LDH (L ⟶ P)

ENZOPAK
Last update 04-2014

(UV-Kinetic)
Reagent kit for quantitative estimation of Lactate Dehydrogenase activity in serum or plasma.

- One of the most reliable LDH reagent in the market. Most of the Reagent kits have moisture (converting **β**-NADH to **α**-NADH) problems.
- Suitable pack sizes and corresponding suitable reconstituted stability.
- L ➔ P Kit as mono (1.1 ml) reagent.
- Most economical with no wastage of reagents.
- Necessary when CK-NAC/CK-MB is inconclusive due to macro CK-BB etc.
- More reliable reagent for CHD follow up after 4 to 6 days.

BACKGROUND & SYNOPSIS :
Lactate dehydrogenase is a cytoplasmic enzyme distributed very widely in the body. It is found in organs like heart, liver, kidney and skeletal muscle. Elevated LDH levels in serum are observed in several hemolytic, neoplastic, cardiac, skeletal muscle and renal diseases. These may also be found in destructive renal disease, progressive muscular dystrophy, megaloblastic anaemia, liver cirrhosis, hepatitis, hepatic metastasis, hepatoma and pulmonary embolism.
Lactate to pyruvate in the presence of lactate dehydrogenase is a reversible reaction. However the L ⟶ P reaction is favoured at a pH of 7.4 and this reaction utilises NAD as a coenzyme which is more stable compared to NADH.
ENZOPAK LDH L ⟶ P is formulated based on the procedure of Gay, Bowers and Mc Combs (1968)

PRINCIPLE :
LDH catalyzes the oxidation of lactate to pyruvate accompanied by the simultaneous reduction of NAD to NADH. LDH activity in serum is proportional to the increase in absorbance due to the reduction of NAD.

DIAGNOSTIC SIGNIFICANCE :
LDH is one of the five (i.e. CK/CPK, CK-MB, GOT/AST, LDH and HBDH) cardiac panel enzymes. Estimation of these enzymes is a powerful tool at the onset of CHD and equally useful in monitoring progress status of the patients.

REAGENT COMPOSITION :

ACTIVE INGREDIENTS	**Concentration**
Reagent-1	
• NAD	5 mmol/L
• Lactate	50 mmol/L
Reagent-2	
• Buffer	100 mmol/L

pH 9.3 ± 0.5 at 25⁰C
Also contains non-reactive fillers & stabilizers.

PRESENTATION :

	No. of Bottles/Blister
Store all reagents at 2-8⁰C	20 x 1.1 ml.
• 1 LDH (Coenzyme)	2 (10 Tablets)
• 2 LDH (Buffered Substrate)	2
Reconstitution Vial	1

An ISO 13485: 2012 & GMP Certified Company

PRECAUTION :
ENZOPAK LDH (L ➔ P) is for *IN-VITRO* diagnostic use only.
Reagent Contains Sodium Azide, DO NOT INGEST.

WORKING REAGENT PREPARATION :
For 20 x 1.1 ml :
Reconstitute one tablet of 1 LDH with 1.1 ml of 2 LDH. Mix gently to dissolve the contents. Use after 5 minutes.

REAGENT STORAGE AND STABILITY :
ENZOPAK LDH (L ➔ P) reagents are stable at 2- 8⁰ C until the expiry date stated on the label. Reconstituted reagent is stable at 2- 8⁰ C for 30 days.

SPECIMEN COLLECTION :
Fresh, clear, serum with no hemolysis is essential.

REACTION PARAMETERS :
- Type of Reaction : Kinetic/Increasing OD
- Wavelength : 340 nm
- Flowcell Temperature : 37⁰C
- Delay Time : 60 Seconds
- Interval Time : 30 Seconds
- Number of Readings : 4
- Reagent Volume : 1.0 ml.
- Sample Volume : 50 µl (0.05 ml)
- Factor : 3376
- Zero setting with : Distilled Water
- Path length : 1.0 cm

PROCEDURE :

PIPETTE INTO TEST TUBES		**TEST**
WORKING REAGENT	(ml)	1.0
SAMPLE	(ml)	0.05

Mix and read first absorbance of the test exactly at one minute & thereafter at 30, 60 & 90 seconds at 340 nm. Determine the mean change in absorbance per minute and calculate test results.

NOTE :
For laboratories using instruments with cuvette capacity more than/ less than 1.0 ml, sample and working reagent volumes should be proportionately increased/decreased.

TEST RESULTS :

LDH activity = ΔA/min X F

$$\text{Where } F = \frac{1}{6.22} \times \frac{T.V.}{S.V.} \times 1000 = 3376$$

T.V. = Total Volume = 1.05 ml.
S.V. = Sample Volume = 0.05 ml.
6.22 = Millimolar Extinction Coefficient of NADH at 340 nm.

NORMAL VALUES :
114 - 240 IU/L at 37⁰C (Adults).

LINEARITY :

The method is linear upto 2000 IU/L . For samples with values higher than 2000 IU/L, dilute the samples suitably with 0.9% saline and repeat the assay. Apply proper dilution factor and calculate the final results.

REFERENCES :

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