



# Pyrazinamidase Agar

### **Intended Use**

Recommended for identification of Yersinia species in accordance with FDA BAM, 1998.

Ingredients	Gms / Litre
Tryptone	11.250
Soya peptone	3.750
Sodium chloride	3.750
Yeast extract	3.000
Pyrazine-carboxamide	1.000
Agar	11.250
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\*\*Formula adjusted, standardized to suit performance parameters

## Directions

Suspend 34.00 grams in 1000 ml 0.2 M Tris-maleate, pH 6.0. Heat to boiling to dissolve the medium completely. Dispense 5ml amount in 16 x 125 mm tubes. Sterilize by autoclaving at 15 lbs pressure ( $121^{\circ}$ C) for 15 min. After sterilization, cool the tubes in slanted position.

# **Principle And Interpretation**

*Yersinia enterocolitica* is a ubiquitous organism, isolated from soil, water, animals, and a variety of foods (7). They comprise a biochemically heterogeneous group that can grow even at refrigerated temperatures. The association of human illness with consumption of *Y.enterocolitica* - contaminated food, animal wastes, and unchlorinated water is well documented (1). Pyrazine

Amidase Agar(M1880) is used for the identification of Yersinia sp. in accordance with FDA BAM, 1998 (4).

Pyrazinamidase activity distinguish potential pathogenic from nonpathogenic strains of *Y. enterocolitica* in epidemiological surveillance programs. Fully grown culture, on pyrazinamidase agar slants at RT is flooded with 1 ml of 1% freshly prepared ferrous ammonium sulphate over slant. Development of pink color within 15 min is positive test, indicating presence of pyrazinoic acid formed by pyrazinamidase enzyme.

Tryptone, soya peptone and yeast extract provides nitrogenous, carbonaceous compounds, long chain amino acids, vitamins and other essential nutrients. Sodium chloride maintains the osmotic balance of the medium. Pyrazine-carboxamide acts as substrate to detect Pyrazinamidase activity.

# Type of specimen

Food and dairy samples - food, animal wastes; Water samples - unchlorinated water

# **Specimen Collection and Handling**

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (2,8,9). For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(3). After use, contaminated materials must be sterilized by autoclaving before discarding.

### **Warning and Precautions**

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

# Limitations

- 1. Some species may show poor growth due to nutritional variations.
- 2. Further biochemical and serological tests must be carried out for complete identification.

**M1880** 

#### **Performance and Evaluation**

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

## **Quality Control**

#### Appearance

Cream to yellow homogeneous free flowing powder

#### Gelling

Firm, comparable with 1.13% Agar gel.

#### Colour and Clarity of prepared medium

Light yellow coloured clear to slightly opalescent gel forms in tubes as slants.

#### Cultural Response

Cultural characteristics observed after an incubation at 25-30°C for 48 hours. \*-After incubation flood 1 ml of 1% freshly prepared ferrous ammonium sulphate solution over the slant.

Organism	Growth	Inoculum (CFU)	Pyrazinamidase Test*
Yersinia enterocolitica ATCC 27729	good-luxuriant	50-100	Variable Positive (development of pink colour within 15 mins)

### **Storage and Shelf Life**

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use.

Product performance is best if used within stated expiry period.

### Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).

#### Reference

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