



SWEMED DIAGNOSTICS

LDH REAGENT KIT

(Mod. IFCC Method)

For the determination of Lactate Dehydrogenase activity in serum

For In vitro diagnostics only

Ref no.

LDH 25

LDH 110

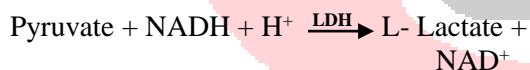
LDH 125

Summary

LDH is found in many body tissues particularly liver, heart, skeletal muscle, kidney & RBC's. LDH found in the form of isoenzymes based on their electrophoretic mobility with each isoenzyme being primarily from different organs. Increased level are found in the myocardial infarction, pulmonary diseases, hepatic diseases, hemolytic anemias, renal diseases and muscular dystrophy.

Principle

LDH catalyzes the reduction of pyruvate with NADH to form NAD. The rate of oxidation of NADH to NAD measured as a decrease in absorbance which is proportional to the LDH activity in the sample.



Kit Contents

Kit size	25ml	110ml	125ml
Ref no.	LDH25	LDH110	LDH125
LDH – R1	1	2	2
LDH– R2	1	2	1
IFU	1	1	1

Material required not provided

Test tubes, yellow tips, blue tips

General laboratory equipment

Storage & Stability of the Reagents

1. All the components of the kit are stable until the expiration date on the label when stored tightly closed at 2-8°C, protected from light and contaminations prevented during their use.
2. Do not freeze the reagents.
3. Ensure the reagents & specimens are brought to Room Temperature.
4. Ensure the reagents shelf life is valid.

5. Do not use haemolysed & lipemic serum.

Reagent preparation

Mix, 4 parts of reagent 1 & 1 part of reagent 2 = working reagent.

Working solution is stable for 1 week at 2 -8°C.

Alternatively 0.8ml of R1 & 0.2ml R2 may also be used instead of 1ml of the working reagent directly during the assay.

The working reagent should have an absorbance above 1.0 against distilled water at 340nm.

Discard the reagent if the absorbance is below 1.0

Reagent composition

Reagent 1	Tris buffer Ph 7.0	6g/L
	EDTA	1.5g/L
	Sodium pyruvate	0.2g/L
	L- alanine	44.5g/L
Reagent 2	KH ₂ PO ₄	0.9g/L
	K ₂ HPO ₄	0.8g/L

Specimen

Serum, heparin-plasma

Specimen collection

- Use non-haemolyzed serum.
- Heparin is the only acceptable anticoagulant. Sodium citrate and EDTA have an inhibitor effect and must not be used.
- RBC's have a very high LDH content & hence hemolysed samples should not be used.
- The biological half-life of LDH in serum is 10 - 54 hours.

Storage & Stability of the Specimen

Serum Stability: 1-3days at 2– 8°C

Freezing inactivates the liver isoenzymes.

Warning & Precautions

1. Do not ingest or inhale.
2. In case of contact with eyes or skin; rinse immediately with plenty of soap and water. In case of severe injuries; seek medical advice immediately.
3. Both reagents (R1) and (R2) contain sodium azide which may react with copper or lead plumbing.
4. Keep out of reach children. Take off immediately all contaminated clothing.
5. Wear suitable gloves and eye /face protection.
6. Always use safety pipettes to pull the reagents into a pipette.



SWEMED DIAGNOSTICS

- Reagents may contain some non-reactive and preservative components. It is suggested to handle carefully, avoid direct contact with skin and do not swallow.
- Perform the test according to the current "Good Laboratory Practice" (GLP) guidelines.
- The reagents contain sodium azide (0.95g/L) as preservative. Do not swallow. Avoid contact with skin and mucous membrane.

Assay procedure

Wave length	: 340 nm	
Temperature	: 37° c	
Light path	: 10 mm	
Pipette into cuvettes	Macro	Semi-Micro
Reagent (R1+R2)	800µl +200µl	400µl+100µl
Sample	25µl	12.5µl

Mix well & read the initial absorbance A_0 after 1 minute and repeat the absorbance reading after every 1, 2, & 3 minutes. Calculate the mean absorbance change per minute ($\Delta A/\text{min}$).

Calculation

$$\text{LDH (U/L)} = \Delta A/\text{min} \times 8199$$

Units: One international unit (IU) is the amount of enzyme that transforms 1 µmol of substrate per minute, in standard conditions. The concentration is expressed in units per litre of sample (U/L).

Performance Characteristics

Measuring range

The reaction is linear up to LDH concentration of 1000 U/L; specimens showing higher concentration should be diluted 1+5 with physiological saline and repeat the assay (result×6).

Linearity

The linearity is 1000U/L

Reference range

Serum	230 -460U/L
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"It is recommended that each laboratory establish its own normal range representing its patient population."

Quick References

Parameter	LDH
Mode	Kinetic
Wavelength	340nm
Unit	IU/L
Temperature	37°C
Factor	8199
Reaction slope	Decreasing
Reagent vol.	1000µl
Sample volume	25µl
Reaction time	180sec
Delay time	60sec
Delta time	60sec
Blanking	Water blank
linearity	1000 U/L

REFERENCES:

- Henry, J.B., Clinical Diagnosis and Management by Laboratory Methods, W.B. Saunders and Company (1979).
- Buhl, S.N. et. al, Clin. Chem. 23:1289.
- Amador, E. et. al. Clin. Chem. 9: 391.

			
Instructions for use	Use by	Batch number	Manufacturer
			
Invitro Diagnostic Medical Device	Date of manufacturer	Temperature limit	Reference number

- Wroblewski, F., La Due, J.S., Proc. Soc. Exp. Biol Med. 90:210.
- Z. KLIN. CHEM. KLIN. BIOCHEM.