

## Violet Red Bile Glucose Agar (VRBGA) (NCM0022)

### Intended Use

Violet Red Bile Glucose Agar is used for the enumeration of *Enterobacteriaceae* in foods. Violet Red Bile Glucose Agar conforms to Harmonized USP/EP/JP requirements and is not intended for use in the diagnosis of disease or other conditions in humans.

### Description

A medium recommended by the Harmonized USP/EP/JP for isolation and identification of bile-tolerant Gram-negative bacteria from non-sterile products. Conforms to Harmonized USP/EP/JP performance specification. Yeast extract provides vitamins and gelatin serves as source of carbon and nitrogen. Glucose is a fermentable carbohydrate and sodium chloride maintains the osmotic balance. Bile salts and crystal violet act as selective agents inhibiting many Gram-positive bacteria. The formulation is a modification of Violet Red Bile Agar by Mossel which substitutes lactose for glucose. *Enterobacteriaceae*, such as *Escherichia coli* and *Salmonella spp.*, are able to ferment glucose. This produces acid which results in a pH drop indicated by neutral red resulting in pink colonies. Enough acid production will cause the precipitation of bile salts resulting in a bile precipitate or halo around glucose fermenting bacteria. Non-glucose fermenting bile tolerant bacteria such as *Pseudomonas aeruginosa* grow but remain colorless with no bile precipitate. According to the Harmonized USP/EP/JP, Enterobacteria Enrichment Broth-Mossel is used as a selective enrichment broth, with subculture performed onto Violet Red Bile Glucose Agar (VRBGA).

### Typical Formulation

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

pH: 7.4 ± 0.2 at 25°C.

Formula may be adjusted and/or supplemented as required to meet performance specifications.

### Precaution

Refer to SDS

### Preparation

1. Suspend 41.5 grams of the medium in 1 liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. DO NOT AUTOCLAVE. Cool to 45-50°C.

### Test Procedures

Inoculate directly on Violet Red Bile Glucose Agar by the streak method from selective or nonselective medium or from the sample itself.

# Technical Specification Sheet



## Quality Control Specifications

**Powder:** Fine, free-flowing, homogeneous, light beige to reddish purple.

**Finished medium:** Reddish purple, clear.

**Expected Cultural Response and USP/EP/JP Growth Promotion Testing:** Cultural response on Violet Red Bile Glucose Agar incubated at Harmonized USP/EP/JP specified temperatures and incubation times.

Microorganism	Approx. inoculum (CFU)	Expected Results	
		Growth	Reaction
<i>Escherichia coli</i> ATCC® 8739	10-100	Growth	Pink colonies w/ red precipitate
<i>Escherichia coli</i> ATCC® 25922	10-100	Growth	Pink colonies w/ red precipitate
<i>Pseudomonas aeruginosa</i> ATCC® 9027	10-100	Growth	Colorless to grey
<i>Salmonella typhimurium</i> ATCC® 14028	10-100	Growth	Pink colonies w/ red precipitate
<i>Staphylococcus aureus</i> ATCC® 6538	>10 <sup>3</sup>	Inhibited	N/A
<i>Enterococcus faecalis</i> ATCC® 29212	>10 <sup>3</sup>	Inhibited	N/A

The organisms listed are the minimum that should be used for quality control testing.

## Results

*Enterobacteriaceae* ferment dextrose, produce acid products, and form pink to reddish colonies with reddish precipitate.

## Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

## Limitations of the Procedures

Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

## Storage

Store dehydrated culture media at 2 – 30°C away from direct sunlight. Once opened and recapped, place the container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

## References

1. Mossel, D.A.A. Media for Enterobacteriaceae (1985) International Journal of Food Microbiology, 2 (1-2), pp. 27-32.
2. European Pharmacopoeia 10<sup>th</sup> Edition (2020)
3. United States Pharmacopoeia National Formulary 2018: USP 41 NF 36
4. Japanese Pharmacopoeia 17<sup>th</sup> Edition (2017)



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