

**VIOLET RED BILE AGAR**  
**Powdered and ready to use medium**  
**for the isolation and enumeration of coliforms in foodstuffs.**

**TYPICAL FORMULA (g/l)**

Peptone	7.000
Yeast Extract	3.000
Bile Salts No.3	1.500
Lactose	10.000
Sodium Chloride	5.000
Neutral Red	0.030
Crystal Violet	0.002
Agar	15.000

**DIRECTIONS FOR POWDERED MEDIUM**

Suspend 41.5g in 1000ml of cold distilled water. Heat to boiling and boil for about 2 minutes, cool in a water bath to approximately 45°C and transfer to inoculated Petri dishes. Sterilisation is not necessary. However, if the plates have to be stored in the refrigerator it is advisable to autoclave the medium at 121°C for 15 minutes. This will not appreciably diminish the fertility of the medium.

Final pH 7.4 ± 0.2

**DIRECTIONS FOR READY TO USE MEDIUM IN FLASKS**

Dissolve the contents of the bottle in a temperature controlled water bath (100°C). Cool to 45-50°C, mix well and distribute into sterile Petri dishes

Final pH 7.4 ± 0.2

**DESCRIPTION**

Violet Red Bile Agar is a selective and differential medium recommended by the ISO 4832 for the isolation and enumeration of coliforms in foodstuffs.

Violet Red Bile Agar contains Bile Salts No.3 and crystal violet, which inhibit the growth of Gram-positive bacteria; the neutral red permits differentiation of lactose-fermenting from non-lactose-fermenting microorganisms. Lactose fermentation causes acidification of the medium, with a consequent colour change of the indicator to violet-red and a precipitation of the bile salts.

**TECHNIQUE**

Prepare the test sample and the decimal dilutions in accordance with the specific Laboratory method using Maximum Recovery Diluent (REF 401691) or other suitable diluent.

Take two sterile Petri dishes and by means of a sterile pipette, transfer 1ml of the test sample, if the product is liquid, or 1ml of the initial suspension in the case of other products.

Take two other sterile Petri dishes and transfer, by means of an other sterile pipette to each dish 1ml of the first decimal dilution ( $10^{-1}$ ) of the test sample, if the product is liquid, or 1ml of the first decimal dilution ( $10^{-2}$ ) of the initial suspension in the case of other products.

If necessary, repeat the procedure with the further dilutions using a fresh sterile pipette for each decimal dilution.

Pour about 15ml of the Violet Red Bile Agar cooled to 45 °C into each Petri dishes.

Carefully mix the inoculum with the medium by rotating the plates and allow to solidify the Petri dishes on a cool horizontal surface.

Pour about 4ml of the Violet Red Bile Agar cooled to 45°C on the surface of inoculated medium and allow to solidify.

Invert the prepared Petri dishes and place them in the incubator at 30°C or 37°C for 24 ± 2 hours.

After incubation for 24 hours coliforms grow on Violet Red Bile Agar with purplish red colonies, 0.5mm or more in diameter.

**USER QUALITY ASSURANCE** (37°C-24 hrs)

Productivity control

*E.coli* ATCC 25922: growth, purplish red colonies

Specificity control

*P.aeruginosa* ATCC 27853 : growth, pale green colonies

Selectivity control

*E.faecalis* ATCC 19433 : inhibited**STORAGE**

Dehydrated medium: 10-30°C

User prepared flasks or plates: 5 days at 2-8°C

**REFERENCES**

- APHA (1995), Standard Methods for the Examination of Dairy Products, 15th edition
- APHA (1992), Compendium of Methods for the Microbiological Examination of Foods, 3<sup>rd</sup> ed.
- ISO 4832:1991 Microbiology-General guidance for the enumeration of coliforms–colony count technique.

**PACKAGING**

<b>4021851</b>	<b>Violet Red Bile Agar</b>	<b>100g (2.4 l)</b>
<b>4021852</b>	<b>Violet Red Bile Agar</b>	<b>500g (12 l)</b>
<b>4021854</b>	<b>Violet Red Bile Agar</b>	<b>5kg (120 l)</b>
<b>542185</b>	<b>Violet Red Bile Agar</b>	<b>20 ready to use plates</b>
<b>5121852</b>	<b>Violet Red Bile Agar</b>	<b>6x100 ml ready to use flasks</b>
<b>5121853</b>	<b>Violet Red Bile Agar</b>	<b>6x200 ml ready to use flasks</b>