Agar medium F

(VRBG/Crystal Violet-Neutral Red-Bile-Glucose agar medium)

355-4239
356-4584

DEFINITION

Medium used for the detection and enumeration of enterobacteria in food products.

STANDARDS

***European Pharmacopeia* 6.0 -** Biological methods - **2.6.13.**: Microbiological test of non-sterile products (Detection of specified micro-organisms).

PRINCIPLE

The principle of the medium relies on the ability of *Enterobacteriaceae* to ferment glucose. The simultaneous presence of crystal violet and bile salts makes the medium inhibit Gram-positive bacteria and some Gram-negative bacteria. Neutral red is an indicator of the pH.

PRESENTATION

- **Ready to use**
  100 ml x 6 bottles **code 355-4239**

- **Dehydrated**
  500 g **code 356-4584**

STORAGE

- Ready to use: +2-8°C
- Dehydrated: +15-25°C, in carefully-sealed bottles in a cool, dry place
- Expiration date and batch number are shown on the package.

TYPICAL FORMULA

Peptone	7 g
Yeast extract	3 g
Sodium chloride	5 g
Bile salts	1.5 g
Glucose	10 g
Neutral red	30 mg
Crystal violet	2 mg
Agar	12 g
Distilled water	1,000 ml

Final pH (25°C) = 7.4 ± 0.2

NB: the formula has been adapted to attain the required performance criteria.

OTHER PRODUCTS REQUIRED (NOT SUPPLIED)

- Diluent(s)
- Distilled water

EQUIPMENT REQUIRED (NOT SUPPLIED)

(non-exhaustive)

- Scales
- Sterile weighing bags
- Grinder
- Hotplate
- Mixer-homogenizer
- 125 ml Pyrex bottles with autoclave-proof stoppers
- Sterile Petri dishes (Ø = 90 mm)
- Sterile pipettes (1 ml, etc)
- Water-bath precise to ±1°C
- Thermostatically-controlled incubator or incubation room, precise to ±1°C
- All usual laboratory equipment

PREPARATION OF DEHYDRATED MEDIUM

Always shake well before use.
Dissolve 38.5 g of powder in 1 liter of distilled water. Wait for 5 minutes, then mix thoroughly until a homogenous suspension is obtained. Heat gently, agitating frequently, then bring to boiling point until completely dissolved.
Dispense.
Do not autoclave the medium.

Reconstitution ratio: 38.5 g/l
500 g of powder makes 13 liters of medium.

PROTOCOL

- **Preparation of samples**

According to the standards or recommendations applicable to the product concerned.

- **Inoculation and incubation**

Inoculate 1 ml of the product to be analyzed, or its decimal dilutions, into sterile Petri dishes. Pour about 15 ml of medium, melted and cooled to 44-47°C, homogenize and leave to solidify.
Pour a second layer (approximately 2 cm thickness) of this medium maintained at 44-47°C and leave to dry again.
Incubate at 30°C, 35°C or 37°C ± 1°C for 24 hr ± 2 hours.

Bio-Rad, 3 boulevard Raymond Poincaré • 92430 Marnes-la-Coquette
Tel. : +33 (0)1 47 95 60 00 • Fax : +33 (0)1 47 41 91 33
www.foodscience.bio-rad.fr

Agar medium F
(VRBG/Crystal Violet-Neutral Red-Bile-Glucose agar medium)

READING AND INTERPRETATION
After 24 hours incubation, enumerate typical *Enterobacteriaceae* colonies on dishes containing between 15 and 150 colonies.
Enterobacteriaceae form pink-red colonies (Glucose positive) with a diameter equal to or exceeding 0.5 mm, with or without zone of bile precipitation.

PRECAUTIONS
- The time lapse between the end of preparation of the stock solution (or the 10^{-1} dilution in the case of a solid product) and the moment when the dilutions come into contact with the culture medium must not exceed 15 minutes.
- Do not autoclave.
- Comply with Good Laboratory Practice.

QUALITY CONTROL
In view of the current harmonization of pharmacopeias, we recommend that you refer to the certificates of analysis for procedures relating to the quality control (performance and selectivity) of media produced by Bio-Rad.

Every product manufactured and marketed by Bio-Rad is subject to a quality-assurance procedure at all stages, from the reception of raw materials to the marketing of the end-product. Each batch of finished product undergoes quality control and is marketed only if it satisfies the acceptability criteria.

Documentation relative to the production and control of each batch is kept on file.

KEY WORDS
VRBG/*Enterobacteriaceae*/Food products/ Detection/Enumeration/Crystal Violet/Bile salts/Glucose/Fermentation/MPN/Medium

BIBLIOGRAPHY
MOSSEL D.A.A., MENGERINK W.H.J. and SCHOLTS H.H. (1962): Use of a modified MacConkey agar medium for the selective growth and enumeration of all *Enterobacteriaceae*. Journal of Bacteriology **84**: 381

Bio-Rad, 3 boulevard Raymond Poincaré • 92430 Marnes-la-Coquette
Tel. : +33 (0)1 47 95 60 00 • Fax : +33 (0)1 47 41 91 33
www.foodscience.bio-rad.fr