# LDH FS*

**DGKC 1970**

**Diagnostic reagent for quantitative in vitro determination of lactate dehydrogenase (LDH) in serum or plasma on photometric systems**

## Order Information

| Cat. No. | Kit size | | | | | |
|---|---|---|---|---|---|---|
| 1 4201 99 10 021 | R1 | 5 x | 20 mL | + R2 | 1 x | 25 mL |
| 1 4201 99 10 026 | R1 | 5 x | 80 mL | + R2 | 1 x | 100 mL |
| 1 4201 99 10 023 | R1 | 1 x | 800 mL | + R2 | 1 x | 200 mL |
| 1 4201 99 10 704 | R1 | 8 x | 50 mL | + R2 | 8 x | 12.5 mL |
| 1 4201 99 10 917 | R1 | 8 x | 60 mL | + R2 | 8 x | 15 mL |
| 1 4201 99 90 305 | R1 | 10 x | 12 mL | + R2 | 2 x | 20 mL |

## Summary [1,2]

Lactate dehydrogenase (LDH) is an enzyme, consisting of five different isoenzymes which catalyze the interconversion of L-lactate and pyruvate. LDH is present in the cytoplasm of all human tissues with higher concentrations in liver, heart and skeletal muscle, and lower values in erythrocytes, pancreas, kidney and stomach. Increased LDH activities are found in a variety of pathological conditions such as myocardial infarction, cancer, diseases of liver, blood or muscle. However, because of the lack of organ specificity, determination of its isoenzymes or other enzymes such as alkaline phosphatase or ALAT/ASAT is necessary for differential diagnosis.

## Method

Optimized test according to German Society of Clinical Chemistry (DGKC) [3].

## Principle

Pyruvate + NADH + $H^+$ <—LDH—> Lactate + $NAD^+$

## Reagents

### Components and Concentrations

| | | | |
|---|---|---|---|
| **R1:** | Phosphate buffer | pH 7.5 | 64 mmol/L |
| | Pyruvate | | 0.80 mmol/L |
| **R2:** | Good's buffer | pH 9.6 | |
| | NADH | | 1.0 mmol/L |

### Storage Instructions and Reagent Stability

The reagents are stable up to the end of the indicated month of expiry, if stored at 2 – 8 °C and contamination is avoided. Do not freeze the reagents!
Reagent 2 must be protected from light.

### Warnings and Precautions

1. The reagents contain sodium azide (0.95 g/L) as preservative. Do not swallow! Avoid contact with skin and mucous membranes.
2. In very rare cases, samples of patients with gammopathy might give false results [7].
3. Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
4. For professional use only!

### Waste Management

Please refer to local legal requirements.

### Reagent Preparation

***Substrate Start***

The reagents are ready to use.

***Sample Start***

Mix 4 parts of R1 + 1 part of R2
(eg. 20 mL R1 + 5 mL R2) = monoreagent
Stability: 5 days at 2 – 8 °C
8 hours at 15 – 25 °C
The monoreagent must be protected from light.

### Materials required but not provided

NaCl solution 9 g/L
General laboratory equipment

## Specimen

Serum, heparin plasma or EDTA plasma
Stability [4]:
4 days at 20 – 25 °C
6 weeks at 4 – 8 °C
Discard contaminated specimens.

## Assay Procedure

***Application sheets for automated systems are available on request.***

| | |
|---|---|
| Wavelength | 340 nm, Hg 365 nm, Hg 334 nm |
| Optical path | 1 cm |
| Temperature | 25 °C/30 °C/37 °C |
| Measurement | Against air |

***Substrate Start***

| | | |
|---|---|---|
| **Temperature** | 25°C/30°C | 37 °C |
| **Sample/Calibrator** | 20 µL | 10 µL |
| **Reagent 1** | 1000 µL | 1000 µL |
| Mix, incubate for approx. 1 – 5 min., then add: | | |
| **Reagent 2** | 250 µL | 250 µL |
| Mix, read absorbance after 1 min. and start stopwatch. Read absorbance again after 1, 2 and 3 min. | | |

***Sample Start***

| | | |
|---|---|---|
| **Temperature** | 25 °C/30 °C | 37 °C |
| **Sample/Calibrator** | 20 µL | 10 µL |
| **Mono-reagent** | 1000 µL | 1000 µL |
| Mix, read absorbance after 1 min. and start stopwatch. Read absorbance again after 1, 2 and 3 min. | | |



