

# **Technical Data**

## **Brilliant Green Agar Base w/ Phosphates**

**M971** 

#### **Intended Use:**

Recommended for selective isolation of Salmonellae while inhibiting Escherichia coli, Proteus and Pseudomonas species.

## Composition\*\*

Ingredients	g/L
Peptone	10.000
HM peptone B #	5.000
Yeast extract	3.000
Lactose	10.000
Sucrose	10.000
Disodium hydrogen phosphate	1.000
Sodium dihydrogen phosphate	0.600
Phenol red	0.090
Brilliant green	0.0047
Agar	12.000
Final pH ( at 25°C)	$6.9 \pm 0.2$

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

## **Directions**

Suspend 25.84 grams in 500 ml purified/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 one vial of S Selective Supplement (FD068). Cool to 45-50°C. Mix well and pour 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 peptone, HM peptone B 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).

## Type of specimen

Clinical: faeces; Food and dairy samples; Water samples

#### **Specimen Collection and Handling**

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (9,10,11).

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (13,14). For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (12).

After use, contaminated materials must be sterilized by autoclaving before discarding.

<sup>#</sup> Equivalent to Beef extract

HiMedia Laboratories Technical Data

## **Warning and Precautions**

In Vitro diagnostic use. For professional use only. 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 clinical specimens. Safety guidelines may be referred in individual safety data sheets.

## Limitations

- 1. Though this medium is selective for Salmonella other species of Enterobacteriaceae may grow.
- 2. Further confirmation has to be carried out on presumptive Salmonella isolates.

#### **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

Light yellow to pink homogeneous free flowing powder.

#### Gelling

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°C for 18 - 24 hours .

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
Escherichia coli ATCC 25922 (00013*)	>=104	inhibited	0%	
Proteus vulgaris ATCC 13315	50-100	none-poor	<=10%	red
Pseudomonas aeruginosa ATCC 10145 (00024*)	50-100	none-poor	<=10%	red
Salmonella Enteritidis ATCC 13076 (00030*)	50-100	luxuriant	>=50%	bright red
Salmonella Typhimurium ATCC 14028 (00031*)	50-100	luxuriant	>=50%	bright red

Key: (\*) Corresponding WDCM numbers.

## Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (13,14).

#### Reference

- 1. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
- 2. Edel W. and Kampelmacher E. H., 1969, Bull. W.H.O., 41:297.
- 3. Edel W. and Kampelmacher E. H., 1969, Bull. W.H.O., 39:487.
- 4. Anon, 1975, International Organization for Standardization, Meat and Meat products Ref. Method, ISO: 3565.
- 5. Anon, 1981, International Organization for Standardization, Microbiology Ref. Methods, ISO: 6579.
- 6. Anon, 1985, International Organization for Standardization, Milk and Milk Products; Ref. Method, ISO: 6785.

**HiMedia Laboratories Technical Data** 

- 7. R. B. and Reyes A. L., 1968, Appl. Microbiol., 16:746.
- 8. Watson U. C. and Walker A. P., 1978, J. Appl. Bacteriol. 45:195.
- 9. Salfinger Y., and Tortorello M.L., 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
- 10. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
- 11. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
- 12. Lipps WC, Braun-Howland EB, Baxter TE, eds. Standard methods for the Examination of Water and Wastewater, 24th ed. Washington DC:APHA Press; 2023.
- 13. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
- Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Revision: 06/2024



HiMedia Laboratories Pvt. Limited, Plot No.C-40, Road No.21Y, MIDC, Wagle Industrial Area, Thane (W) -400604, MS, India



CEpartner4U, Esdoornlaan 13, 3951DB Maarn, NL www.cepartner4u.eu



In vitro diagnostic medical device

**CE Marking** 



Storage temperature



Do not use if package is damaged

## Disclaimer:

User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related HiMedia™ publications. The information contained in this publication is based on our research and development work and is to the best of our knowledge true and accurate. HiMedia<sup>TM</sup> Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are not intended for human or animal or therapeutic use but for laboratory, diagnostic, research or further manufacturing use only, unless otherwise specified. Statements contained herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.