



EnzyChrom™ NAD/NADH Assay Kit

Pyridine nucleotides play an important role in metabolism and, thus, there is continual interest in monitoring their concentration levels. Quantitative determination of NAD⁺/NADH has applications in research pertaining to energy transformation and redox state of cells or tissue.

Simple, direct and automation-ready procedures for measuring NAD⁺/NADH concentration are very desirable. BioAssay Systems' EnzyChrom™ NAD⁺/NADH assay kit is based on an alcohol dehydrogenase cycling reaction, in which a tetrazolium dye (MTT) is reduced by NADH in the presence of phenazine methosulfate (PMS). The intensity of the reduced product color, measured at 565 nm, is proportionate to the NAD⁺/NADH concentration in the sample. Our assay is a convenient method to measure NAD, NADH and their ratio.

KEY FEATURES

Sensitive and accurate. Detection limit 0.2 μM, linearity up to 10 μM NAD⁺/NADH in 96-well plate assay.

Convenient. The procedure involves adding a single working reagent, and reading the optical density at time zero and 15 min at room temperature. No 37°C heater is required.

High-throughput. Can be readily automated as a high-throughput 96-well plate assay for thousands of samples per day.

APPLICATIONS

Direct Assays: NAD⁺/NADH concentrations and ratios in cell or tissue extracts.

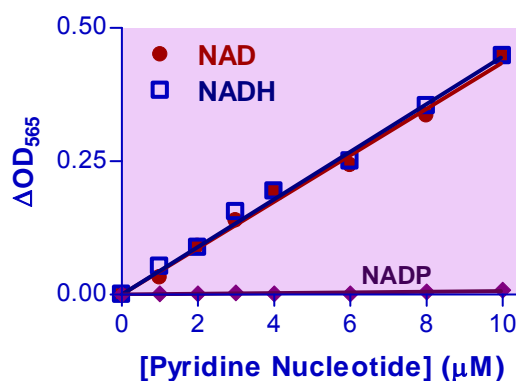
PRODUCT INFORMATION:

EnzyChrom™ NAD/NADH Assay Kit

ECNP-100

Each kit is sufficient for 100 assays in 96-well plate. Kit includes:

- 1 x 10 mL Assay Buffer
- 1 x 2 mL Ethanol (1vol%)
- 1 x 2 mL PMS Solution
- 1 x 2 mL MTT Solution
- 1 x 120 μL Enzyme
- 1 x 0.5 mL NAD⁺ Standard
- 1 x 0.5 mL NADH Standard
- 1 x 12 mL NAD⁺ Extraction Buffer
- 1 x 12 mL NADH Extraction Buffer



Standard Curves in 96-well plate assay

REFERENCES:

- [1]. Zhao, Z, Hu, X and Ross CW (1987). Comparison of Tissue Preparation Methods for Assay of Nicotinamide Coenzymes. *Plant Physiol.* 84: 987-988.
- [2]. Matsumura, H. and Miyachi S (1980) Cycling assay for nicotinamide adenine dinucleotides. *Methods Enzymol.* 69: 465-470.
- [3]. Vilcheze, C et al. (2005). Altered NADH/NAD⁺ Ratio Mediates Coresistance to Isoniazid and Ethionamide in Mycobacteria. *Antimicrobial Agents and Chemotherapy.* 49(2): 708-720.