ANA BIO CLR LDH

(Kinetic Method)

Intended Use

LDH is a reagent set for determination of lactate dehydrogenase activity in serum and plasma based on Kinetic method.

Principle

Lactate dehydrogenase (LD or LDH) catalyzes the reduction of pyruvate by NADH to form lactate and NAD^+. The catalytic concentration is determined from the rate of decrease of NADH measured at 340 nm.

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

Reagents provided

1. Reagent R_1
2. Reagent R_2

Working reagent preparation

Prepare working solution by mixing Reagent R_1 and R_2 in the ratio 4:1 as per requirement.

Reagent storage and stability

The reagent kit should be stored at 2 - 8℃ and is stable till the expiry date indicated on the label.
R_1 and R_2 reagents are stable till expiry at 2 - 8℃.
The working solution ($4R_1 + 1R_2$) is stable for 30 days at 2 - 8℃.
DO NOT FREEZE THE REAGENT. Contamination of the reagents should be strictly avoided.

Specimen collection and preservation

Collect the specimen in a clean dry container. Although serum is preferred, plasma with Heparin or EDTA can be used. Hemolyzed samples should not be used since LDH activity in erythrocytes is 160 fold higher than in serum. The serum should be separated from the clot promptly. Samples should be assayed soon after collection. LDH is stable in serum or plasma for four days at 2 - 8℃. Do not freeze or expose the serum to high temperature as this may inactivate thermolabile LDH isoenzymes.

Assay guidelines for Analyzer

Reaction type	Kinetic
Reaction slope	Decreasing
Wavelength	340 nm
Flow cell temperature	37℃
Zero setting with	Distilled water
Delay time	60 seconds
Measuring time	90 seconds
Blank Absorbance limit	≥ 1.000
Sample Volume	20 µl (0.02ml)
Reagent Volume	1000 µl (1.0ml)
Factor	8109
Linearity	2000 IU/L

Assay guidelines for Manual procedure

Prewarm at 37℃ the required amount of working solution before use.

Dispense	Sample
Working solution	1000 µl (1.0 ml)
Sample solution	20 µl (0.02ml)

Mix thoroughly and transfer the assay mixture immediately to the thermostated cuvette and start the stop watch simultaneously. Record the first reading at 60^{th} second and subsequently three more readings with 30 seconds interval at 340 nm.

Calculation

Activity of LDH in IU/L = ΔAbs/min. x 8109

Normal Range

		25℃	30℃	37℃
IU/L	Adults	120-240	161-322	240-480
µkat/l	Adults	2.00-4.00	2.68-5.37	4.00-8.00

The following factors are used for conversion:
From 25℃ to 30℃: 1.34
From 25℃ to 37℃: 2.00
Note: Expected range varies from population to population and each laboratory should establish its own normal range.

Limitations

- Working reagent is considered unsatisfactory & should not be used if its absorbance is less than 1.000 at 340 nm against distilled water.
- If LDH activity is above 2000 IU/L then dilute the specimen suitably with normal saline. In such case the results obtained should be multiplied by the dilution factor to obtain correct LDH activity.

Quality Control

To ensure adequate quality control, it is recommended that each batch should include a normal and an abnormal commercial reference control serum. It should be realized that the use of quality control material checks both instrument and reagent functions together. Factors which might affect the performance of this test include proper instrument function, temperature control, cleanliness of glassware, and accuracy of pipetting.

References

1. Thomas L. Clinical laboratory Diagnostics. 1st ed.Frankfurt: TH-Books verlagsegellschaft; 1998:89-94.
2. NCCLS Document "Evaluation of precisión performance of clinical chemistry Devices",2nd ed. (1992).
3. Moss DW; Henderson AR. Clinical Enzymology In: Burtis CA,Ashwood ER, editors. Tietz Textbook of Clinical chemistry.3rd ed.Philadelphia. W.B.Saunders Company; 1999:617-721.

Symbols

IVD In Vitro Diagnostics. | Caution. | Keep away from sun light. | Date of Manufacture.

LOT Batch No. | Read Instructions. | Fragile. | Product Expiry Date.

CONT Content. | Storage Temperature. | Keep Dry. | Manufactured By.

REF Catalogue No.

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