Halophilic Broth

M591

Intended Use:

Recommended for isolation and cultivation of extremely halophilic bacteria.

Composition**

Ingredients	Gms / Litre
Acicase™ #	10.000
Yeast extract	10.000
Proteose peptone	5.000
Trisodium citrate	3.000
Potassium chloride	2.000
Magnesium sulphate	25.000
Sodium chloride	250.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters
Equivalent to Casein acid hydrolysate

Directions

Suspend 30.5 grams in 100 ml distilled water. Heat if necessary to dissolve the medium completely. Dispense in tubes or flasks as desired and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle And Interpretation

Halophilic media are formulated for isolation and cultivation of extreme halophilic species of *Halobacterium* and *Halococcus* from foods (1, 2). For optimum growth they require high salt concentration of about 20-30%. In general, the requirement for salt by halophilic microorganisms is not an exclusive need for NaCl since many species require low levels of K +, Mg++ and other ions in addition to NaCl (5,6). The level of salt required by microorganism varies greatly. Therefore the microbial types associated with a particular salted food depend on the concentration and type of salt and food. The most recent classifications of halophilic microorganisms are based on the level of salt required (2, 5). These bacteria can cause pink discoloration on the outer surface accompanied by putrefaction and decomposition of fish, bacon and hides preserved in sea salts.

Halophilic Broth contains Acicase™; proteose peptone and yeast extract which provide all the necessary nutrients, mainly nitrogenous and vitamins to the halophilic bacteria. Trisodium citrate is added to avoid the losses (2). Magnesium sulphate, sodium chloride and potassium chloride are essential ions required for the growth of extreme halophiles.

Type of specimen

Water samples from high-salinity water body

Specimen Collection and Handling:

10 gm sample is added to 90 ml Halophilic Broth and incubated at 35°C for upto 12 days. The organisms are then isolated onto Halophilic Agar (M590) from this enriched culture.

Warning and Precautions :

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 specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. This medium is general purpose medium and may not support the growth of fastidious organisms.
2. Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.

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
Off-white to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium
Amber coloured, slightly hazy solution with heavy precipitate at the bottom in tubes.

Reaction
Reaction of 30.5% w/v solution at 25°C. pH : 7.2±0.2

pH
7.00-7.40

Cultural Response
Cultural characteristics observed after an incubation at 35-37°C for 12 days.

Organism	Growth
Halobacterium salinarium ATCC 33171	luxuriant
Halococcus morrhuae ATCC 17082	luxuriant

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. 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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Reference

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