

## Specification

Fluid medium used for determining the oxidative and/or fermentative metabolism of Gram negative bacilli (Enterobacteriaceae).

## Presentation

20 Tubes  
Tube 16 x 113 mm  
with: 10 ± 0.3 ml

### Packaging Details

1 box with 20 tubes, 16x113 mm glass tubes, ink  
labelled and metal-Non injectable cap.

**Shelf Life**      12 months  
**Storage**      8-25 °C

## Composition

Composition (g/l):
Casein peptone..... 2.00
Sodium chloride..... 5.00
D(+) Glucose..... 10.0
Dipotassium phosphate..... 0.20
Bromothymol Blue..... 0.08
Agar..... 2.50

## Description /Technique

### Description

Using this medium Hugh and Leifson were able to differentiate Gram negative bacteria into three categories: fermentative, oxidative and inactive.

### Technique

The inoculation technique is stab inoculation. One tube is covered with vaseline® layer to induce an anaerobic environment that forces the strain to carry out fermentation.

Fermentative organisms produce a large amount of acid in both the tubes, and this is indicated by the yellow colouration of the Bromothymol Blue indicator. Bacteria that utilise an oxidative metabolic pathway carry out this reaction only in the tube without the vaseline ®. Inactive strains do not use sugars and therefore do not induce any change in either tube.

In some instances a slight blue colouration, probably due to alkalinization by peptone degradation, can occur.

Note: pH decrease may appear over time without affecting its performance.

## Quality control

### Physical/Chemical control

Color : Green      pH: 7.1 ± 0.2 at 25°C

### Microbiological control

Inoculate by stabbing with and without vaseline

Analytical methodology according to ISO 11133:2014/A1:2018; A2:2020.

Aerobiosis. Incubation at 35 ± 2 °C. Reading at 24 hours.

Microbiological control according to ISO 11133:2014/A1:2018; A2:2020.

### Microorganism

*Ps. aeruginosa* ATCC® 27853, WDCM 00025

*Escherichia coli* ATCC® 25922, WDCM 00013

*Salmonella typhimurium* ATCC® 14028, WDCM 00031

*Salmonella enterica* ATCC® 13076, WDCM 00030

*Ps. fluorescens* ATCC® 13525, WDCM 00115

### Growth

Good growth- O/F: Pos./Neg.

Good growth- O/F: Pos./Pos. Yellow medium

Good growth- O/F: Pos./Pos. Yellow medium

Good growth- O/F: Pos./Pos. Yellow medium

Good growth- O/F: Pos./Neg.

### Sterility Control

Incubation 48 h at 30-35 °C and 48 h at 20-25 °C: NO GROWTH.

Check at 7 days after incubation in same conditions.

Revision date: 20/06/23

## Bibliography

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- ISO 11133:2014/ Adm 1:2018/ Adm 2:2020/ Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.

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