

# **Technical Data**

Gelatin Agar M920

Gelatin Agar is recommended for cultivation and identification of Vibrio species.

## Composition\*\*

Ingredients	Gms / Litre
Gelatin	30.000
Casein enzymic hydrolysate	10.000
Sodium chloride	10.000
Agar	15.000
Final pH ( at 25°C)	7.2±0.2

<sup>\*\*</sup>Formula adjusted, standardized to suit performance parameters

## **Directions**

Suspend 65 grams in warn preheated 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Mix well and pour into sterile Petri plates.

# **Principle And Interpretation**

Members of the genus *Vibrio* are facultative anaerobes capable of both respiratory and fermentative metabolism. The natural habitat for *Vibrio* species is aquatic, in both fresh water and salt water. The growth and biochemical reactivity of most species are enhanced in different test media supplemented with 1-2% sodium chloride. *Vibrios* are fairly easy to isolate from both clinical and environmental material, though some species may require growth factors and /or vitamins. Media can be made selective for *Vibrio* 's by adding appropriate selective agents (1). High concentrations of NaCl and alkaline pH have also been used to select certain *Vibrio* species, based on the ability of most *Vibrio* 's to grow at pH values above 8.0 and at 3% or higher concentrations of NaCl. Gelatin Agar is formulated in accordance with APHA (2) for the cultivation and characterization of Vibrio species from foods and faeces. Clinical specimens must be obtained early in the disease as possible because the duration of excretion of the pathogen is short.

Weigh 25 grams of sample such as seafood or vegetables either blended or cut into small pieces and add into 2 flasks. Add 225 ml Alkaline Peptone Water (M618) to one flask and 225 ml of Glucose Phosphate Broth (M070) in another flask. Mix well. Incubate at  $35^{\circ} \pm 2^{\circ}$ C for 6 to 8 hours. Inoculate one loopful from each flask on the non-selective Gelatin Agar.

*V.cholerae* appear transparent and usually have a characteristic cloudy zone around colony, which becomes more definite after few minutes of refrigeration. When these colonies are viewed in oblique light they appear iridescent green to bronze coloured and finely granular.

## **Quality Control**

## **Appearance**

Cream to yellow homogeneous free flowing powder

#### Gelling

Firm, comparable with 1.5% Agar gel and 3.0% Gelatin gel

#### Colour and Clarity of prepared medium

Yellow coloured, clear to slightly opalescent gel forms in Petri plates

#### Reaction

Reaction of 6.5% w/v aqueous solution at 25°C. pH :  $7.2\pm0.2$ 

#### pН

7.00-7.40

### **Cultural Response**

Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours.

## **Cultural Response**

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Organism	Inoculum (CFU)	Growth	Recovery	Gelatin liquefaction
Cultural Response				
Vibrio cholerae ATCC 15748	50-100	luxuriant	>=50%	positive reaction, clear zone around the colony within 24-48 hours
Vibrio parahaemolyticus ATCC 17802	50-100	luxuriant	>=50%	positive reaction, clear zone around the colony within 24-48 hours

# **Storage and Shelf Life**

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

## Reference

1.Bruno Gomez-Gil and Ana Roque, Isolation, Enumeration and Preservation of the Vibrionaceae, F.L. Thompson, B. Austin and J. Swings. The Biology of Vibrios, ASM Press.

2.Downes F. P. and Ito K., (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., APHA, Washington, D.C.

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