Pantothenate Assay Medium, AOAC

M1281

Recommended by AOAC for microbiological assay of pantothenic acid or its salts using *Lactobacillus plantarum* ATCC 8014 as the test organism.

Composition**

Ingredients	Gms / Litre
Casein acid hydrolysate	10.000
Dextrose	40.000
Sodium acetate	20.000
Dipotassium phosphate	1.000
Monopotassium phosphate	1.000
L-Cystine	0.400
L-Tryptophan	0.100
Magnesium sulphate	0.400
Sodium chloride	0.020
Ferrous sulphate	0.020
Manganese sulphate	0.020
Adenine sulphate	0.020
Guanine hydrochloride	0.020
Uracil	0.020
Riboflavin (Vitamin B2)	0.0004
Thiamine hydrochloride	0.0002
Biotin	0.0000008
p-Amino benzoic acid (PABA)	0.0002
Nicotinic acid	0.001
Pyridoxine hydrochloride	0.0008
Sorbitan monooleate complex	0.100
Final pH (at 25°C)	6.7±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 7.3 grams in 100 ml distilled water. Heat to boiling to dissolve the medium completely. Mix well to distribute the slight precipitate evenly. Dispense in 5 ml amounts to each assay tube in increasing amounts of the standard or the unknown. Adjust the volume of each tube to 10 ml with distilled water. Sterilize by autoclaving at 15 lbs pressure (121°C) for 10 minutes. Cool the medium immediately. Generally satisfactory results are obtained with Pantothenic acid at levels of 0.0, 0.005, 0.01, 0.015, 0.02 and 0.025 microgram per assay tube (10ml).

Principle And Interpretation

Pantothenate Assay Medium is a dehydrated medium free from pantothenic acid or pantothenate but containing all other nutrients and vitamins essential for the cultivation of *L. plantarum* ATCC 8014 as recommended by AOAC (1). To obtain a standard curve pantothenate is added in particular increasing concentrations giving a growth response that can be turbidimetrically measured. A standard curve is drawn on graph paper the in the following way. Plot the absorbance as a function of the pantothenate concentration and the best curve is drawn through the points. The concentration of pantothenate is calculated by interpretation on the standard curve.

Quality Control

Appearance

Off-white to light yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear solution, which may have a slight precipitate.

Reaction
Reaction of 7.3% w/v aqueous solution at 25°C. pH : 6.7±0.2

pH
6.50-6.90

Cultural Response
Microbiological Assay of Pantothenate is carried out by using L.plantarum ATCC 8014 after an incubation at 35-37°C for 18-24 hours.

Growth
Good growth is obtained. Gradual increase in growth with increasing concentration of pantothenate standard levels of 0.0, 0.005, 0.01, 0.015, 0.02, 0.025 mcg per assay tube is recorded as equivalent increase in absorbance at 620 nm.

Storage and Shelf Life
Store below 8°C and use freshly prepared medium. Use before expiry date on the label.

Reference
1. Williams, (Ed.), 2005, Official Methods of Analysis of the Association of Official Analytical Chemists, 19th Ed., AOAC, Washington, D.C.

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