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Brilliant Green Agar

REF **PO0171A**

Intended Use
Brilliant Green Agar (Modified) (PO0171A) device is a selective and diagnostic agar for Salmonellae other than *Salmonella typhi* or *Salmonella paratyphi A* for clinical samples such as faecal samples, and food/environmental samples. Brilliant Green Agar (Modified) (PO0171A) device is used in diagnostic workflow to aid clinicians in determining potential treatment options for patients suspected of having Salmonellae infections.
The device is for professional use only, is not automated, nor is it a companion diagnostic.

Summary and Explanation
Salmonella species are rod shaped motile bacteria with peritrichous flagella with 2 to 5µm length and 0.7 to 1.5µm diameter of Enterobacteriaceae family[1]. Salmonella species is broadly classified into two major groups that are *Salmonella enterica* and *Salmonella bongori* and the former subdivided into typhoidal and paratyphoid serovar. Almost 70% of the Salmonella infections in the UK are caused by the non-typhoidal serovar *Salmonella enteritidis*[1]. *Salmonella enterica* often causes gastroenteritis in humans and the mode of transmission can be food like undercooked meat, egg, dairy products, contaminated water, infected people and animals[2].

The implications of Salmonella infections are reported more in immunocompromised patients. In patients with HIV, bacteremia due to non-Salmonella infections is common. Studies report that in immunocompromised HIV patients the Salmonella bacteremia can also spread to other tissues and organs especially lungs[3]. Consequently, it is important to distinguish Salmonella species in the clinical samples as it is the third most common cause of bacterial gastroenteritis in the UK[1].

Principle of Method
Brilliant Green Agar (Modified) (PO5033A) contains peptones, 'Lab Lemco' and yeast extract to support the growth of salmonellae. Lactose and sucrose are energy sources, the fermentation of which causes a drop in pH which results in the indicator dye, phenol red, changing from red to yellow at acid pH. Brilliant green is a selective dye that inhibits Gram-positive bacteria and many Gram-negative bacilli for the selective isolation of salmonellae, with the exception of *Salmonella typhi*. Colony morphology may vary depending on the strain and the length of incubation. Agar is the solidifying agent.

Typical Formula

	grams per litre
'Lab-Lemco'	5.0
Peptone	10.0
Yeast extract	3.0
Disodium hydrogen phosphate	1.0
Sodium dihydrogen phosphate	0.6
Lactose	10.0
Sucrose	10.0
Phenol red	0.09
Brilliant green	0.0047
Agar	12.0

Physical Appearance

Colour	Green/Brown
Clarity	Clear
Fill weight	19.0 ± 2.0g
pH	6.9 ± 0.2

Materials Provided
PO0171A: 10 x 90mm Brilliant Green agar plates

Each plate should only be used once.

Materials Required but Not Supplied
- Inoculating loops
- Swabs
- Collection containers
- Incubators
- Quality control organisms

Storage
- Store product in its original packaging at 2–10°C until used.
- The product may be used until the expiry date stated on the label.
- Store away from light.
- Allow product to equilibrate to room temperature before use.
- Do not incubate prior to use.

Warnings and Precautions
- For in vitro diagnostic use only.
- For professional use only.
- Inspect the product packaging before first use.
- Do not use the product if there is any visible damage to the packaging or plates.
- Do not use the product beyond the stated expiry date.
- Do not use the device if signs of contamination are present.
- Do not use the device if the colour has changed or there are other signs of deterioration.
- It is the responsibility of each laboratory to manage waste produced according to their nature and degree of hazard and to have them treated or disposed of in accordance with any federal, state and local applicable regulations. Directions should be read and followed carefully. This includes the disposal of used or unused reagents as well as any other contaminated disposable material following procedures for infectious or potentially infectious products.

Refer to the Safety Data Sheet (SDS) for safe handling and disposal of the product (www.thermofisher.com).

Serious Incidents
Any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the relevant regulatory authority in which the user and/or the patient is established.

Specimen Collection, Handling and Storage
Specimens should be collected and handled following local recommended guidelines, such as the UK Standards for Microbiology Investigations (UK SMI) ID 01, ID 24, S 7 and Q5.

Procedure

- Allow product to equilibrate to room temperature.
- Inoculate and streak the specimen onto the medium using a standard loop.
- Incubate plates aerobically for 18–24 hours at 37 ± 2°C.
- Visually inspect plates to assess colony growth and colour under good lighting.

