



ISO 13485:2016 ISO 9001:2015

Ver.240817

Lactate Dehydrogenase (LDH) Assay kit

BC6603-01(50 Tests)

FOR RESEARCH USE ONLY, DO NOT USE IT IN CLINICAL DIAGNOSIS

Product Description

Lactate Dehydrogenase (LDH or LD) is an enzyme found in nearly all living cells (animals, plants and prokaryotes). LDH catalyzes the conversion of lactate to pyruvic acid and back, as it converts NAD^+ to NADH and back. A dehydrogenase is an enzyme that transfers a hydride from one molecule to another.

Kinetic determination of lactate dehydrogenase according to the following reaction.



Kit components

Reagent / Component	Volume	Storage
Extraction Reagent	1 × 60 mL	2-8°C
LDH (S.L) R1	2 × 24 mL	2-8°C
LDH (S.L) R2	2 × 6 mL	2-8 °C

Open Vial Stability

Once opened, the reagent is stable up to 4 weeks if contamination is avoided.

Reagent Deterioration

Turbidity or precipitation in any kit component indicates deterioration and the component must be discarded.

Reagent Preparation

Mix 4 volumes of Reagent 1 (R1) with 1 volume of Reagent 2 (R2)

The working reagent is stable for 21 days at 2-8°C.

NOTE: Discard the working reagent if the blank absorbance is less than 1.0 at 340 nm.

Precaution

- To avoid contamination, use clean laboratory wares. Use clean, dry disposable pipette tips for dispensing. Close reagent bottles immediately after use.
- Avoid direct exposure of reagent to light.
- Do not blow into the reagent bottles.

Operation Procedures

Sample Preparation

1. Bacteria or cells

Harvest the cells and wash twice with PBS. Ideal to use 5 million cells for the assay. Add 1mL Extraction Reagent to 5 million cells and ultrasonicate (200W, work time 3 second / interval 10 second repeat for 30 times) for complete lysis. Perform ultrasonication while keeping the cells in ice bath. Centrifuge at 8000 rpm, 4°C for 10 minutes and collect the supernatant. The supernatant should be kept on ice.

Note: Ideal proportion of Cells / Bacteria to Extraction Reagent is 1:5 - 10.

2. Tissue

Prepare 10% tissue homogenate by adding 1mL Extraction Reagent to 0.1g tissue. Grind completely to make a homogenate. Centrifuge at 8000 rpm, 4°C for 10 minutes and collect the supernatant.

3. Serum or Plasma

Directly use for the assay.

Interferences

No interference for

Bilirubin up to 20 mg/dL ,

Note: Haemolysed sera should not be used since significant haemolysis may increase LDH concentration because of high levels of LDH in erythrocytes.

Materials Required but Not Provided

Homogenizer, centrifuge, water bath, micro pipettes, spectrophotometer, 1ml cuvette, ice and distilled water

Unit Conversion

Traditional Unit	SI Unit	Conversion from Traditional to SI
U/L	uKat/L	$\times 0.017$

Procedure Notes

Switch on the spectrophotometer 30 minutes prior to reading, adjust wavelength to 340 nm, set zero with distilled water.

Reagent	Volume
Working Reagent	1000 μ L
Sample	10 μ L
Mix and Incubate at 37°C for 1 minute. Measure the change in absorbance per minute (Δ OD/minutes) during 3 minutes.	

Calculation

Plasma or Serum

$$\text{LDH activity (U/L)} = (\Delta \text{ OD/minutes}) \times 16030$$

Protein Concentration

$$\text{LDH activity (U/L)} = (\Delta \text{ OD/minutes}) \times 16030 \div \text{Cpr}$$

Tissue Weight

$$\text{LDH activity (U/L)} = (\Delta \text{ OD/minutes}) \times 16030 \div W$$

Cpr: Sample protein concentration, mg/ mL

W : Sample weight, g

Performance

Linearity

This reagent is linear up to 2400 U/L

If the concentration is greater than linearity (2400 U/L) dilute the sample with normal saline and repeat the assay. Multiply the result with dilution factor.

Sensitivity

Lower detection limit is 7 U/L.