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# **ProductInformation**

# L-Leucine Dehydrogenase from Bacillus cereus

Product Number L 5135 Storage Temperature -0 °C

## **Product Description**

Enzyme Commission (EC) Number: 1.4.1.9

CAS Number: 9082-71-7 Molecular Weight: 245 kDa<sup>1</sup>

Extinction coefficient:  $E^{1\%} = 9.18 (280 \text{ nm})^{1}$ 

Leucine dehydrogenase is a hexamer consisting of 6 identical subunits. The enzyme catalyzes the following reaction:

L-Leucine +  $H_2O$  +  $\beta$ -NAD  $\rightarrow \alpha$ -Ketoisocaproate +  $NH_3$  +  $\beta$ -NADH

The  $K_m$  values for this reaction are: L-leucine (1mM),  $\beta$ -NAD (0.39 mM),  $\beta$ -NADH (0.035 mM),  $\alpha$ -ketoisocaproate (0.31 mM), and ammonia (200 mM). Isoleucine, valine, norvaline, and norleucine may also be utilized as substrates. The following  $K_m$  values and activities have been reported<sup>1</sup>:

Substrate	Relative Activity	$K_{m}$ (mM)
L-Leucine	100	1.0
L-Valine	74	1.7
L-Isoleucine	58	1.8
L-Norleucine	10	6.3

Leucine dehydrogenase is a sulfhydryl containing enzyme which is strongly inhibited by p-chloromercuribenzoate and HgCl<sub>2</sub>. The enzyme is also competitively inhibited by D-enantiomers of the substrate amino acids and by pyridoxal phosphate.<sup>1,2</sup>

Leucine dehydrogenase is useful for the enzymatic detrmination of L-leucine. Leucine dehydrogenanase can be used in conjunction with urease to quantitate urea in serum and urine,<sup>3</sup> and can be used in an enzymatic oxidation system for the synthesis of enantiomerically pure D-tert-leucine.<sup>4</sup>

#### **Precautions and Disclaimer**

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

## **Preparation Instructions**

This enzyme is soluble in 25 mM potasssium phosphate buffer, pH 7.2 (0.2 mg/ml), yielding a clear, colorless solution.

### Storage/Stability

The enzyme is stable to 60 °C when heated for 5 minutes in 10 mM potassium phosphate buffer (pH 7.2) and the enzyme was very stable at pH 6.5 to 9.0 when incubated at 50 °C for 5 minutes. 1

### References

- Ohshima, T., et al., Properties of crystalline leucine dehydrogenase from *Bacillus sphaericus*.
   J. Biol. Chem., 253, 5719-5725 (1978).
- Matsuyama, T., et al., Leucine dehydrogenase from *Bacillus stearothermophilus*: identification of active lysine by modification with pyridoxal phosphate. J. Biochem., 112, 258-265 (1992).

- 3. Morishita, Y., et al., Kinetic assay of serum and urine for urea with use of urease and leucine dehydrogenase. Clin. Chem., **43**, 1932-1936 (1997).
- 4. Hummel, W., et al., An efficient and selective enzymatic oxidation system for the synthesis of enantiomerically pure D-tert-leucine. Org. Lett., **5(20)**, 3649-3650 (2003).

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