Interpretation

Rose shiny colonies can indicate the presence of either *Salmonella* Typhimurium or *Salmonella* Enteritidis

Quality Control

It is the responsibility of the user to perform Quality Control testing taking into account the intended use of the medium, and in accordance with any local applicable regulations (frequency, number of strains, incubation temperature etc.).

The performance of this medium can be verified by testing the following reference strains.

Incubation Conditions:18 – 48 h @ 36 ± 1°C aerobic

Positive Controls Inoculum level: 10-100 cfu Colony count is ≥ 50% of the control medium count.	
Salmonella typhimurium ATCC® 14028™	Red colonies and medium
Salmonella nottingham ATCC® 14028™	Red colonies and medium
Negative Controls Inoculum level: 100-1000 cfu	
Escherichia coli ATCC® 25922™	≥ One log reduction when compared to non-selective control medium. Yellow/green colonies or no growth.
Inoculum level: 10^4-10^5 cfu	
Proteus hauseri ATCC® 13315™	No growth or pinpoint green colonies, no swarming

Limitations

Organisms with atypical enzyme patterns may give anomalous reactions on Brilliant Green Agar.

It should be noted that lactose fermenting salmonellae will not produce typical red colonies. Some salmonellae such as *Salmonella typhi* and *Salmonella paratyphi* are inhibited by the selective agents in the medium and will not grow. Not all non-target organisms may be completely inhibited and *Proteus citrobacter* and *Pseudomonas* spp., when able to grow mimic enteric pathogens producing red colonies. Identifications are presumptive and should be confirmed by appropriate serological/biochemical methods.

The medium should be protected from light at all times except during inoculation and after incubation.

Performance Characteristics

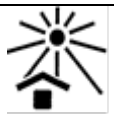
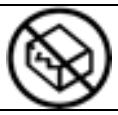
Accuracy has been demonstrated through review of the QC data. Correct detection of Salmonella strains is confirmed by the inclusion of a well-characterised isolate in the QC processes performed as part of the manufacture of each batch of the devices, which must meet the defined acceptance criteria. The precision of Brilliant Green Agar (Modified) (PO0171A) was demonstrated by an overall pass rate of 100% obtained for the product over three months of testing (February 2022 – May 2022; 10 batches). This shows that the performance is reproducible.

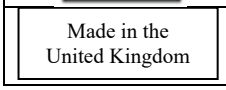
Brilliant Green Agar (Modified) (PO0171A) is tested in-house as part of QC process since the product was launched in 2001. For target organisms, when using 10-100 cfu inoculum of *Salmonella typhimurium* ATCC® 14028™ and *Salmonella nottingham* NCTC 7832 and incubating the device at 37 ± 2°C for 18-24 hours the user can recover organisms with colony size and morphology as listed in this document. For non-target organisms, when using 10^3 cfu of *Escherichia coli* ATCC® 25922 or 10^5 of *Proteus hauseri* and incubating the device at 37 ± 2°C for 18-24 hours the user can recover organisms with colony size and morphology as listed in this document.

Bibliography

1. Public Health England. 2021. 'Standards for Microbiology Investigations Identification of Salmonella species.' Issue no: 4 | Issue date: 12.03.21. https://www.gov.uk/government/publications/smi-id-24-identification-of-salmonella-species
2. Ford, L., Glass, K., Veitch, M., Wardell, R., Polkinghorne, B., Dobbins, T., ... & Kirk, M. D. (2016). Increasing incidence of Salmonella in Australia, 2000-2013. PLoS one, 11(10), e0163989.
3. Ridha, A. G., Malbrain, M. L. N. G., Mareels, J., Verbraeken, H., & Zachee, P. (1996). Lung abscess due to nontyphoid salmonella in an immunocompromised host: case report with review of the literature. Acta Clinica Belgica, 51(3), 175-183.

Symbol Legend

Symbol	Definition
	Catalogue number
	In Vitro Diagnostic Medical Device
	Batch code
	Temperature limit
	Use-by date
	Keep away from sunlight
	Do not re-use
	Consult instructions for use or consult electronic instructions for use
	Contains sufficient for <n> tests
	Do not use if packaging damaged and consult instructions for use

	Manufacturer
	Authorized representative in the European Community/ European Union
	European Conformity Assessment
	UK Conformity Assessment
	Unique device identifier
	Made in the United Kingdom

Oxoid Limited, Wade Road, Basingstoke, Hampshire, RG24 8PW, UK

For technical assistance please contact your local distributor.

Revision Information

Version	Modifications Introduced
1.0	2022-07-27 Original Document