Vibrio Parahaemolyticus Sucrose Agar

M1153

Vibrio Parahaemolyticus Sucrose Agar (VPSA) is used for isolation and enumeration of *Vibrio parahaemolyticus* from seafood in accordance with APHA.

Composition**

Ingredients	Gms / Litre
Tryptose	5.000
Casein enzymic hydrolysate	5.000
Yeast extract	7.000
Sucrose	10.000
Sodium chloride	30.000
Bile salts mixture	1.500
Bromo thymol blue	0.025
Agar	15.000
Final pH (at 25°C)	8.6±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 73.52 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Vibrio parahaemolyticus is a halophilic estuarine organism. This organism can be isolated from a variety of seafood products and marine environments. The organism, when isolated from fresh seafood, is usually found in low numbers (< 100/g) and is sensitive to refrigeration and heat.

Vibrio parahaemolyticus Sucrose Agar (VPSA) is recommended by APHA (1) for isolating and enumerating *V. parahaemolyticus* from seafoods. It is a differential medium (and also selective to some extent) that distinguishes *V.parahaemolyticus* from other marine Vibrios. This medium is employed in the final steps of Hydrophobic Grid Membrane Filtration enumeration procedure (HGMF) (2).

Tryptose, casein enzymic hydrolysate and yeast extract provide the necessary nitrogen compounds, growth factors and vitamin B complex for the growth of *V.parahaemolyticus* . Sucrose is the fermentable carbohydrate. Bromothymol blue is the pH indicator. Bile salts mixture inhibits the contaminating gram-positive bacteria. High salt content and alkaline pH of the medium provides conditions that facilitate easy recovery of *Vibrio* 's. *V. parahaemolyticus* does not ferment sucrose and forms green to blue colonies which differentiates it from other sucrose fermenting *Vibrio* species.

Suspected seafood sample when diluted and blended with sterile peptone tween salt diluent, is filtered through HGMF using sterile diluent as a carrier. HGMF is then aseptically transferred to the Tryptic Soya Salt Agar with Magnesium Sulphate (TSAMS) (M990) plates and incubated for 4 hours at 35°C. HGMF is then transferred from TSAMS to the dry VPSA (M1153) plate and incubated for 18-20 hours at 42°C.

Quality Control

Appearance

Light yellow to pale green homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel.

Colour and Clarity of prepared medium

Blue coloured clear to slightly opalescent gel forms in Petri plates.

Reaction

Reaction of 7.35% w/v aqueous solution at 25°C. pH : 8.6±0.2

pH
8.40-8.80

Cultural Response
M1153: Cultural characteristics observed after an incubation at 42°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
Cultural Response				
Staphylococcus aureus ATCC 25923	>=10^3	inhibited	0%	-
Vibrio parahaemolyticus ATCC 17802	50-100	luxuriant	>=50%	blue-green

Storage and Shelf Life
Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

Reference
1. Downes F. P. and Ito K., (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., APHA, Washington, D.C.
2. Entis P. and Boleszczuk P., 1983, J. Food Prot., 46:783.

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