# **Violet Red Bile Glucose Agar Plate (Harmonized)**

#### **Intended Use**

Violet Red Bile Glucose Agar Plate (Harmonized) is used for isolation and cultivation of *Enterobacteriaceae* from pharmaceutical products in accordance with microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP.

## **Summary**

Enterobacteriaceae includes lactose-fermenting coliform bacteria, non-lactose-fermenting strains of Escherichia coli, and other non-lactose-fermenting species of Salmonella and Shigella involved in food spoilage. Because of their potential contamination of food and dairy products, it is important to detect members of the Enterobacteriaceae rather than traditional coliform bacteria.

Violet Red Bile Glucose Agar Plate (Harmonized) is a modification of Violet Red Agar. Mossel *et al.*, modification lactose containing violet red bile agar by adding glucose. Further work by Mossel *et al.*, showed that the lactose could be omitted resulting in the formulation of Violet Red Bile Glucose Agar. In media glucose is fermented by all members of the *Enterobacteriaceae* thus Violet Red Bile Glucose Agar (VRBGA) gives a presumptive *Enterobacteriaceae* count. VRBGA is recommended by APHA for enumeration of *Enterobacteriaceae* in food samples.

### **Principle**

Pancreatic digest of casein and yeast extract supply nutrients, amino acids, carbon compounds, vitamin B complex, minerals and trace elements. Glucose is an energy source. Bile Salts and Crystal Violet inhibit Gram-positive bacteria. Neutral red is a pH indicator. Agar is the solidifying.

#### Formula\*

Ingredients	g/L
Pancreatic Digest of Gelatin	7.0
Yeast Extract	3.0
Glucose Monohydrate	10.0
Bile Salts	1.5
Sodium Chloride	5.0
Neutral Red	0.03
Crystal Violet	0.002
Agar	15.0

<sup>\*</sup>Adjusted to suit performance parameters.

#### **Additional Material Required**

Bacteriology Incubator.

#### Instruction for use

- 1. Open the sterile pack and remove Violet Red Bile Glucose Agar Plate aseptically.
- 2. Inoculate/streak the plate and Incubate in inverted position as per standard procedure.

## Reading and interpretation

- 1. After incubation, observe the microbial growth and count the colonies.
- 2. Interpretation is assured by user.
- 3. User is responsible to define the action limits as per standard guidelines and alert limits on the basis of trend analysis & other relevant data.

## **Quality Control**

**Appearance:** Gel with smooth and even surface without any cracks, bubbles and drying or shrinking of media.

Colour of Medium: Red with purplish tinge coloured medium.

**Quantity of Medium:**  $27 \pm 2$  g in 90 mm petriplate.

**pH at 25°C \pm 2°C:** 7.4  $\pm$  0.2

**Growth Promotion Test:** Growth promotion is carried out in accordance with the harmonized method of USP/EP/BP/JP/IP and growth is observed after an incubation at 30°C-35°C for 18-24 hours.

**Growth Promoting Properties:** The test results observed are within the specified temperature and shortest period of time, inoculating ≤ 100 cfu of appropriate microorganism.

**Indicative Properties:** The test results observed are within the specified temperature and time, inoculating  $\leq 100$  cfu of appropriate microorganisms.

**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 atleast 100 cfu of the appropriate microorganism.

Organism (ATCC)	Growth	Colour of colony	Incubation Temperature	Incubation Period
Escherichia coli (8739)	Good	Pinkish red with bile precipitate	30°C-35°C	18 Hours
Pseudomonas aeruginosa (9027)	Good	Pink	30°C-35°C	18 Hours
Inhibitory Staphylococcus aureus subsp. aureus (6538)	Inhibited		30°C-35°C	24 Hours

**Note:** For inhibition no growth of test microorganism should occur.

#### **Precautions**

Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or other signs of deterioration.

# Storage and Shelf Life

- 1. Store between 15°C-25°C to avoid water condensation. Condensation can be prevented by avoiding quick temperature shifts and mechanical stress.
- Under optimal conditions, the medium has a shelf life of 6 months. Use before expiry mentioned on the label.

#### Reference

- 1. American Public Health Association. *Standard Methods for the Examination of Dairy Products*, APHA, Washington, D.C.
- 2. APHA Technical Committee on Microbiological Methods for Foods. *Compendium of Methods for the Microbiological Examination of Foods*, APHA, Washington, D.C.
- 3. Draft Standard Methods for Microbiological Examination of Meat Products. 1977. Part 3: *Detection and enumeration of Enterobacteriaceae*. BS5393: Part 3, ISO/DIS 5552.
- 4. Mossel, D.A.A. 1985. Media for Enterobacteriaceae. Int. J. Food. Microbiol.; 2:27.
- 5. Mossel, D.A.A., W.H.J. Mengerink, and H.H. Scholts. 1962. Use of a modified MacConkey agar medium for the selective growth and enumeration of *Enterobacteriaceae*. *J. Bacteriol*.; 84:381.
- 6. Mossel, D.A.A., I. Eelderink, M. Koopmans, and F. van Rossem. 1978. Lab Practice; 27:1049-1050.
- Mossel, D.A.A., I. Eelderink, M. Koopmans, and F. van Rossem. 1979. Influence of carbon source, bile salts and incubation temperature on recovery of *Enterobacteriaceae* from food using MacConkey-type agars. *J. Food Protect*; 42:470.
- 8. USP Chapter 61: Microbiological Examination of Nonsterile Products: Microbial enumeration Tests.
- 9. USP Chapter 62: Microbiological Examination of Nonsterile Products: Tests for Specified Microorganism.
- 10. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

# **Product Presentation:**

Cat No.	Product	Pack Size
205220140100	Violet Red Bile Glucose Agar Plate (Harmonized)	100 Plates

#### Disclaimer

Information provided is based on our inhouse technical data on file, it is recommended that user should validate at his end for suitable use of the product.