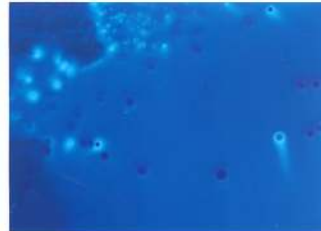
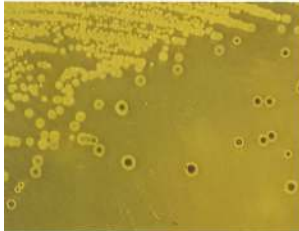


INSTRUCTIONS FOR USE

MUCAP TEST



Salmonella sp. and *Proteus mirabilis* mixed culture on Hektoen Enteric Agar, flooded with MUCAP Test reagent.
At left: normal light, at right under Wood's lamp

1 - INTENDED USE

In vitro diagnostic. Liquid reagent for rapid differentiation of *Salmonella* spp. colonies directly on selective enteric plating media.

2 - COMPOSITION – BOTTLE CONTENT

4- methylumbellyferil caprylate dissolved in heptane 8 mL

3 - PRINCIPLE OF THE METHOD AND EXPLANATION OF THE PROCEDURE

Gastrointestinal infections are still a global public health problem, being a leading cause of medical consultation. Among the bacteria responsible for these syndromes, *Salmonella* spp. are one of the most common causes of bacterial diarrheal diseases.

The fluorogenic substrates, derived from methylumbelliferone and coumarins, have been widely tested for the rapid identification of various microbial species and have demonstrated high sensitivity.^{1,2}

The MUCAP Test reagent, developed and synthesized by Biolife and earlier described in some scientific papers,^{3,4} is an easy and rapid screening test for presumptive identification of *Salmonella* colonies, directly on selective enteric plating media,^{5,6} using an eight-carbon-atom ester conjugated with methylumbelliferone.

The MUCAP Test reagent detects the C₈ esterase enzyme, encoded by the *apeE* gene, present in *Salmonella* spp.⁹ The MUCAP Test reagent consists of 4-methylumbellyferil caprylate dissolved in heptane; this substrate interacts with the *Salmonella* C₈ esterase in a specific manner, leading to the rapid release on the medium of umbelliferone, strongly fluorescent under Wood's lamp at 366 nm.

4 - PHYSICAL CHARACTERISTICS

Reagent appearance colourless, limpid

5 - MATERIALS PROVIDED - PACKAGING

Product	Type	REF	Pack
MUCAP Test CND: W0104080208, EDMA: 14.02.02.08, RDM: 1483858/R	Identification reagent	191500	8 mL (160 tests) Primary packaging: glass bottle with dropper Secondary packaging: cardboard box

6 - MATERIALS REQUIRED BUT NOT PROVIDED

Wood's lamp (366 nm), ancillary culture media and reagents for the identification of the colonies.

7 - SPECIMENS

In clinical and non-clinical microbiology, the specimens consist of suspected *Salmonella* colonies grown on selective enteric plating media. MUCAP Test reagent cannot be used for the direct testing of clinical specimens.

8 - TEST PROCEDURE

The test is carried-out by flooding with one drop of MUCAP reagent all the suspect *Salmonella* colonies (lactose negative and H₂S positive or negative colonies) cultivated on a selective enteric plating medium.

The following scheme should be used:

1. Observe the colonies under a Wood's lamp (366nm) before adding the reagent to ensure that no spontaneous fluorescence occurs.
2. Add a drop of reagent to each isolated colony or to a group of colonies.
3. After 3-5 minutes observe the plates under the Wood's lamp (wavelength 366 nm) in semi-darkness.

9 - READING AND INTERPRETATION

Positive result: appearance of a blue fluorescence over the whole colony or on the edge of black centred colonies.

Negative result: no development of fluorescence

The fluorescent colonies can be presumptively identified as *Salmonella* and subjected to a complete identification with appropriate tests.

Fluorescence negative colonies can be considered non-*Salmonella* and therefore plates are discarded.

10 - 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, it is responsibility of the end-user to perform Quality Control testing in accordance with the local applicable regulations, in compliance with accreditation requirements and the experience of the Laboratory. Here below are listed the test strains useful for the quality control.

Positive control: *S.Typhimurium* ATCC 14028

Negative control: *P.mirabilis* ATCC 25933

ATCC is a trademark of American Type Culture Collection





11 - PERFORMANCES CHARACTERISTICS

Several experimental works have been published for the evaluation of the specificity and sensitivity of MUCAP Test in identifying *Salmonella* colonies. The sensitivity values are almost constantly close to 100%;⁷⁻¹⁹ the specificity values are more variable and sometimes dependent on the isolation and enrichment media combination and vary between 80% and 100%.⁷⁻¹⁹

The specificity and sensitivity of the detection lactose negative, H₂S positive and MUCAP positive colonies are respectively of 100% and 99.8%.¹⁵

Prior to release for sale a representative sample of all lots of MUCAP Test is tested with positive and negative strains.

C₈ esterase positive strains (fluorescent under Wood's lamp within 5 minutes): *S.Typhimurium* ATCC 14028, *S.Enteritidis* NCTC 5188, *S.Gallinarum* CB 506, *S.arizonae* CB 302, *S.Derby* CB 1.5, *S.Dublin* CB 9.2 *S. Choleraesuis* CB X4.

