

Niacin Assay Medium is devoid of nicotinic acid but contains all other nutrients and vitamins essential for the cultivation of *L. plantarum* ATCC 8014. Addition of Niacin (Nicotinic acid) in specified increasing concentrations gives a corresponding growth response that can be measured turbidimetrically or titrimetrically.

Stock cultures of *L. plantarum* ATCC 8014 are maintained on Lactobacilli Agar, AOAC (M366). Culture for assay is obtained by inoculating Lactobacilli Agar, AOAC (M366) and incubating at 35-37°C for 24-48 hours. These cultures are then inoculated into Lactobacilli Broth, AOAC (M367), to prepare the inoculum. Following an incubation at 35-37°C for 18-24 hours, the cells are centrifuged and washed thrice with 0.85% saline. The appropriate dilution of cells so obtained is used to inoculate tubes of Niacin Assay Medium (M040), containing increasing concentrations of Niacin. Using standard Niacin concentration, a standard curve is obtained. This standard is used to extrapolate the unknown niacin concentration. For detailed procedure, refer standard procedures (4, 5).

Extreme care must be taken to avoid contamination of media or glassware used for microbiological assay procedures. Detergent-free clean glassware should be used. Even small amount of contamination by foreign material can lead to erroneous results.

Type of specimen

Pure isolates.

Specimen Collection and Handling

Stock cultures of *L. plantarum* ATCC 8014 are maintained on Lactobacilli Agar, AOAC (M366). Culture for assay is obtained by inoculating Lactobacilli Agar, AOAC (M366) and incubating at 35-37°C for 24-48 hours. These cultures are then inoculated into Lactobacilli Broth, AOAC (M367), to prepare the inoculum. Following an incubation at 35-37°C for 18-24 hours, the cells are centrifuged and washed thrice with 0.85% saline. The appropriate dilution of cells so obtained is used to inoculate tubes of Niacin Assay Medium (M040), containing increasing concentrations of Niacin. Using standard Niacin concentration, a standard curve is obtained. This standard is used to extrapolate the unknown niacin concentration. For detailed procedure, refer standard procedures (4, 5).

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. Further biochemical testing is required for complete identification.

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

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 7.51% w/v aqueous solution at 25°C. pH : 6.8±0.2

pH

6.60-7.00

Cultural Response

Microbiological assay of Niacin is carried out using *L. plantarum* ATCC 8014 after an incubation at 35-37°C for 16-18 hrs.

Organism

Growth

Growth

Good growth is obtained.

Gradual increase in growth with increasing conc. of Std Niacin-0.0,0.025,0.05,0.075,0.1,0.125, 0.15,0.2 & 0.25mcg per assay tube is recorded asequential increase in absorbance at 620nm.

Reference

1. Snell and Wright, 1941, J. Biol. Chem. 13:675.
2. Krehl, Strong and Elvehjem, 1943, Ind. & Eng. Chem., Ann. Ed. 15:471.
3. Barton-Wright, 1944, J. Biochem., 38:314.
4. Williams, (Ed.), 2005, Official Methods of Analysis of the Association of Official Analytical Chemists, 19th Ed., AOAC, Washington, D.C.
5. The United States Pharmacopoeia, 2006, USP29/NF24, The United States Pharmacopoeial Convention, Rockville, MD.

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