LACTATEDEHYDROGENASE-P

Cat. No.	Pack Name	Packaging (Content)
BLT00037	LDH 100	R1: 4 x 20 ml, R2: 1 x 20 ml

EN

CE IVD

INTENDED USE
Diagnostic reagent for quantitative *in vitro* determination of LDH in human serum and plasma (DGKCH method).

CLINICAL SIGNIFICANCE
The enzyme Lactate dehydrogenase (LDH) is concentrated in heart, kidney, liver, muscle and body tissues. Consequently, damage to these results in increased serum levels of LDH. Elevated levels are associated with myocardial infarction, renal damage, hepatitis, anemias, malignancies and muscular disease or damage.

PRINCIPLE
The LDH method is based on the recommendations of DGKCH (from pyruvate). This reagent uses pyruvate and is based on the method of Henry et al.

Pyruvate + NADH ⟶ Lactate + NAD+

LDH catalyses the reduction of pyruvate to lactate oxidising reduced nicotinamide adenine dinucleotide (NADH) to NAD. The activity of LDH can be determined by the rate of decrease in absorbance at 340 nm as NAD is produced.

REAGENT COMPOSITION
R1

Tris Buffer (pH 7.5)	100 mmol/l
Pyruvat	2.0 mmol/l

R2

NADH	1.66 mmol/l

REAGENT PREPARATION
Reagents are liquid, ready to use.

STABILITY AND STORAGE
The unopened reagents are stable till the expiry date stated on the bottle and kit label when stored at 2–8°C.

Two reagents method
Reagents are ready to use.
After opening, reagents are stable until expiry date at 2–8°C if stored at appropriate conditions, closed carefully and without any contamination.

Monoreagent method
Mix 4 portion of Reagent R1 and 1 portion of Reagent R2.
Stability:

24 hours	at 15–25°C at dark
5 days	at 2–8°C at dark

SPECIMEN COLLECTION AND HANDLING
Use serum, plasma (heparin, EDTA).
It is recommended to follow NCCLS procedures (or similar standardized conditions).

Loss of activity:

within 24 hours	at 15–25°C	< 2%
within 3 days	at 2–8°C	< 8%
Stability at least 6 weeks	at –20 °C	

CALIBRATION
Calibration with calibrator XL MULTICAL, Cat. No. XSYS0034 is recommended.

QUALITY CONTROL
For quality control ERBA NORM, Cat. No. BLT00080 and ERBA PATH, Cat. No. BLT00081 are recommended.

UNIT CONVERSION
U/l x 0.017 = µkat/l

EXPECTED VALUES [2]
At 37°C: 225 - 450 U/l
It is recommended that each laboratory verify this range or derives reference interval for the population it serves.

PERFORMANCE DATA
Data contained within this section is representative of performance on ERBA XL systems. Data obtained in your laboratory may differ from these values.

Limit of quantification: 43.8 U/l
Linearity: 1200 U/l
Measuring range: 43.8 - 1200 U/l

Intra-assay precision Within run (n=20)	Mean (U/l)	SD (U/l)	CV (%)
Sample 1	76.74	3.6	0.49
Sample 2	76.02	7.8	0.99

Inter-assay precision Run to run (n=20)	Mean (U/l)	SD (U/l)	CV (%)
Sample 1	562.8	13.8	2.43
Sample 2	312.0	5.4	1.88

COMPARISON
A comparison between XL-Systems LDH (y) and a commercially available test (x) using 40 samples gave following results:
y = 1.982 x + 0.06 U/l
r = 0.996

INTERFERENCES
Following substances do not interfere:
bilirubin up to 20 mg/dl, triglycerides up to 500 mg/dl, haemoglobin up to 5.0 g/l.
Significant hemolysis may increase LD concentration because of high levels of LD in the erythrocytes.

WARNING AND PRECAUTIONS
For *in vitro* diagnostic use. To be handled by entitled and professionally educated person.

