

Mac Conkey/Agar (Lactose with crystal violet)

356-3617
356-9084

DEFINITION

Medium used to isolate and enumerate coliforms bacilli, and particularly *Escherichia coli* in the testing of microbial contamination of non-sterile Pharmacopeia products (2.6.13.).

STANDARDS

FOOD MICROBIOLOGY

- **NF EN ISO 21567 (March 2005):** Microbiology of food and animal feeding stuffs - Horizontal method for the detection of *Shigella* spp.

PRINCIPLE

The principle of the medium rests on the ability of coliforms to ferment lactose (brick-red colonies).

Due to the presence of bile salts and crystal violet this medium inhibits Gram-positive bacteria.

PRESENTATION

- **Ready-to-use**
90 mm x 20 dishes **code 356-3617**
- **Dehydrated**
500 g **code 356-9084**

STORAGE

- Ready-to-use: + 2°C to 20°C
- Dehydrated: + 15°C to 25°C, in carefully-sealed bottles in a cool, dry place.
- Expiration date and batch number are shown on the package.

THEORETICAL FORMULA

Bacteriological peptone	20 g
Bile salts	1.5 g
Sodium chloride	5 g
Lactose	10 g
Neutral red	30 mg
Crystal violet	1 mg
Agar	15 g
Distilled water	1,000 ml
Final pH (25°C) = 7.1 ± 0.2	

OTHER PRODUCTS REQUIRED (NOT SUPPLIED)

- 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
- Autoclave
- All usual laboratory equipment.

PREPARATION OF DEHYDRATED MEDIUM

Always shake before use

Dissolve 51.5 g of powder in 1 liter of distilled water. Bring to the boil until completely dissolved.

Dispense 100 ml per bottle and sterilize in autoclave at 121°C ± 1°C for 15 minutes. Cool to 44°C - 47°C then pour into Petri dishes. After solidification, leave to dry in incubator at 37°C ± 1°C (partially closed).

Reconstitution ratio: 51.5 g/l.
500 g of powder makes 9.7 liters of medium.

PROTOCOL

Inoculation and incubation

After presumptive screening, carry out sub-culture on Mac Conkey medium. Incubate at 44°C ± 1°C for 18-24 hours.

READING AND INTERPRETATION

- "Lactose-negative colonies": colorless.
- "Lactose-positive colonies": brick-red, sometimes surrounded by an opaque halo of precipitated bile salts.

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.
- Comply with Good Laboratory Practice.

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PERFORMANCES / QUALITY CONTROL OF THE TEST

The growth performances of the media are verified with the following strains:

STRAINS	Results of 24h culture at 37°C
<i>Salmonella Enteritidis</i> ATCC 13076	Colorless colonies Lactose-negative
<i>Proteus mirabilis</i> ATCC 25933	Colorless colonies Lactose-negative
<i>Escherichia coli</i> ATCC 25922	Pink to red colonies * Lactose-positive
<i>Klebsiella pneumoniae</i> ATCC 13883	Pink to red colonies Lactose-positive
<i>Pseudomonas aeruginosa</i> ATCC 27853	Colorless colonies Lactose-negative

* Precipitation halo

QUALITY CONTROL OF MANUFACTURER

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

Mac Conkey with crystal violet / Coliforms / *Escherichia coli* / Food products / Isolation / Enumeration / Lactose / Medium.