction
Clostridium sporogenes ATCC 11437	50-100	good-luxuriant	>=50%	reaction, opaque zone around the colony	positive reaction, irridescent sheen on the colony

Storage and Shelf Life

Dried media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.
Prepared Media: 2-8^0 in sealable plastic bags for 2-5 days.

Further Reading

1. Centre for Disease Control, 1982, CDC Surveillance Summaries, 35:7SS-16SS, 1986.
2. Czeczulin J. R., Hanna P. C., Mcclane B., Infect. Immun., 61: 3429-3439 (1993).
3. Traci P. A., and Duncan C. L.,1974 , Appl. Microbiol., 28:815
4. Downes F. P. and Ito K., (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., APHA, Washington, D.C.
5. Finegold S. M. and Baron E. J., 1986, Bailey and Scotts Diagnostic Microbiology, 7th Ed., The C.V. Mosby Company, St. Louis.
6. Murray P. R., Baron E. J., Jorgensen J. H., Pfaller M. A., Yolken R. H., (Eds.), 8th Ed., 2003, Manual of Clinical Microbiology, ASM, Washington, D.C.

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