

# Instructions for use

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# TAYLOR LYSINE DECARBOXYLASE BROTH

Dehydrated culture medium



#### 1 - INTENDED USE

The medium is used, together with other biochemical tests, for the confirmation of *Salmonella* colonies, isolated from samples of the food chain.

| <b>2 - COMPOSITION TYPICAL FOR</b>         | MULA  |  |  |
|--|-------|--|--|
| (AFTER RECONSTITUTION WITH 1 L OF WATER) * |       |  |  |
| Yeast extract                              | 3 g   |  |  |
| Glucose                                    | 1 g   |  |  |
| L-lysine                                   | 5 g   |  |  |
| Bromocresol purple                         | 15 mg |  |  |

\*The formula may be adjusted and/or supplemented to meet the required performances criteria.

Taylor Lysine Decarboxylase Broth – from the left: uninoculated tube, *P. vulgaris* Lys -, *S.* Enteritidis Lys +

# **3 - PRINCIPLE OF THE METHOD AND EXPLANATION OF THE PROCEDURE**

The amino acid decarboxylation test was developed by Moeller<sup>1</sup> in 1955 for the determination of lysine and ornithine decarboxylase and arginine dehydrolase. Falkow<sup>2</sup> in 1958 modified Moeller's formula for differentiating *Salmonella* and *Shigella*. Taylor<sup>3</sup> in 1961 modified the Falkow medium by omitting the peptone from the formulation, because it is the origin of false positive results given by *Citrobacter* and other bacteria, due to its oxidation and deamination with the formation of an alkaline environment. The good results obtained by Taylor have been confirmed by Bonev<sup>4</sup> in a comparative study of the three media with 2764 strains of *Enterobacteriaceae*.

Taylor Lysine Decarboxylase Broth corresponds to Lysine decarboxylation medium (LDC), recommended by ISO 6579<sup>5</sup> as a confirmatory test for *Salmonella* colonies cultivated on selective medium, together with TSI and urease test. Yeast extract provide nitrogen, carbon, vitamins and trace elements for bacterial growth. Glucose is the fermentable carbohydrate and bromocresol purple is the pH indicator. The amino acid L-lysine is included to detect the production of the specific enzyme lysine decarboxylase that removes COOH group from the lysine to produce CO<sub>2</sub> and cadaverine, an alkaline polyamine. Facultatively fermenting bacteria ferment glucose resulting in the production of acid, which lower the pH of the medium and activate the enzyme lysine decarboxylase. When the medium containing lysine is inoculated with a glucose-fermenting and decarboxylase positive strain, it turns first to yellow for the production of acids then to purple for the production of alkalinity, based on the formation of amines. The positive test for lysine decarboxylase is therefore indicated by the formation of a purple color, the negative test by the presence of a yellow color.

# 4- DIRECTIONS FOR DEHYDRATED MEDIUM PREPARATION

Suspend 9 g in 1000 mL of cold purified water. Heat with frequent agitation to dissolve the medium completely. Distribute 2-5 mL in screw-capped tubes, and sterilize by autoclaving at 121°C for 15 minutes.

# **5 - PHYSICAL CHARACTERISTICS**

Dehydrated medium appearancewhitish, fine, homogeneous, free-flowing powderPrepared medium appearanceviolet, limpid or slightly opalescentFinal pH at 20-25 °C $6.8 \pm 0.2$ 

## 6 - MATERIALS PROVIDED - PACKAGING

| Product Type REF Pack   |  |
|---|--|
|   |  |
| Taylor Lysine Decarboxylase Broth         Dehydrated medium         401367L2         500 g (55.5 L) |  |

# 7 - MATERIALS REQUIRED BUT NOT PROVIDED

Autoclave, water-bath, sterile microbiological loops or needles, incubator and laboratory equipment as required, Erlenmeyer flasks, ancillary culture media and reagents.

# 8 - SPECIMENS

The specimens consist of bacteria strains isolated from food sample on *Salmonella* primary isolation agar and purified on appropriate medium (e.g., Nutrient Agar).

# 9 - TEST PROCEDURE

With an inoculating needle or loop, transfer one colony into the tube just below the surface of the medium. Incubate the tubes, with the caps tightened, at  $37^{\circ}$ C for  $24 \pm 3$  hours.

#### **10 - READING AND INTERPRETATION**

After incubation, observe the presence of growth (turbidity) and the colour change of the medium. Turbidity and a purple colour after incubation indicate a positive reaction (decarboxylation of lysine). A yellow colour indicates a negative reaction (fermentation of glucose). The majority of typical *Salmonella* cultures show a positive reaction.

#### **11 - USER QUALITY CONTROL**

All manufactured lots of the product are released for sale after the Quality Control has been performed to check the compliance with the specifications. However, the end user can perform its own Quality Control in accordance with the local applicable regulations, in compliance with accreditation requirements and the experience of the Laboratory. Here below are listed some test strains useful for the quality control.

| <u></u> |
|---------|
|---------|

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CONTROL STRAINS *P. vulgaris* ATCC 9484 *S.* Enteritidis ATCC 13076 INCUBATION T°/ T / ATM 37° / 24 H /A 37° / 24 H /A

EXPECTED RESULTS negative test (yellow colour) positive test (purple)

A: aerobic incubation; ATCC is a trademark of American Type Culture Collection

# **12 - PERFORMANCES CHARACTERISTICS**

Prior to release for sale, a representative sample of all lots of dehydrated Taylor Lysine Decarboxylase Agar is tested for specific performance characteristics by comparing the results with a previously approved Reference Batch.

