

(KINETIC)

CLINICAL SIGNIFICANCE

Elevated levels are found in myocardial infarction, liver diseases, hemolytic anemias, pernicious anemia, leukemia & pulmonary diseases.

PRINCIPLE

Lactate is oxidised to pyruvate in the presence of NAD by the action of Lactate dehydrogenase. The rate of formation of NADH is measured at 340 nm & it is directly proportional to LDH activity.

REAGENTS COMPOSITION

Tris buffer	80 mMol/L
Sodium chloride	200 mMol/L
Lithium Lactate	65 mMol/L
NAD	5.25 mMol/L

REAGENT SUPPLIED

R 1 Buffer	3 x 9 mL
R 2 Enzyme	3 x 1 mL

Preparation of Working Reagent

Mix 9mL of R1 Buffer with 1mL of R2 Enzyme. A uniform solution may form after 10 minutes which is ready to use.

STORAGE AND STABILITY

Unopened reagents are stable till the expiry as mentioned on the label when stored at 2-8°C.

STABILITY OF WORKING RGT:

Working reagent is stable for two week at 2-8°C, When stored at room temp. & protected from light.

SAMPLE

Use fresh serum, or (heparinised or EDTA Na⁺) plasma which shows no sign of haemolysis. No prior-patient preparation is required for sample collection.

PROCEDURE FOR AUTOANALYSERS

Working Reagent	µL	500
Sample	µL	10

Mix well. Take one minute reading at 340 nM after a delay of 60 seconds at 37°C.

NOTE: Programme the analyser using system parameters. A specific programme data sheet may be provided for each analyser upon request. **Reagent, sample volume can be altered proportionately.**

SYSTEM PARAMETERS

Reaction	UV-Kinetic
Direction of Reaction	Increasing
Temperature	37°C
Wavelength	340 nM
Factor	8095
Absorbance Range	0-2°A
Cuvette Path Length	1 cm
Delay Time	60 Seconds
Interval	30 Seconds
No. of Readings	4
Linearity	800 U/L
Blank	DW

Reagent Volume	500 µL
Sample Volume	10 µL

RESULTS

Compute average rate of absorbance per minute x 8095.

EXPECTED VALUES

TEMPERATURE	37°C
Adults	70-280 U/L

As with all diagnostic methods, the final diagnosis should not be made on the result of a single test as well as laboratory diagnosis must be confirmed with clinical manifestations.

LIMITATIONS

If the test exceeds the linearity limit dilute the sample with 0.9% Sodium Chloride and rerun the test and multiply the result with dilution factor. This methodology highly depends on accuracy and precision of instrument and professionals.

This assay is linear up to 800 U/L.

QUALITY CONTROL

To ensure adequate quality control, each kit should be tested against a standard control sera. It should be realised that the use of quality control material checks both instrument and reagent function together. Factors which might affect the performance of this test include proper instrument function, temperature control, cleanliness of glasswares and accuracy of pipetting.

It is appropriate to establish each laboratory's accuracy constant and interpret values accordingly. Similarly, laboratory findings should be established by clinical manifestations.

WARNING

This reagent system is for *in vitro Diagnostic* use only.

This reagent system is containing preservatives and components that have not established for safety if contacted on broken skin or eye or taken orally. In case of such incidents wash off with plenty of water, or consult a physician.

BIBLIOGRAPHY

1. Jung K. Fechner C, Egger E, J. Cil Biochem (1976) 14, 53.
2. Thefeld W, et. al., Dtsch. Med. Wschr. (1974) 99, 343.