

## Minerals Modified Glutamate Broth (MMBG) ISO

Cat. 1365

 for the enumeration of  $\beta$ -glucuronidase-positive *Escherichia coli*.

### Practical information

| Applications                       | Categories              |
|------------------------------------|-------------------------|
| Selective enumeration              | <i>Escherichia coli</i> |
| Selective enrichment               | <i>Escherichia coli</i> |
| Industry: Water / Food             |                         |
| Regulations: ISO 11133 / ISO 16649 |                         |



### Principles and uses

Minerals Modified Glutamate Broth (MMBG) ISO is recommended by ISO 16649-3 for enumeration of *E.coli* using the most probable number Method (MPN). MMGB Broth is also an alternative broth used for the presumptive identification of coliforms in water.

In comparison to other media, this broth is better as it gives fewer false positive results. Sodium glutamate and sodium formate are the basis of the medium for the enumeration of coliform organisms in water. Lactose is the source of carbohydrates. The addition of vitamins, aminoacids and heptahydrate magnesium sulfate increase fermentation, whereas the addition of ferric ammonium citrate permits increase of gas production. The bromocresol purple is a pH indicator.

### Formula in g/L

|                                 |       |                            |       |
|---------------------------------|-------|----------------------------|-------|
| Bromocresol purple              | 0,01  | Dipotassium phosphate      | 0,9   |
| Ferric ammonium citrate         | 0,01  | Lactose                    | 10    |
| L-Arginine                      | 0,02  | L-Cystine                  | 0,02  |
| Magnesium sulfate heptahydrated | 0,1   | Thiamine                   | 0,001 |
| Sodium glutamate                | 6,35  | Sodium formate             | 0,25  |
| L (-) aspartic acid             | 0,024 | Calcium chloride dihydrate | 0,01  |
| Panthenic acid                  | 0,001 | Nicotinic acid             | 0,001 |

### Preparation

Suspend 17,7 grams of the medium in one liter of distilled water. Add 2,5 grams of Ammonium Chloride. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Dispense into appropriate containers and sterilize in autoclave at 116 °C for 10 minutes.

### Instructions for use

Enumeration of  $\beta$ -glucuronidase-positive *Escherichia coli* according to ISO 16649:

- Inoculate three tubes of a single strength and three tubes of a double strength medium.
- The larger volumes of sample (10 ml) are added to equal volumes of double strength medium, whereas the 1 ml volumes ( or dilution of them) are added to 5 ml of single strength medium.
- Incubate the tubes at 37±1 °C and examine after 24±2 hours, for the acid production (lactose fermentation).
- Each tube of selective enrichment medium showing acid production (yellow color), is subcultured to TBX Chromogenic Agar (Cat. 1151).
- Incubate the TBX plates at 44±1 °C for 22±2 hours.
- The presence of characteristic blue colonies on TBX indicated the presence of *Escherichia coli* in the original MMGB tube.

\* Those strains of *Escherichia coli* that do not grow at 44°C and, in particular, those that are  $\beta$ -glucuronide negative, such as *Escherichia coli* O0157: H7,

will not be detected

## Quality control

| Solubility | Appearance  | Color of the dehydrated medium | Color of the prepared medium | Final pH (25°C) |
|------------|-------------|--------------------------------|------------------------------|-----------------|
| w/o rests  | Fine powder | White with black particles     | Clear purple                 | 6,7 ± 0,1       |

## Microbiological test

According to ISO 11133:

Incubation conditions: Productivity, Selectivity (37±1 °C / 24±2 h).

Inoculation conditions: Productivity qualitative (<=100 CFU) / Selectivity (10<sup>4</sup>-10<sup>6</sup> CFU).

| Microorganisms                   | Specification    | Characteristic reaction |
|----------------------------------|------------------|-------------------------|
| Escherichia coli ATCC 25922      | Acid production  | Colour change to yellow |
| Enterococcus faecalis ATCC 29212 | Total inhibition |                         |
| Escherichia coli ATCC 8739       | Acid production  | Colour change to yellow |

## Storage

Temp. Min.:2 °C

Temp. Max.:25 °C

## Bibliography

ISO 16649-3 Microbiology of food and animal feeding stuffs-Horizontal method for the enumeration of β-glucuronidase-positive Escherichia coli- Part 3: Most probable number technique using 5-bromo-4-chloro-3-indolyl-β-D-glucuronide. Departments of the Environment, Health & Social Security, and P.H.L.S. 1982. The bacteriological examination of drinking water supplies. Report on public Health and Medical Subjects No. 71., H.M.S.O., London, England.