

Standard or the unknown and total volume 10 ml per tube is adjusted by addition of distilled water. Sterilize by autoclaving at 15lbs pressure (121°C) for 10 minutes.

Principle And Interpretation

Pediococcus acidilactici is gram positive organism and used as a probiotic. Cystine Assay Medium contains all the essential growth factors required for growth of *Pediococcus acidilactici* ATCC 8042 except L-Cystine. The addition of the amino acid in increasing concentrations gives growth response are prepared for use in the microbiological assay.

Three types of media used for the microbiological assay of amino acids are the maintenance media used for carrying the stock culture, the inoculum media for preparation of the inoculum and the assay media for quantitation of the amino acid (L-Cystine) under test.

Cystine Assay Medium is prepared as per the formulation of Steel et.al (1). It contains all other vitamins and nutrients essential for the growth of *Pediococcus acidilactici* ATCC 8042 except L-cystine (amino acid under study).

Stock culture of *Pediococcus acidilactici* ATCC 8042 is prepared by stab inoculation into tubes of Lactobacilli Agar AOAC. The cultures are incubated at 35-37°C for 24 hours and stock cultures are maintained at 2-8°C. The inoculum is prepared by subculturing in 10 ml Lactobacilli Broth AOAC. Incubate at 35-37°C for 16-24 hours. After incubation, centrifuge the cells under aseptic conditions, decant the liquid supernatant. Wash the cells thrice with sterile 10 ml of sterile 0.85% NaCl solution. Then resuspend in 10 ml 0.85% NaCl solution. Dilute the solution as per use. The growth response obtained is turbidometrically or acidimetrically measured.

A standard curve is plotted with absorbance as a function of the L-cystine concentration. The concentration of L-cystine in the test sample is calculated based on the interpretation of the standard curve.

Extreme care should be taken to avoid contamination of media or glassware used for the assay. Detergent-free clean glassware

Pure Isolates

Pure isolates.

Specimen Collection and Handling

For pure isolate 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.

Limitations:

For pure isolate samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards. (3)

Limitations:

A4

Quality Control

Appearance

Off-white to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light amber coloured, clear solution, which may contain a slight precipitate.

Reaction

Reaction of 10.5% w/v aqueous solution at 25°C. pH : 6.7±0.2

pH

6.50-6.90

Cultural Response

M1936: Microbiological Assay of Cystine was carried out using *Pediococcus acidilactici* ATCC 8042 after an incubation at 35-37°C for 16-20 hours .

Organism

Growth

Cultural Response

Pediococcus acidilactici
ATCC 8042

Good growth is obtained. Gradual increase in growth with increasing conc.of standard L- Cystine 0, 5, 10, 15, 20, 25 mcg per assay tube was recorded as equivalent increase in absorbance at 660 nm.

Storage and Shelf Life

Store below 8°C, preferably in dessicators and use freshly prepared medium. Use before expiry date on the label.

Reference

1. Steel,Sauberlich, Reynolds and Baumann 1949, J.Biol. Chem. 177:533

Revision : 0/ 2014

Disclaimer :

User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related HiMedia™ publications. The information contained in this publication is based on our research and development work and is to the best of our knowledge true and accurate. HiMedia™ Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are not intended for human or animal or therapeutic use but for laboratory,diagnostic, research or further manufacturing use only, unless otherwise specified. Statements contained herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.