



## Violet Red Bile Glucose Agar w/o Lactose

M581

Violet Red Bile Glucose Agar w/o Lactose is used for detection and enumeration of *Enterobacteriaceae* in raw foods and clinical samples.

### Composition\*\*

| Ingredients                    | Gms / Litre |
|--------------------------------|-------------|
| Peptic digest of animal tissue | 7.000       |
| Yeast extract                  | 3.000       |
| Sodium chloride                | 5.000       |
| Bile salts mixture             | 1.500       |
| Glucose                        | 10.000      |
| Neutral red                    | 0.030       |
| Crystal violet                 | 0.002       |
| Agar                           | 12.000      |
| Final pH ( at 25°C)            | 7.4±0.2     |

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 38.53 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE. Mix well and pour into sterile Petri plates.

### Principle And Interpretation

Violet Red Bile Agar, a modification of MacConkeys original formulation (1) is used for the enumeration of coli-aerogens bacterial group.

Violet Red Bile Glucose Agar w/o Lactose, a modification of VRBA (M049), was designed for the enumeration of *Enterobacteriaceae* (2). It employs the selective inhibitory components crystals violet and bile salts and the indicator system glucose and neutral red. Sought bacteria will dissimilate glucose and produce purple zones around the colonies (3). ISO committee has also recommended this medium (4). Selectivity of VRBGA can be increased by incubation under anaerobic conditions and/ or at elevated temperature, i.e. equal to or above 42°C (5-7).

Peptic digest of animal tissue and yeast extract serve as sources of carbon, nitrogen, vitamins and other essential growth nutrients. Glucose is the fermentable carbohydrate, utilization of which leads to the production of acids. Neutral red indicator detects the acidity so formed. Crystal violet and bile salts mixture help to inhibit the accompanying gram-positive and unrelated flora. Sodium chloride maintains the osmotic equilibrium. Further biochemical tests are necessary for positive identification (8).

### Quality Control

#### Appearance

Light yellow to pinkish beige homogeneous free flowing powder

#### Gelling

Firm, comparable with 1.2% Agar gel.

#### Colour and Clarity of prepared medium

Reddish purple coloured clear to slightly opalescent gel forms in Petri plates.

#### Reaction

#### pH

7.20-7.60

#### Cultural Response

Cultural characteristics was observed after an incubation at 35-37°C for 18-24 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

#### Cultural Response

| Organism                                 | Inoculum (CFU)    | Growth         | Observed Lot value (CFU) | Recovery | Colour of colony               | Incubation temperature |
|--|-------------------|----------------|--------------------------|----------|--------------------------------|------------------------|
| <i>Escherichia coli</i> ATCC 8739        | 50 -100           | luxuriant      | 25 -100                  | >=50 %   | pink-red with bile precipitate | 18 -24 hrs             |
| <i>Pseudomonas aeruginosa</i> ATCC 9027  | 50 -100           | luxuriant      | 25 -100                  | >=50 %   | pink to red                    | 18 -24 hrs             |
| <i>Escherichia coli</i> NCTC 9002        | 50 -100           | good-luxuriant | 25 -100                  | >=50 %   | pink-red with bile precipitate | 18 -24 hrs             |
| <i>Escherichia coli</i> ATCC 25922       | 50 -100           | good-luxuriant | 25 -100                  | >=50 %   | pink-red with bile precipitate | 18 -24 hrs             |
| <i>Salmonella Enteritidis</i> ATCC 13076 | 50 -100           | good-luxuriant | 25 -100                  | >=50 %   | light pink                     | 18 -24 hrs             |
| <i>Enterobacter aerogenes</i> ATCC 13048 | 50 -100           | good-luxuriant | 25 -100                  | >=50 %   | pink-red                       | 18 -24 hrs             |
| <i>Staphylococcus aureus</i> ATCC 25923  | >=10 <sup>3</sup> | inhibited      | 0                        | 0%       |                                | >=24 hrs               |
| <i>Staphylococcus aureus</i> ATCC 6538   | >=10 <sup>3</sup> | inhibited      | 0                        | 0%       |                                | >=24 hrs               |

### Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

### Reference

1. MacConkey A., 1905, J. Hyg., 5, 333-379.
2. Mossel D. A. A., Eclerink I., Koopmans M. and Van Rossem F., 1978, Lab. practice, 27 No. 12: 1049.
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4. International Organization for Standardization (ISO), 1993, Draft ISO/DIS 7402.
5. Mossel D. A. A. and Vega C. L., 1973, Hlth. Lab. Sci., 11:303
6. Mossel D. A. A., Eclerink I., Koopmans M. and Van Rossem F., 1979, Food Protect., 42 : 470
7. Mossel D. A. A. et al, 1986, J. Appl. Bacteriol., 60:289.
8. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore

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