

# **Technical Data**

# **Brilliant Green Agar with Phosphates**

Brilliant Green Agar with Phosphates is used for selective isolation and identification of Salmonellae from mixed flora while inhibiting *Escherichia coli*, *Proteus* and *Pseudomonas* species.

### **Composition\*\***

Ingredients	Gms / Litre			
Peptic digest of animal tissue	10.000			
Beef extract	5.000			
Yeast extract	3.000			
Lactose	10.000			
Sucrose	10.000			
Disodium phosphate	1.000			
Monosodium phosphate	0.600			
Phenol red	0.090			
Brilliant green	0.0047			
Agar	12.000			
Final pH ( at 25°C)	$6.9 \pm 0.2$			
**Formula adjusted, standardized to suit performance parameters				

## **Directions**

Suspend 25.84 grams in 500 ml distilled water. Heat with occasional agitation and bring just to the boil to dissolve the medium completely. DO NOT AUTOCLAVE. For more selectivity and maximum recovery aseptically add the rehydrated contents of 1 vial of Sulpha Supplement (FD068). Mix well before pouring into sterile Petri plates.

# **Principle And Interpretation**

*Salmonella* species cause many types of infections, from mild self-limiting gastroenteritis to life threatening typhoid fever. The most common form of *Salmonella* disease is self-limiting gastroenteritis with fever lasting less than 2 days and diarrhoea lasting less than 7 days (1).

Brilliant Green Agar Base w/phosphates is formulated as per the recommendation of Rijks Institute Voorde Volksgezondheld (National Institute for Public Health), Utrecht (2,3). It is also recommended by the ISO Committee (4,5,6), because of its improved performance with respect to recovery of smaller numbers of *Salmonella* species, inhibition of *Escherichia coli* 

, Proteus species and Pseudomonas species (7).

The medium contains peptic digest of animal tissue, beef extract and yeast extract as sources of carbon, nitrogen, vitamins, amino acids and essential nutrients. The two sugars namely lactose and sucrose serve as energy sources. Fermentation of lactose and / or sucrose in the medium results in the formation of acidic pH which is detected by phenol red indicator. Phosphates (M971) buffer the medium. Brilliant green helps to inhibit the contaminating microflora. The medium can further supplemented with sulphacetamide (1g/l) and sodium mandelate (0.25g/l) to inhibit contaminating microorganisms when the sample is suspected to contain large number of competing organisms along with *Salmonella* species (8).

Brilliant Green Agar w/Phosphates being highly selective is recommended to be used along with a less inhibitory medium to improve the chances of recovery. Often cultures are enriched in Selenite Cystine Broth (M025) or Tetrathionate Broth (M032). These enriched cultures are then isolated simultaneously on Brilliant Green Agar Base (M016/M971), SS Agar (M108), Bismuth Sulphite Agar (M027) and MacConkey Agar (M081).

## **Quality Control**

Appearance Light yellow to pink homogeneous free flowing powder Gelling

# **M971**

Firm, comparable with 1.2% Agar gel.

#### Colour and Clarity of prepared medium

Greenish brown coloured clear to slightly opalescent gel forms in Petri plates.

#### Reaction

Reaction of 5.2% w/v aqueous solution at 25°C. pH : 6.9±0.2

#### pН

6.70-7.10

#### **Cultural Response**

Cultural characteristics observed after an incubation at 35 -  $37^{\circ}$ C for 18 - 24 hours .

#### **Cultural Response**

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
Cultural Response				
Escherichia coli ATCC 25922	>=103	inhibited	0%	
Proteus vulgaris ATCC 13315	50-100	none-poor	<=10%	red
Pseudomonas aeruginosa ATCC 10145	50-100	none-poor	<=10%	red
Salmonella Enteritidis ATCO 13076	250-100	luxuriant	>=50%	bright red
Salmonella Typhimurium ATCC 14028	50-100	luxuriant	>=50%	bright red

#### **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

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Revision : 1 / 2011

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