Violet Red Bile Glucose Agar without Lactose

Intended Use
Violet Red Bile Glucose Agar without Lactose is used for enumeration of *Enterobacteriaceae*.

Summary
Violet Red Bile Agar, a modification of MacConkey original formulation is used for the enumeration of coli-aerogenes bacterial group. Violet Red Bile Glucose Agar w/o Lactose, a modification of VRBA, was designed for the enumeration of *Enterobacteriaceae*. 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. ISO committee has also recommended this medium. Selectivity of VRBGA can be increased by incubation under anaerobic conditions and/ or at elevated temperature, i.e. equal to or above 42°C.

Principle
Peptic digest of gelatin and yeast extract serve as sources of carbon, nitrogen, vitamins and other essential growth nutrients. Glucose Monohydrate 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.

Formula*

Ingredients	**g/L**
Peptic Digest of Gelatin	7.0
Yeast Extract	3.0
Sodium Chloride	5.0
Bile Salts Mixture	1.5
Glucose Monohydrate	10.0
Neutral Red	0.03
Crystal Violet	0.002
Agar	15.0
Final pH (at 25°C)	7.4 ± 0.2

*Adjusted to suit performance parameters.

Storage and Stability
Store dehydrated medium below 30°C in tightly closed container and the prepared medium at 2°C-8°C. Avoid freezing and overheating. Use before expiry date on the label. Once opened keep powdered medium closed to avoid hydration.

Type of Specimen
Clinical samples; Food and Dairy samples; Water samples.

Specimen Collection and Handling
Ensure that all samples are properly labelled. Follow appropriate techniques for handling samples as per established guidelines. Some samples may require special handling, such as immediate refrigeration or protection from light, follow the standard procedure. The samples must be stored and tested within the permissible time duration. After use, contaminated materials must be sterilized by autoclaving before discarding.

Directions
1. Suspend 38.53 g of the (the equivalent weight of dehydrated medium per litre) powder in 1000 mL purified / distilled water.
2. Mix thoroughly.
3. Boil with frequent agitation to dissolve the powder completely. DO NOT AUTOCLAVE.
4. Cool to 45°C and pour into sterile petridishes.

Quality Control

Dehydrated Appearance: Light yellow to pinkish beige coloured, homogeneous, free flowing powder.
Prepared Appearance: Light pinkish purple coloured, clear to slightly opalescent gel forms in petridishes.
Growth Promotion Test: Growth promotion is carried out in accordance with the harmonized method of USP/EP/JP and growth is observed after an incubation at 30°C-35°C for 18 to 24 hours.
Growth Promoting Properties: The test results observed are within the specified temperature and shortest period of time specified in the test, inoculating ≤ 100 cfu of appropriate microorganism at 30°C-35°C for 18 hours.
Indicative Properties: The test results observed are within the specified temperature and time, inoculating ≤100 cfu of appropriate microorganism.
Inhibitory Properties: No growth of the test microorganism occurs for the specified temperature and not less than the longest period of the time specified, inoculating >100 cfu of the appropriate microorganism at 30-35°C for ≥ 24 hours.

Growth Promoting + Indicative

Organism (ATCC)	**Growth**	**Colour of Colony**
Escherichia coli (8739)	Good	Pinkish red with bile precipitate
Pseudomonas aeruginosa (9027)	Good	Pink
Inhibitory		
Staphylococcus aureus subsp. *aureus* (6538)	Inhibited	-

Note:
1. For good growth - Growth obtained on test media should not differ by a factor greater than 2 from calculated value for a standardized inoculum.
2. For inhibition no growth of test microorganism should occur.
3. Inoculum for good growth is 10 -100 cfu and that for Inhibition is greater than 100 cfu.

Performance and Evaluation

Performance of the product is dependent on following parameters as per product label claim:
1. Directions
2. Storage
3. Expiry

Precautions / Limitations

1. Over incubation may result in reverting of reaction.
2. Further biochemical tests must be carried out for confirmation.

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

Reference

1. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
2. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
3. Corry J. E. L., Curtis G. D. W. and Baird R. M., (Ed.), 1995, Culture Media for Food Microbiology, Vol. 34, Progress in Industrial Microbiology, Elsevier, Amsterdam.
4. International Organization for Standardization (ISO), 1993, Draft ISO/DIS 7402.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat. No.	Product Description	Pack Size
201220060100	Dehydrated Culture Media	100 g
201220060500	Dehydrated Culture Media	500 g