Pure cultures, grown for 18–24 h on Tryptic Soy Agar, of lysine decarboxylase positive and negative strains are inoculated directly into the tubes. Lysine decarboxylase positive strains: S.Typhimurium ATCC 14028, S.Enteritidis ATCC 13076, S.arizonae ATCC 13314, S.Dublin CB 9.5; Lysine decarboxylase negative strains: C.freundii ATCC 8090, P.vulgaris ATCC 9484, P.mirabilis ATCC 10005, P.stuartii ATCC 33672. The tubes are incubated with loosened caps at 37 °C for 24 hours. For all strains the reactions are conform to the specifications. CB: Biolife microbial collection

# **13 - LIMITATIONS OF THE METHOD**

- With the tubes held vertically during incubation, the decarboxylase test may show two layers of different colour; shake the tube gently before attempting to make an interpretation.<sup>6</sup>
- Taylor medium with lysine is indicated to differentiate *Citrobacter* (generally negative) from *Salmonella* (generally positive); however, *Salmonella* Paratyphi A gives negative reactions (yellow test tube).<sup>6</sup>
- Lysine decarboxylation is one of the tests necessary for the identification of *Salmonella*. The result of the decarboxylation test must be interpreted together with other tests for a correct identification of the strains.

#### **14 - PRECAUTIONS AND WARNINGS**

- This product is for microbiological control and for professional use only; it is to be used by adequately trained and qualified laboratory personnel, observing approved biohazard precautions and aseptic techniques.
- Dehydrated media must be handled with suitable protection. Before use, consult the Safety Data Sheet.
- Apply Good Manufacturing Practice in the production process of prepared media.
- All laboratory specimens should be considered infectious.
- The laboratory area must be controlled to avoid contaminants such as culture medium or microbial agents.
- Sterilize all biohazard waste before disposal. Dispose the unused medium and the sterilized medium inoculated with samples or microbial strains in accordance with current local legislation.
- Do not use the culture medium as active ingredient for pharmaceutical preparations or as production material intended for human and animal consumption
- The Certificates of Analysis and the Safety Data Sheet of the product are available on the website www.biolifeitaliana.it.
- The information provided in this document has been defined to the best of our knowledge and ability and represents a guideline for the
  proper use of the product but without obligation or liability. In all cases existing local laws, regulations and standard procedures must be
  observed for the examination of samples collected from human and animal organic districts, for environmental samples and for products
  intended for human or animal consumption. Our information does not relieve our customers from their responsibility for checking the
  suitability of our product for the intended purpose.

#### **15 - STORAGE CONDITIONS AND SHELF LIFE**

Upon receipt, store at +10°C /+30°C away from direct light in a dry place. If properly stored, it may be used up to the expiration date. Do not use beyond this date. Avoid opening the bottle in humid places. After use, the container must be tightly closed. Discard the product if the container and/or the cap are damaged, or if the container is not well closed, or in case of evident deterioration of the powder (colour changes, hardening, large lumps).

The user is responsible for the manufacturing and quality control processes of prepared media and the validation of their shelf life, according to the type (plates/tubes/bottles) and the applied storage conditions (temperature and packaging).

According to ISO 6579 the self-prepared tubes may be stored at 2-8 for up to three months.<sup>5</sup>

#### 16 - REFERENCES

- 1. Moeller V. Simplified tests for some amino acid decarboxylases and for the arginine dihydrolase system. Acta Pathol Microbiol Scand 1955;36(2):158-72.
- Falkow S. Activity of lysine decarboxylase as an aid identification of *Salmonella* and *Shigella*. Amer J Clin Path1958; 29: 589-600.
   Taylor, WI. Isolation of Salmonellae from Food Supplies. V. Determination of the Method of Choice for Enumeration of *Salmonella*. Appl Microbiol 1961; 9:487-490.
- Bonev SI, Zakhariev Z, Gentchev P. Comparative Study of Media for Determination of Lysine Decarboxylase Activity. App Microbiol 1974; 27: 464-468.
   ISO 6579:2017 Microbiology of the food chain Horizontal method for the detection, enumeration and serotyping of Salmonella —Part 1: Detection of
- Salmonella spp.
   MacFaddin JF. Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria. Baltimore: Williams & Wilkins; 1985.

#### TABLE OF APPLICABLE SYMBOLS

| ADEL OF ATTEICADEL STINE       |   |                                    |                                   |        |
|--------------------------------|---|------------------------------------|-----------------------------------|--------|
| REF or REF<br>Catalogue number | LOT Batch code                              | Manufacturer                       | Store in a dry place              | Use by |
| Temperature<br>limitation      | Contents<br>sufficient for<br><n> tests</n> | Consult<br>Instructions for<br>Use | Keep away<br>from direct<br>light |        |

#### **REVISION HISTORY**

| Version    | Description of changes                                  | Date    |
|------------|---|---------|
| Revision 1 | Updated layout and content                              | 2020/05 |
| Revision 2 | Update of chapter 6, 14 and 15; inclusion of chapter 12 | 2022/09 |

Note: minor typographical, grammatical, and formatting changes are not included in the revision history.

