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## Product Information

### **β-Nicotinamide adenine dinucleotide hydrate Cell Culture Tested**

Product Number **N3014**  
Storage Temperature -20 °C

#### **Product Description**

Molecular Formula:  $C_{21}H_{27}N_7O_{14}P_2 \cdot xH_2O$   
Molecular Weight: 663.4 (anhydrous basis)  
CAS Number: 53-84-9  
Extinction Coefficient:  $E^{mM} = 18.0$  (260 nm, pH 7)<sup>1</sup>  
Synonym: β-NAD, DPN

This product is cell culture tested (7 mg/L) and is appropriate for use in cell culture applications.

β-NAD, a pyridine nucleotide and biologically active form of nicotinic acid, is a coenzyme necessary for the catalytic reaction of certain enzymes. β-NAD is a carrier for hydride ion, forming β-NADH. Hydride ion is enzymatically removed from a substrate molecule by the action of dehydrogenases such as malic dehydrogenase and lactic dehydrogenase. Such enzymes catalyze the reversible transfer of a hydride ion from malate or lactate to β-NAD to form the reduced product, β-NADH. Unlike β-NAD which has no absorbance at 340 nm, β-NADH absorbs at 340 nm ( $E^{mM} = 6.22$ ). The increase in absorbance at 340 nm with the formation of β-NADH is the basis for measurement of activity of many enzymes.<sup>2,3</sup>

Many metabolites and enzymes of biological interest are present in tissues at low concentrations. With the use of β-NAD as a catalyst intermediate and several enzymes in a multistep system, known as enzyme cycling, much greater sensitivity for detection of these components is achieved. The reduced form, β-NADH, is fluorescent whereas β-NAD is not. This difference in fluorescence provides a sensitive fluorescent measurement of the oxidized or reduced pyridine nucleotides at concentrations down to  $10^{-7}$  M.<sup>3,4</sup>

#### **Precautions and Disclaimer**

For Laboratory Use Only. Not for drug, household or other uses.

#### **Preparation Instructions**

β-NAD is soluble in water (50 mg/ml), yielding a clear solution (gentle heating and sonication may be necessary). Aqueous solutions can also be prepared by adjusting the pH to 7 with either sodium hydroxide or sodium bicarbonate buffer.

#### **Storage/Stability**

β-NAD is very hygroscopic and should be stored desiccated.<sup>5</sup> Aqueous solutions between pH 2 - 6, stored as single-use aliquots at -70 °C, are stable for at least 6 months. Neutral or slightly acidic solutions are stable at 0 °C for at least 2 weeks. Solutions are rapidly degraded upon heating and are very labile in alkaline solutions, especially in the presence of phosphate, maleate, or carbonate. The rates of degradation of solutions at different pH and temperature conditions have been reported. Solutions are also sensitive to light.<sup>6,7</sup>

#### **References**

1. Specifications and Criteria for Biochemical Compounds, 3rd ed., National Academy of Sciences (Washington, DC: 1972), p. 87.
2. Methods of Enzymatic Analysis, Volume 4, Hans Ulrich Bergmeyer, Academic Press, Inc., (New York, NY:1974), 2066-2072.
3. Enzymatic Analysis, A Practical Guide, Passonneau, J.V., and Lowry, O.H., The Humana Press, Inc., (Totowa, NJ:1993), 3-4.
4. Enzymatic Analysis, A Practical Guide, Passonneau, J.V., and Lowry, O.H., The Humana Press, Inc., (Totowa, NJ:1993), 85-110.

5. The Merck Index, 13th, Entry# 6370.
6. Data for Biochemical Research, 3rd ed., Dawson, R. M. C., et al., Oxford University Press (New York, NY: 1986), p 130-131.
7. Lowry, O. H., et al., The stability of pyridine nucleotides. J. Biol. Chem., **236**, 2756 (1961).

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