Instructions for Use

REF - BM0270 **Plate Count Agar**

Bottled Media

1. Intended Use

BM0270 Plate Count Agar is intended for use in food, dairy, and water microbiology to perform Total Viable Counts.

2. Composition*

Ingredient	g/L
Tryptone	5.0
Yeast extract	2.5
Glucose	1.0
Agar	15.0

*Adjusted/supplemented as needed to meet performance requirements

3. Summary and Explanation

Plate Count Agar is formulated to the American Public Health Association [1, 2] specification developed by Buchbinder *et al.* [3]. It complies with the standards set out in ISO 11133 [4] and ISO 4833-1 and 2 [5 6]. The agar is of high gel strength and is therefore suitable for use in pour plate techniques as well as surface inoculation.

4. Principle

Tryptone and yeast extract supply the required nitrogen and other essential nutrients. Glucose is a fermentable carbohydrate. Agar is the solidifying agent.

5. Physical Characteristics

	Appearance and Colour	pH
Medium	Straw firm gel	7.0 ± 0.2

6. Materials Provided

BM0270 Plate Count Agar is available in the formats detailed below. Each bottle is ink-jet printed or labelled with (abbreviated) product name, product code, lot number and expiry date.

Product Code	Product Format
BM0270-A300-250	10 x 250ml in 300ml Sirop Bottles
BM0270-A500-400	6 x 400ml in 500ml Sirop Bottles
BM0270-M002-20	50 x 20ml in Thin Wall Universals

7. Materials Needed but not Provided

Standard microbiological laboratory materials e.g., sterile loops or swabs, collection containers, incubators, media for isolation and quality control organisms.

8. Specimens

BM0270 Plate Count Agar is suitable for the testing of the following specimens:

- Food and Environmental: products for human consumption, animal feed and environmental samples.
- Water samples from various sources.

Sampling and transport equipment must be used in accordance with the end user's suppliers' recommendations. Refer to appropriate standard method or local guidance on sample collection and subsequent processing.

9. Test Procedures and Interpretation of results

Heat Agar in a boiling water bath until it has entirely melted. Mix and allow to cool to approximately 45°C.

For the pour-plate method, add 1.0ml of the sample to the empty plate then pour the agar into the plate and mix by gentle rotation [5]. Alternatively, pour the agar into the plate, allow to set then inoculate the surface of the agar with 0.1ml of the sample and spread evenly over the surface of the agar [6].

Food and Environmental Specimens:

For solid samples, prepare a homogenised suspension using a ratio of 1:10 of sample to Maximum Recovery Diluent (E&O BM0760). With homogenised and liquid specimens, the medium should be inoculated with the specimen (as above) and incubated at 30 ± 1°C aerobically for 72 ± 3 hours.

Water Specimens:

A suitable volume of the sample is passed through a filter membrane of 0.45µm pore and the membrane is then placed on the surface of the medium, ensuring there are no air bubbles underneath, before incubating aerobically at 30 ± 1°C aerobically for 72 ± 3 hours.

After incubation, examine agar for colonies (typical colony appearance outlined in Quality Control table below). Perform further biochemical, serological, molecular, or mass spectroscopy testing to confirm identity of presumptive positive isolates. Refer to relevant local guidelines.

10. Quality Control

Organism	Incubation	Result (Specificity)
E. coli (NCTC 12241)	30 ± 1°C aerobically for 24-48 hours	Growth: Cream colonies
S. aureus (NCTC 12981)	30 ± 1°C aerobically for 24-48 hours	Growth: White colonies
B. subtilis (NCTC 10400)	30 ± 1°C aerobically for 24-48 hours	Growth: Cream colonies

It is the responsibility of the user to perform Quality Control testing taking into consideration the intended use of the medium and in agreement with any local relevant guidelines (e.g., frequency, strains used, atmosphere, incubation temperature).

11. Performance

To fully verify BM0270 Plate Count Agar performance, samples were tested to assess colony morphology and recovery level (where an acceptable range is ≥70% and ≤120%) compared to a non-selective reference medium. Prepared samples were inoculated with 30-150cfu and incubated at 30 ± 1°C aerobically for 72 ± 3 hours. All samples showed good recovery and the correct morphology of the required test organisms: *Escherichia coli* (NCTC 12241), *Staphylococcus aureus* (NCTC 12981) and *Bacillus subtilis* (NCTC 10400). Therefore, it can be concluded that BM0270 Plate Count Agar, meets performance criteria when used according to the instructions outlined above.

12. Limitations of the Medium

- Biochemical, serological, molecular, or mass spectrometry testing using pure cultures may be necessary for complete identification.
- Due to natural variation, some strains may grow poorly on this medium.

13. Precautions and Warnings

This product is considered non-hazardous under CLP regulations. Wear such PPE as recommended by laboratory COSHH assessment. During and after use, always handle all materials in a manner conforming to Good Laboratory Practices and consider that material under test should be regarded as a potential biohazard if mishandled.

Refer to E&O BM0270 Bottled Media Material Safety Data Sheet.

14. Storage conditions and Shelf life

Store BM0270 in the original bottle with the lid tightly closed at between 15 and 30°C. Kept under these conditions it may be used up to date of expiry shown on product label.

Dispose of in accordance with local and national authority requirements. Do not use medium if it is contaminated, discoloured or out of date. Additionally, do not use medium if it has been stored inappropriately or the packaging has been damaged.

15. References

Wehr, H., and Frank, J. (2004) Standard Methods for the Examination of Dairy Products. 17th ed. Washington DC: American Public Health Association.

2. Bridgewater, L. (2012) Standard Methods for the Examination of Water. 22nd ed. Washington DC: American Public Health Association, American Water Works Association, Water Environment Federation.

3. Buchbinder, L., Baris, Y., Alff, E., Reynolds, E., Dillon, E., Pessin, V., Pincus, L. and Strauss, A. (1951) Studies to formulate new media for the standard plate count of dairy products. Public Health Reports, 66, pp.327-340.

4. International Organization for Standardization (2014) 11133:2014 Microbiology of food, animal feed and water — Preparation, production, storage and performance testing of culture media. Geneva: ISO.

5. International Organization for Standardization (2013) 4833-1:2013 Microbiology of the food chain - Horizontal method for the enumeration of microorganisms - Part 1: Colony count at 30 °C by the pour plate technique. Geneva: ISO.

6. International Organization for Standardization (2013) 4833-2:2013 Microbiology of the food chain - Horizontal method for the enumeration of microorganisms - Part 2: Colony count at 30 °C by the surface plating technique. Geneva: ISO.

7. International Organization for Standardization (2006) 16266:2006 Water quality - Detection and enumeration of Pseudomonas aeruginosa - Method by membrane filtration. Geneva: ISO.

Version History*

001 18/10/23 - New Document Created

*Note: minor typographical, grammatical, and formatting changes are not included in the revision history.

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TABLE OF APPLICABLE SYMBOLS

REF o REF Catalogue number	LOT Batch code		Manufacturer	Use by
Temperature limitation	Σ Contents sufficient for <n> tests	Consult Instructions for Use	Keep away from direct light	Store in a dry place