



## PRODUCT DATA SHEET

### EE BROTH, MOSSEL (as per ISO)

TM 1804

For selective enrichment for Enterobacteriaceae

### Composition

| Ingredients                    | Gms/Ltr. |
|--------------------------------|----------|
| Dehydrated Ox-bile             | 20.00    |
| Pancreatic digest of Gelatin   | 10.00    |
| Disodium hydrogen phosphate    | 6.450    |
| Glucose monohydrate            | 5.00     |
| Potassium dihydrogen phosphate | 2.00     |
| Brilliant green                | 0.014    |

\*Dehydrated powder, hygroscopic by nature, store in a dry place, in tightly-sealed containers below 25°C and protect from direct sunlight.

### Instructions for Use

Dissolve 43.5gms in 1000ml of distilled water. Dispense 100ml portions in 250ml flasks or in 10ml portions. Gently heat to boiling at 100°C for 30 minutes with gentle swirling and dissolve the medium completely. **DO NOT AUTOCLAVE**. Cool rapidly in cold water and dispense into sterile test tubes as desired.

**Appearance:** Emerald green colour, clear solution.

**pH (25°C):**  $7.2 \pm 0.2$

### Principle

**EE BROTH, MOSSEL** is also commonly known as EE Broth Mossel Medium. Enterobacteria Enrichment Broth Mossel is a highly enrichment medium used for bile-tolerant Gram-negative bacteria. Enterobacteria Enrichment Broth Mossel was developed by Mossel, Visser, and Cornelissen to facilitate the growth of *Enterobacteriaceae*.<sup>4</sup> Enterobacteria Enrichment Broth Mossel is used as an enrichment broth, providing a rich environment for the recovery of damaged or injured cells. Although injured cells may not form colonies on selective media, they can cause infection if ingested.<sup>5</sup> Pancreatic digest of Gelatin provides nitrogen, vitamins, and amino acids in Enterobacteria Enrichment Broth Mossel. Glucose is the carbon source to enhance organism growth. Dehydrated Ox-bile and Brilliant green are the selective agents inhibiting Gram-positive bacteria, particularly bacilli and fecal streptococci. Sodium phosphate and Potassium phosphate are strong buffering agents. Inoculate 10ml. Enterobacteria Enrichment Broth Mossel (TM 1804) medium with a small number (not more than 100 cfu) of the appropriate microorganism and incubate at 30-35°C for 24-48 hours. To prepare a sample for the product to be examined, use a 1 in 10 dilution of not less than 1 gm of the product and use 10 ml or the quantity corresponding to not less than 1gm or 1ml to inoculate 100 ml of Enterobacteria Enrichment Broth Mossel (TM 1804) medium and mix. Incubate at 30-35°C for 18-48 hours. Subculture on a plate of Violet Red Bile Glucose Agar (TM 426) and incubate at 30-35°C for 24-48 hours.

**Note:** Acid production causes the change of Enterobacteria Enrichment Broth Mossel (TM 1804) to yellow while, if the reaction is negative, the medium remains green.

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### Interpretation

Cultural characteristics observed after incubation at 30-35°C for 24 - 48 hours.

| Microorganisms                | ATCC  | Inoculum (CFU/ml) | Growth    | Appearance      | Acid Production |
|-------------------------------|-------|-------------------|-----------|-----------------|-----------------|
| <i>Escherichia coli</i>       | 25922 | 10-100            | Luxuriant | Yellow coloured | +               |
| <i>Pseudomonas aeruginosa</i> | 9027  | 10-100            | Luxuriant | Yellow coloured | +               |
| <i>Enterobacter aerogenes</i> | 13048 | 10-100            | Luxuriant | Yellow coloured | +               |
| <i>Staphylococcus aureus</i>  | 25923 | 10-100            | Inhibited | --              | --              |

### References

1. The United States Pharmacopoeia. Amended Chapters 61, 62 & 111, The United States Pharmacopoeial Convention Inc., Rockville, MD. (2009).
2. Directorate for the Quality of Medicines of the Council of Europe (EDQM). The European Pharmacopoeia, Amended Chapters 2.6.12, 2.6.13, 5.1.4, Council of Europe, 67075 Strasbourg Cedex, France. (2007).
3. Mossel, Vissar, and Cornellisen. J. Appl. Bacteriol. 26:444. (1963).
4. Sorrells, K. M., M. L. Speck, and J. A. Warren. Pathogenicity of *Salmonella gallinarum* after metabolic injury by freezing. Appl. Microbiol. 19:39-43. (1970).
5. ISO 8523: 1991 - Microbiology- general guidance for the detection of Enterobacteriaceae with preenrichment 1991-10-01.