

## Specification

Selective supplement for the cultivation of Brucella in diverse clinical specimens, foods and other materials of sanitary interest.

## Presentation

10 Freeze dried vials  
Vial  
with:  $6 \pm 2$  ml

### Packaging Details

$22 \pm 0.25 \times 55 \pm 0.5$  mm glass vials, tag labelled, White plastic cap - 10 vials per box.

### Shelf Life

36 months

### Storage

2-8 °C

## Composition

Composition (g/vial):

Natamycin.....	0.0250
Nalidixic acid.....	0.0025
Bacitracin.....	12,500 IU
Vancomycin.....	0.0100
Polymixin B sulphate.....	2,500 IU
Nystatin.....	50,000 IU

**Note:** Each vial is sufficient to supplement 500 ml of Brucella Agar

Reconstitute the original freeze-dried vial by adding

1:1 solution, methanol:

Sterile Distilled Water.....5 ml

## Description /Technique

### Description:

Brucella Medium Base (Cat. 1374) is prepared according to the formula described by Jones and Brinley Morgan for the cultivation and isolation of Brucella, including fastidious types. It is a medium rich in nutritive elements and growth factors that make it adequate for the growth and isolation of Brucella spp.

Beef extract and peptone provide nitrogen, vitamins, minerals and amino acids essential for growth. Glucose is the fermentable carbohydrate providing carbon and energy. Sodium chloride supplies essential electrolytes for transport and osmotic balance. Bacteriological agar is the solidifying agent. The addition of the supplement enhances the medium's selectivity for the growth of Brucella.

Brucella species are level 3 pathogens and cause brucellosis, a zoonotic disease. It is usually transmitted through milk, dairy products, meat and direct contact with infected animals. It is widely used for the isolation of Brucella in highly contaminated materials, food materials and clinical samples.

### Technique:

Aseptically reconstitute 1 vial with 10 ml of a solution 1:1 of ethanol/sterile distilled water. Incubate at 37 °C for 10-15 minutes. Mix until completely dissolved and aseptically add to 500 ml Brucella Medium Base (cat. 1374) cooled to 50 °C and if desired, add 5-10% of inactivated horse serum and 1-5% of sterile dextrose solution. Mix well and distribute into sterile containers.

### Instructions for use:

#### Streak plate method:

- In a Petri dish, add 12-15 ml of molten agar and let it solidify.
- Inoculate 10 µl of the initial suspension and/or diluted sample.
- Extend the inoculum with a sterile loop on the agar surface.
- Incubate the plates in an inverted position at a temperature of  $35 \pm 2$  °C in an atmosphere of 5-10% CO<sub>2</sub> and observe after 72 hours.

**Quality control****Physical/Chemical control**

Color: yellow

pH: at 25°C

**Microbiological control**

Reconstitute 1 vial as indicated in COMPOSITION; shake and dissolve completely

Add 1 vial to 500 ml of medium base. DO NOT HEAT once supplemented.

Distribute the complete medium, cooled to 50 °C, into 90 mm plates

Aerobiosis. Incubation at 35 ± 2 °C, reading after 48-72 hours

**Microorganism***Stph. aureus* ATCC® 25923, WDCM 00034*Escherichia coli* ATCC® 25922, WDCM 00013**Growth**

Inhibited

Inhibited

**Sterility Control**

Add 5ml of the sample to 100 ml of TSB and to 100 ml Thioglycollate.

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

Check at 7 days after incubation in same conditions.

**Bibliography**

Kzudas and Mors, J.Bact. 66:502. 1953 Rennoux G. Ann. Inst. Pasteur, 87:325. 1954 Standard Methods for Examination of Dairy Products. 10 th Ed. APHA, Inc. New York 1960 Smith Louis Ds.

The pathogenic anaerobic Bacteria. C. Thomas Pub. Springfield, Il, 1975.