LDH FL DGKC

LD F060 CH	6 x 10 ml
LD F120 CH	12 x 10 ml
LD F245 CH	12 x 20 ml

INTENDED USE

Reagent for quantitative in vitro determination of LDH in biological fluids.

SUMMARY OF TEST

Lactate dehydrogenase (LDH) is present in high levels in kidneys, heart, liver, and skeletal muscle, besides in other human tissues. An increase of circulating level of LDH is an index of myocardial infarction, renal failure, hepatitis, anemia, malignancies, and affections of skeletal muscles.

PRINCIPLE OF THE METHOD

Lactate dehydrogenase (EC 1.1.1.27.; L-lactate:NAD^+ oxidoreductase; LDH) catalyzes the conversion of pyruvate to L-lactate in presence of NADH, which is converted to NAD^+. The rate of conversion of NADH/NAD^+, monitored at 340 nm, is proportional to LDH activity.

KIT COMPONENTS

For in vitro diagnostic use only.
The components of the kit are stable until expiration date on the label.
Keep away from direct light sources.

LDH R1 **F060: 6 x 8 ml (liquid) blue cap**
F120: 12 x 8 ml (liquid) blue cap
F245: 12 x 16 ml (liquid) blue cap

LDH R2 **F060: 1 x 12 ml (liquid) red cap**
F120: 2 x 12 ml (liquid) red cap
F245: 3 x 16 ml (liquid) red cap

Composition in the test: phosphate buffer pH 7.50 50 mM, sodium pyruvate 0.60 mM, NADH 0.18 mM.

Store all components at 2-8°C.

MATERIALS REQUIRED BUT NOT SUPPLIED

Current laboratory instrumentation. Spectrophotometer UV/VIS with thermostatic cuvette holder. Automatic micropipettes. Glass or high quality polystyrene cuvettes. Saline solution.

REAGENT PREPARATION

Serum as starter procedure:
Codes F060/F120: add 2 ml of reagent R2 to a vial of reagent R1.
Code F245: add 4 ml of reagent R2 to a vial of reagent R1.
Stability of working reagent: 30 days at 2-8°C, away from light sources.

Reagent as starter procedure:
use separate reagents ready to use.
Stability: up to expiration date on labels at 2-8°C;
Stability since first opening of vials: preferable within 60 days at 2-8°C.

PRECAUTIONS

LDH R1: Warning. Causes serious eye irritation (H319). Causes skin irritation (H315). Wear protective gloves. Eye protection (P280). IF ON SKIN: Wash with plenty of water (P302+P352). IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing (P305+P351+P338). If eye irritation persists: get medical advice (P337+P313).

LDH R2: It is not classified as hazardous.

SPECIMEN

Serum, plasma heparinate or EDTA. Avoid hemolysis.
LDH activity is stable 3 days in samples stored at 2-8°C.

TEST PROCEDURE (sample as starter)

Wavelenght:	340 nm
Ligthpath:	1 cm
Temperature:	37°C
dispense in cuvette working reagent:	1 ml
preincubate at 37°C for 5 minutes.	
add sample:	10 µl
Mix, execute a first reading of absorbance after 1 minute, incubating at 37°C. Perform other 3 readings at 60 seconds intervals. Calculate the ΔA/min.	

TEST PROCEDURE (reagent as starter)

Wavelenght:	340 nm
Ligthpath:	1 cm
Temperature:	37°C
dispense in cuvette reagent R1:	1 ml
add sample:	10 µl
incubate at 37°C for 5 minutes.	
dispense in cuvette reagent R2:	250 µl
Mix, execute a first reading of absorbance after 1 minute, incubating at 37°C. Perform other 3 readings at 60 seconds intervals. Calculate the ΔA/min.	

RESULTS CALCULATION

Perform calculation in units per litre, multiplying the ΔA/min by the factor as it is indicated.

Calculation in U/l: ΔA/min x 16030 (sample starter)
Calculation in U/l: ΔA/min x 20080 (reagent starter)

Activity in µkat/l: U/l x 0.0167 = µkat/l

EXPECTED VALUES

225 - 450 U/l (3.75 - 7.51 µkat/l)

Each laboratory should establish appropriate reference intervals related to its population.

QUALITY CONTROL AND CALIBRATION

It is suggested to perform an internal quality control. For this purpose the following human based control sera are available:
QUANTINORM CHEMA
with normal or close to normal control values
QUANTIPATH CHEMA
with pathological control values.
If required, a multiparametric, human based calibrator is available:
AUTOCAL H

Please contact Customer Care for further information.

TEST PERFORMANCE

Linearity
the method is linear up to 4000 U/l.
If a ΔA/min of 0.100 is exceeded, it is suggested to dilute sample 1+9 with saline and to repeat the test, multiplying the result by 10.

Sensitivity/limit of detection (LOD)
the limit of detection is 31 U/l.

Interferences
no interference was observed by the presence of:

hemoglobin	≤ 150 mg/dl
bilirubin	≤ 40 mg/dl
lipids	≤ 500 mg/dl

Precision

intra-assay (n=10)	mean (U/l)	SD (U/l)	CV%
sample 1	329.90	6.33	1.90
sample 2	531.90	7.75	1.50

inter-assay (n=20)	mean (U/l)	SD (U/l)	CV%
sample 1	331.51	7.39	2.20
sample 2	546.04	11.76	2.20

Methods comparison
a comparison between Chema and a commercially available product gave the following results:

LDH Chema = x
LDH competitor = y
n = 99

$y = 0.99x + 2.41$ U/l $r^2 = 0.99$

WASTE DISPOSAL

This product is made to be used in professional laboratories.
P501: Dispose of contents according to national/international regulations.

REFERENCES

HU Bergmeyer - Methods of enzymatic analysis, Vol. III (1987).
DGKC - Eur.J.Clin.Chem.Clin.Biochem., 31 (1993).
Kreutzer H.H. et al. - Clin. Chim. Acta 9,64 (1964)
Young D.S., et al. - Clin. Chem. 21 ID, 432D (1975)

MANUFACTURER

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SYMBOLS

IVD	*in vitro* diagnostic medical device
LOT	batch code
REF	catalogue number
	temperature limit
	use by date
	caution
	consult instructions for use

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