C₈ esterase negative strains (not fluorescent under Wood's lamp within 5 minutes): *P.mirabilis* ATCC 25933, *P.vulgaris* ATCC 13315, *C.freundii* ATCC 8090, *S.marcescens* ATCC 8100, *H.alvei* CB 910.

12 - LIMITATIONS OF THE METHOD

- Before carrying out the test it is advisable to examine the plates under Wood's lamp for the presence of natural fluorescence developed by *Pseudomonas* spp. Strains with natural fluorescence should not be subjected to the MUCAP Test
- Lactose positive *Salmonella* strains, with atypical colonies on conventional lactose-containing media such as SS Agar, Hektoen Enteric Agar etc, may not be detected with MUCAP Test; however, these strains can be detected with the reagent on lactose-free media.
- Since most of the false positive strains are oxidase positive,⁶ the H₂S negative and fluorescence positive colonies can be tested with the oxidase reagent or strips. The combination of MUCAP and oxidase tests raises the specificity of C₈ esterase detection from 94.1% to 98.2%.¹⁶
- The reagent does not affect the viability of the flooded colonies.
- Do not take any reading after 5 minutes because the reagent is susceptible to autolysis due to the watery nature of the medium and so false positive results might occur.
- The MUCAP Test can be carried-out on the most commonly used culture media (e.g. Mac Conkey Agar, SS Agar, Hektoen Enteric Agar, Brilliant Green Agar, Desoxycholate Agar, XLD Agar etc.). MUCAP Test cannot be performed on colonies cultivated on Bismuth Sulphite Agar.
- It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed for complete identification of the MUCAP positive colonies. If relevant, perform antimicrobial susceptibility testing.
- This culture medium is intended as an aid in the diagnostic procedures of infectious diseases; the interpretation of the results must be made considering the patient's clinical history, the origin of the sample and the results of other diagnostic tests.

13 - PRECAUTIONS AND WARNINGS

- This product is a qualitative *in vitro* diagnostic, for professional use only; it is to be used by adequately trained and qualified laboratory personnel, observing approved biohazard precautions and aseptic techniques.
- This product is classified as flammable according to the current European legislation. Consult the Safety Data Sheet before the use.
- Apply good laboratory practice guidelines when performing the test.
- All laboratory specimens should be considered infectious.
- The laboratory area must be controlled to avoid contaminants such as the reagent, the culture media or the microbial agents.
- Sterilize all biohazard waste before disposal. Dispose the unused reagent and the plates inoculated with samples or microbial strains in accordance with current local legislation.
- Do not use this product 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 are available on the website www.biolifeitaliana.it.
- Notify Biolife Italiana Srl (complaint@biolifeitaliana.it) and the relevant Authorities of any serious incident occurring in connection with the use of the *in vitro* diagnostic
- 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.

14 - STORAGE CONDITIONS AND SHELF LIFE

Upon receipt, store the reagent in the original pack at 2-8°C away from direct light. If properly stored, the reagent may be used up to the expiration date. Do not use the reagent beyond this date. Opened bottle can be used up to the expiration date. Repeated openings of the bottle do not affect the performances and do not cause contamination of the reagent. Do not use the reagent with signs of deterioration (turbidity, precipitate, atypical colour).

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191500 MUCAP TEST

SDS rev 4

Regulation (EU) 2020/878

Classification

The product is classified as hazardous

Hazard classification and indication:

Flammable liquid, category 2	H225	Highly flammable liquid and vapour.
Aspiration hazard, category 1	H304	May be fatal if swallowed and enters airways.
Skin irritation, category 2	H315	Causes skin irritation.
Specific target organ toxicity - single exposure, category 3	H336	May cause drowsiness or dizziness.
Hazardous to the aquatic environment, acute toxicity, category 1	H400	Very toxic to aquatic life.
Hazardous to the aquatic environment, chronic toxicity, category 1	H410	Very toxic to aquatic life with long lasting effects.

Hazard labelling

Pictogram



Hazard statements:

H225	Highly flammable liquid and vapour.
H304	May be fatal if swallowed and enters airways.
H315	Causes skin irritation.
H336	May cause drowsiness or dizziness.
H410	Very toxic to aquatic life with long lasting effects.

Precautionary statements:

P210	Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
P331	Do NOT induce vomiting.
P280	Wear protective gloves/ protective clothing / eye protection / face protection.
P301+P310	IF SWALLOWED: Immediately call a POISON CENTER / doctor / . . .
P370+P378	In case of fire: use . . . to extinguish.
P273	Avoid release to the environment.
Contains:	HEPTANE

TABLE OF APPLICABLE SYMBOLS

REF or REF Catalogue number	LOT Batch code	IVD <i>In vitro</i> Diagnostic Medical Device	Manufacturer	This side up	
Temperature limitation	Content sufficient for <n> tests	Consult Instructions for Use	Use by	Keep away from direct light	Fragile

REVISION HISTORY

Version	Description of changes	Date
Revision 1	Updated layout and content	2022/04

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

