

# **Product Data Sheet**

# C. BOTULINUM ISOLATION AGAR BASE Product No. GB-DCM-00142-1A

#### INTENDED USE

For isolation of Clostridium botulinum from food and clinical samples.

# **Product Description**

Clostridium botulinum is an anaerobic, spore forming bacteria that produces a neurotoxin protein botulin. Severe food poisoning results from the consumption of this protein (toxin), which may be produced in foods contaminated with Clostridium botulinum. C. botulinum Isolation Agar Base is formulated as per the recommendation of APHA for the selective isolation of C. botulinum from food samples. The antibiotics supplement containing the broad spectrum antibiotics namely Cycloserine, sulphamethoxazole and trimethoprim makes the medium very selective. Egg yolk emulsion helps in detecting lecithinase, lipase and photolytic activity. Lecithinase degrades lecithin present in the egg yolk producing an insoluble, opaque precipitate in the medium surrounding the growth. Lipase break down free fats present in the egg volk causing an iridescent (oil on water) sheen to form on the surface of the colonies. Botulinal toxin is heat-labile. Therefore, the test samples and cultures should be maintained at refrigeration temperatures. The pH of the toxic material should also be maintained at a slightly acidic pH since botulinal toxin is less stable at alkaline ph. Inoculate 1-2 grams of solid or 1-2 ml of liquid food per 15 ml of enrichment broth. The enrichment broth employed is Cooked Meat Medium. After incubation at 35°C for 7 days, observe for turbidity, gas production and meat digestion. Carry out gram staining and spore staining. To isolate Botulinum mix enrichment broth with equal amount of sterile ethanol (alcohol treatment). The alcohol treated culture is further streaked on C.botulinum Isolation Agar Base. Alternatively untreated enrichment cultures or stool can be streaked directly on Botulinum Isolation Agar Base.

# **Product Specifications**

Ingredients	Gms / Ltr
Tryptone	40.000
Yeast extract	5.000
Dextrose (Glucose)	2.000
Disodium hydrogen phosphate	5.000
Sodium chloride	2.000
Magnesium sulphate	0.010
Agar	20.000

## **QUALITY CONTROL SPECIFICATIONS**

Appearance of Powder: A Light yellow to pink homogeneous free flowing powder. Appearance of prepared medium Red coloured, clear to slightly opalescent gel forms in Petri plates.

PH (at 25°C): 7.1±0.2

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## **PRINCIPLE**

Tryptone and yeast extract supply amino acids and other nitrogenous substances and vitamin B complex. Dextrose is the fermentable carbohydrate. Disodium phosphate helps in buffering the medium while magnesium sulphate helps for the sporulation of the organisms. Sodium chloride maintains the osmotic equilibrium of the medium.

Microorganism	ATCC	Inoculum (CFU)	Growth	Recovery	Lecithinase	Incubation Temperature	Incubation Period
Clostridium botulinum	25763	50-100	Good- luxuriant	450%	Positive reaction, opaque zone around the colony	35-37°C	48 Hours

#### **NSTRUCTION FOR USE**

- Dissolve 37.0 grams in 450 ml purified / distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.
- Cool to 45-50°C and aseptically add sterile 50 ml Egg Yolk Emulsion and reconstituted contents of 1 vial of CBI Supplement.

#### STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

**Product Deterioration:** Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

### **DISPOSAL**

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

This product is for research use only.