WASTE MANAGEMENT
Please refer to local legal requirements.

ASSAY PROCEDURE
Wavelength 340 nm, Hg 334 nm, Hg 365 nm
Cuvette 1 cm

Two reagents method – substrate start

	Reagent blank	Calibrator	Sample
Reagent 1	0.800 ml	0.800 ml	0.800 ml
Sample	–	–	0.020 ml
Calibrator	–	0.020 ml	–
Distilled water	0.020 ml	–	–

Mix and after 1 min. incubation (at 37°C) add:

Reagent 2	0.200 ml	0.200 ml	0.200 ml

Mix, incubate 1 min. at 37°C and then measure the initial absorbance of calibrator and sample against reagent blank. Measure the absorbance change exactly after 1, 2 and 3 min. Calculate 1 minute absorbance change (ΔA/min).

Monoreagent method – sample start

	Reagent blank	Calibrator	Sample
Working reagent	1.000 ml	1.000 ml	1.000 ml
Sample	–	–	0.020 ml
Calibrator	–	0.020 ml	–
Distilled water	0.020 ml	–	–

Mix, incubate 1 min. at 37°C and then measure the initial absorbance of calibrator and sample against reagent blank. Measure the absorbance change exactly after 1, 2 and 3 min. Calculate 1 minute absorbance change (ΔA/min).

CALCULATION
1. $\text{LDH (U/l)} = \frac{\Delta A_{sam}/\text{min}}{\Delta A_{cal}/\text{min}} \times C_{cal}$ C_{cal} = calibrator concentration

2. Using factor:
LDH (U/l) = f x ΔA/min
f = factor
f = 8095 (at 340 nm)

Applications for automatic analysers are available on request.

ASSAY PARAMETERS FOR PHOTOMETERS

Mode	Kinetic
Wavelength 1 (nm)	340
Sample Volume (µl)	10/20
Reagent Volume (µl)	500/1000
Lag Time (sec.)	60
Kinetic Interval (sec.)	60
No. of readings	3
Kinetic factor	8095
Reaction temperature (°C)	37
Reaction direction	Decreasing
Normal Low (U/l)	225
Normal High (U/l)	450
Linearity Low (U/l)	43.8
Linearity High (U/l)	2000
Absorbance Limit (Max.)	0.8
Blank with	Water
Units	U/l

Erba Lachema s.r.o., Karásek 1d, 621 00 Brno, CZ
e-mail: diagnostics@erbalachema.com, www.erbamannheim.com

N/52/13/A/INT Date of revision: 27. 11. 2013

12000058

REFERENCES

1. Searcy, R.L., Diagnostic Biochemistry, McGraw-Hill, New York, NY, 1969.
2. Tietz Textbook of Clinical Chemistry and Molecular Diagnostics. Burtis, C.A., Ashwood, E.R., Bruns, D.E.; 5th edition, WB Saunders Comp., 2012.
3. Henry, R.J., Chiamori N., Golub O.J., and Berkman S., Am.J. Clin. Path. 34(341), 1960.
4. Lum, G., Gambino, S.R., Am.J. Clin. Pathol. 61(108), 1974.
5. Bergmeyer, H. W., Methods of Enzymatic Analymatic Analysis, Ed.2, Verlog Chemie, 1965.
6. Young DS, Effects of Drugs on Clinical Laboratory Tests. Third Edition. 1990; 3 : 221-4.

SYMBOLS USED ON LABELS

REF Catalogue Number	Manufacturer	See Instruction for Use
LOT Lot Number	CE Mark - Device comply with the Directive 98/79/EC	Storage Temperature
Expiry Date	IVD In Vitro Diagnostics	CONT Content

QUALITY SYSTEM CERTIFIED
ISO 9001 ISO 13485

Erba Lachema s.r.o., Karásek 1d, 621 00 Brno, CZ
e-mail: diagnostics@erbalachema.com, www.erbamannheim.com