LACTATE DEHYDROGENASE
What it is

- Lactate dehydrogenase (LDH) is an enzyme catalysing the reaction:

  \[ \text{Pyruvate} + \text{NADH} \rightleftharpoons \text{Lactate} + \text{NAD}^{+} \]

- LDH is in almost all of the body's cells and is released into the blood when cells are damaged or destroyed

  ➔ the blood level of LDH is a general indicator of tissue and cellular damage
What it is

- LDH exists in five forms, or isoenzymes.
- Each isoenzyme has a slightly different structure and is found in different tissues.
- The results of each LDH isoenzyme concentration indicates which tissue may be damaged or injured.
Isoenzymes are multiple enzyme isomers of enzyme.
There are five isoenzymes of LDH.
• LDH-1 found in heart and in RBC as well as in brain.
• LDH-2 found in the reticuloendothelial system.
• LDH-3 found in the lungs.
• LDH-4 found in the kidneys, placenta and pancreas.
• LDH-5 found in the liver and striated muscle.
• METHOD:- KING’S (COLORIMETRIC) METHOD

• PRINCIPLE:-
LDH catalyzes the reaction.

Lactate +NAD → Pyrvate + NADH2

By the addition of dinitrophenyl hydrazine (DNPH), Dinitrophenyl hydrazone gives measures of LDH concentration and can be measured after alkaline dilution at 440 nm (violet filter)
Stability of the reagents
Reagent 1, 3 & 6 are stable at 2-8°C for one year.
Reagent 2 is stable at 0-4°C for 6 months.
Reagent 4 is stable at 0-4°C for one week.
Reagent 5 is stable at room temperature (25°C ± 5°C) for 2 years.

Procedure
Wavelength : 440 nm; (Violet filter, 405 - 450 nm)
Incubation time : 15 minutes
Incubation temp. : 37°C
One blank is sufficient for each assay series.
Pipette in the tubes labelled as follows:

<table>
<thead>
<tr>
<th>Test</th>
<th>Blank</th>
</tr>
</thead>
<tbody>
<tr>
<td>1) Substrate, ml</td>
<td>1.0</td>
</tr>
<tr>
<td>Incubate at 37°C for .5 minutes</td>
<td></td>
</tr>
<tr>
<td>2) NAD solution, ml</td>
<td>0.2</td>
</tr>
<tr>
<td>3) Serum, ml</td>
<td>0.02</td>
</tr>
<tr>
<td>Incubate at 37°C for exactly 15 minutes</td>
<td></td>
</tr>
<tr>
<td>4) D.N.P.H., ml</td>
<td>1.0</td>
</tr>
<tr>
<td>5) Serum, ml</td>
<td></td>
</tr>
<tr>
<td>Mix, keep at room temperature (25°C ± 5°C) for 15 minutes.</td>
<td></td>
</tr>
<tr>
<td>6) 0.4 N sodium hydroxide, ml</td>
<td>10</td>
</tr>
</tbody>
</table>

Mix thoroughly & measure optical density of test against blank at 440 nm (violet filter).
CLINICAL SIGNIFICANCE OF LDH

LDH may be used as a general indicator for the severity of acute and chronic tissue damage.
LDH may be used to detect and monitor progressive conditions such as anemia including hemolytic anemia, megaloblastic anemia and severe infections.
LDH determine prognosis or monitor treatment i.e chemotherapy of cancer such as germ cell tumors e.g testicular and ovarian cancer, lymphoma, leukemia and neuroblastoma.
 Conditions in which it is elevated:

- Blood flow deficiency
- Hemolytic anemia
- Infectious mononucleosis
- Liver disease
- Low blood pressure
- Muscle injury
- Muscle weakness, loss of muscle tissue (muscular dystrophy)
Cancer
Pancreatitis
Stroke
Tissue death
If LDH level is raised may order an LDH isoenzyme test to determine the location of tissue damage.