* fluid stable

## Calculation

**With factor**

From absorbance readings calculate ΔA/min and multiply by the corresponding factor from table below:

**ΔA/min x factor = LDH activity [U/L]**

| Substrate Start | 25 °C/30 °C | 37 °C |
|---|---|---|
| 340 nm | 10080 | 20000 |
| 334 nm | 10275 | 20390 |
| 365 nm | 18675 | 37060 |
| | | |
| Sample Start | 25 °C/30 °C | 37 °C |
| 340 nm | 8095 | 16030 |
| 334 nm | 8250 | 16345 |
| 365 nm | 15000 | 29705 |

**With calibrator**

$$\text{LDH [U/L]} = \frac{\Delta A/\text{min Sample}}{\Delta A/\text{min Calibrator}} \text{ x Conc. Calibrator [U/L]}$$

**Conversion factor**

LDH [U/L] x 0.0167 = LDH [µkat/L]

## Calibrators and Controls

For the calibration of automated photometric systems, DiaSys TruCal U calibrator is recommended. This method is traceable to the molar extinction coefficient. DiaSys TruLab N and P controls should be assayed for internal quality control. Each laboratory should establish corrective action in case of deviations in control recovery.

| | Cat. No. | Kit size |
|---|---|---|
| TruCal U | 5 9100 99 10 063 | 20 x 3 mL |
| | 5 9100 99 10 064 | 6 x 3 mL |
| TruLab N | 5 9000 99 10 062 | 20 x 5 mL |
| | 5 9000 99 10 061 | 6 x 5 mL |
| TruLab P | 5 9050 99 10 062 | 20 x 5 mL |
| | 5 9050 99 10 061 | 6 x 5 mL |

## Performance Characteristics

**Measuring range**

On automated systems the test is suitable for the determination of LDH activities up to 1200 U/L.
In case of a manual procedure, the test is suitable for LDH activities which correspond to a maximum of ΔA/min of 0.15 at 340 and 334 nm or 0.08 at 365 nm.
If these values are exceeded the sample should be diluted 1+10 with NaCl solution (9 g/L) and results multiplied by 11.

**Specificity/Interferences**

No interference was observed by ascorbic acid up to 30 mg/dL, bilirubin up to 40 mg/dL and lipemia up to 2,000 mg/dL triglycerides. Hemolysis interferes because LDH is released by erythrocytes. For further information on interfering substances refer to Young DS [5].

**Sensitivity/Limit of Detection**

The lower limit of detection is 5 U/L.

**Precision (at 25 °C)**

| Intra-assay precision n = 20 | Mean [U/L] | SD [U/L] | CV [%] |
|---|---|---|---|
| Sample 1 | 142 | 5.50 | 3.86 |
| Sample 2 | 245 | 4.95 | 2.01 |
| Sample 3 | 497 | 8.39 | 1.69 |

| Inter-assay precision n = 20 | Mean [U/L] | SD [U/L] | CV [%] |
|---|---|---|---|
| Sample 1 | 144 | 3.09 | 2.13 |
| Sample 2 | 248 | 4.53 | 1.82 |
| Sample 3 | 492 | 6.23 | 1.26 |

**Method Comparison**

A comparison of DiaSys LDH FS (y) with a commercially available test (x) using 78 samples gave following results:
y = 1.03 x + 2.13 U/L; r= 0.999.

## Reference Range [6]

| | 25 °C | 30 °C | 37 °C | Unit |
|---|---|---|---|---|
| Adults: | < 240 | < 346 | < 480 | [U/L] |
| | < 4 | < 5.77 | < 8 | [µkat/L] |

Each laboratory should check if the reference ranges are transferable to its own patient population and determine own reference ranges if necessary.

## Literature


1. Thomas L. Clinical laboratory diagnostics. 1st ed. Frankfurt: TH-Books Verlagsgesellschaft;1998.p.89–94.
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3. Deutsche Gesellschaft für klinische Chemie. Empfehlungen der deutschen Gesellschaft für Klinische Chemie (DGKC). Standardisierung von Methoden zur Bestimmung von Enzymaktivitäten in biologischen Flüssigkeiten. Z Klin Chem Klin Biochem 1972;10:182-92.
4. Guder WG, Zawta B et al. The Quality of Diagnostic Samples. 1st ed. Darmstadt: GIT Verlag; 2001; p. 36-7.
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7. Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: Mechanism, detection and prevention. Clin Chem Lab Med 2007; 45(9): 1240–1243.


## Manufacturer


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