Tribioscience

Lactate Dehydrogenase, Enzyme Activity

Catalog	Unit
TBP0074-10KU	10000 U
TBP0074-50KU	50000 U

Product Details

Form: Freeze-dried powder

Solubility: Soluble in distilled water or dilute buffer

Stability: Store at -20° C (-4° F)

Activity: 350 U/mg protein

Protein: 80-90%

Catalog No.: 092A0350

Unit Definition

The amount of enzyme which will reduce one micromole of pyruvate to L-lactate per minute at 25°C in 0.1 M phosphate buffer at pH 7.0.

Assay Method

The rate of decrease in the absorbancy at 340 nm, resulting from the oxidation of NADH, is a measure of LDH activity.

Applications

Lactate dehydrogenase (LDH) (EC 1.1.1.27) catalyzes the following reaction:

$$\text{Pyruvate} + \text{NADH} \xrightarrow{\text{LDH}} \text{Lactate} + \text{NAD+} + \text{H+}$$

LDH is a metabolic enzyme which is widely distributed in nature. Mammalian LDH exists as five tetrameric isozymes composed of combinations of two different subunits. The isozymes differ in physical, immunological and catalytic properties. LDH has a molecular weight of about 140,000.

LDH is of clinical significance in that the serum level of certain isozymes suggests pathological changes in particular tissues. It is also used extensively for the determination of lactate and in coupled systems for detection of biological metabolites.

Reagents

1. 0.1 M Sodium phosphate buffer, pH 7.0.
2. Sodium pyruvate solution, (2.5 mg/ml) in distilled water.
3. NADH solution (5 mg/ml). Dissolve 5 mg NADH, sodium salt in 1.0 ml distilled water. Always prepare fresh.
4. 1% Bovine serum albumin (BSA) solution. Dissolve 1.0 g BSA in 100 ml distilled water. Albumin should be of highest purity.
5. LDH solution (0.5-1.0 U/ml) - Dilute 0.1 ml enzyme suspension to 5 ml with cold 1% BSA solution. Use an aliquot from this stock enzyme solution and dilute to a final concentration of 0.5-1.0 U/ml with cold 1% BSA. This solution must be used as soon as it is prepared and must be made fresh for each run.

Calculation

$$\text{Activity (U/mg)} = \frac{(\Delta E_{304nm/min})(\text{Total Vol.})(\text{Enz. Diln.})}{(6.22)(\text{Enz. Vol.})(\text{mg Enz./ml})}$$

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