DIRECT LDL-CHOLESTEROL (LDL-1000A/B)

D-LDL-C

Test for the quantitative determination of LDL-Cholesterol in human serum and plasma. Liquid. Dual reagents. Store at 2°C - 8°C. Do not freeze. For in Vitro Diagnostic Use.

INTENDED USE

The test is applied for the quantitative determination of LDL (Low Density Lipoprotein)-cholesterol in human serum and plasma.

TEST PRINCIPLE

The assay consists of distinct reaction steps:

1. The LDL complexes with polyanion. The detergent 1 in Reagent 1 is soluble only in the non-LDL lipoprotein particles (CM, HDL, VLDL). The cholesterol released will be used up by enzymatic reagent and be in a non-color forming reaction without the chromogenic coupler.
2. The cholesterol released from LDL-C by detergent 2 in Reagent 2 reacts with chromogenic coupler for the colour formation.

TEST PARAMETERS

Method	: Colorimetric
Wavelenght	: Main 572-600 nm /Sub 700-750 nm
Temperature	: 37 °C*
Sample	: Serum, plasma
Linearity	: 5-600 mg/dL*

REAGENT COMPOSITION

Reagent 1:

Polyanion detergent 1

Cholesterol esterase	:	≤ 200.000 U/L
Cholesterol oxidase	:	≤ 200.000 U/L
Peroxidase	:	≤ 200.000 U/L

4-aminoantipyrine

TOOS

Reagent 2:

Detergent 2

TOOS

Tris Buffer

Testing of human serum used in the preparation of the standard is resulted as negative for the presence of antibodies anti-HIV and anti-HCV, beside for HBs antigen. Because of the possibility of being infectious, standard should be used cautiously and with GLP rules.

REAGENT PREPARATION

Reagents are ready to use, liquid.

REAGENT STABILITY AND STORAGE

Store at 2-8ºC. Reagents are stable till the expiry date stated on the label when they stored in closed vials and avoiding contamination during their usage.

There is a strong relation between on board stability and auto analysers cooling specification and carry-over values.

SAMPLE

Samples: Fresh Serum or EDTA and heparinized plasma on an empty stomach are the recommended specimens.
Note: Separate the serum or plasma as soon as possible after collection (within 3 hours). Store serum no more than 12 hours at room temperature, no more 7 days at 2-8 ºC. Serum is stable for 30 days at (-60)-(-80)°C.

TEST PROCEDURE

Sample Start
In case of request, ready application procedures dedicated to different kind of photometers and ready manual working procedures can be supplied.

In case of request, ready application procedures dedicated to different kind of biochemistry auto analyzers can be supplied.

Substrate Start
In case of request, ready application procedures dedicated to different kind of biochemistry auto analyzers can be supplied.

CALCULATIONS

The LDL-Direct concentration in the sample is calculated using the following general formula:

$$\text{Cholesterol (mg/dL)} = \frac{A_{\text{calibrator}}}{A_{\text{sample}}} \times \text{Conc. of Std./Cal (mg/dL)}$$

LDL=Cholesterol-(HDL + Triglyserid/5)
Unit Conversion
mmol/Lx38.61=mg/dL

REFERENCE INTERVALS (NORMAL VALUES)

Optimal :<100 mg/dL (<2.59 mmol/L)
Near optimal,
Above optimal : 100 – 129mg/dL (2.59 – 3.34 mmol/L)
Borderline high : 130 - 159 mg/dL (3.37 – 4,12 mmol/L)
High : 160 – 189 mg/dL (4,14 – 4,89 mmol/L)
Very high : ≥ 190 mg/dL (≥4,92 mmol/L)

It is recommended that each laboratory establish its own reference range.

QUALITY CONTROL AND CALIBRATION

Commercially available control material with established values determined by this method may be used. We recommend:

Assayed Control Serum Normal

Assayed Control Serum Pathological

The assay requires the use of a HDL/LDL Standard (Calibrator). Any commercially available Standard

or Calibrator suitable for this method may be used. We recommend:

Standard (Lipids HDL/LDL Calibrator) (Recommended)

Calibration Stability: It is strongly depend of application to auto analyzers and auto analyzers specification. Calibration stability is 20 days.

*Each laboratory should establish its own internal Quality Control scheme and procedures for corrective action if controls do not recover within the acceptable tolerances.

Quality control is recommended every morning. Calibration is not recommended if QC control values are acceptable. Reagent should be calibrated after lot changes.

PERFORMANCE CHARACTERISTICS

***Low Linearity (LOQ)**(LOQ values are based on CV% ≤ 20%): 5 mg/dL.

***High Linearity:** The test is linear up to 600 mg/dL.

Precision Studies (Based on CLSI EP05A3 Doc.):

Repeatability (within run)(intra-assay):

Mean concentration	CV	n
45 mg/L	4.6 %	20
65.0 mg/L	3.4 %	20

Reproducibility (run to run)(inter-assay):

Mean concentration	CV	n
45 mg/L	4.2 %	25
65 mg/L	3.9 %	25

***Sensitivity (LOD)**:4.5 mg/dL.

Trueness: No systematic differences seen in results obtained with this reagent when compared with reference reagents. It's available to get details of comparison experiments in case of requirement.

***Interferences:**

The acceptable interference limit is set 10% below the highest interferent concentration that is within ±10% recovery of the target.
Hemoglobin up to 6.3g/L, bilirubin up to 13.5mg/dL, lipemia (Triglycerides) up to 2250 mg/dL do not interfere. Other drugs and substances may interfere.
Significant interference may be observed with hemolyzed samples. Reference observed results in the table below.[10, 11]

Interferent and Concentration	LDL Direct Cho. Target (mg/dL)	N	Observed Recovery %
Bilirubin Total 15 mg/dL	48.7	3	92
	70.2	3	108
Triglyceride 2500mg/dL	47	3	101
	98.5	3	98
Hemoglobin 7 g/L	44.4	3	109
Hemoglobin 10 g/L	104	3	108

The effect of interfering substances has only been evaluated for those listed in this labeling.

An analyzer has been used to obtain these performance characteristics. Usage of different analyzer or a manual procedure may cause the variance in results.

***NOTES**

1. For in vitro diagnostic use only. Do not pipette by mouth. Avoid contact with skin and mucous membranes.
2. All the calibrators, controls and some reagents must be considered as human & animal sample, so potentially infectious; all the protection actions must be applied to avoid any potential biological risk.
3. Material safety data sheet will be supplied on request.
4. Exercise the normal precautions required for handling laboratory reagents.
5. Caps of the reagents bottles cannot be used between two different kind of reagent and between R1&R2.
6. Reagents with different lot numbers should not be interchanged or mixed.
7. The reagents contain sodium azide (< 0.1%) as a preservative.

***PRECAUTIONS AND WASTE DISPOSAL**

This product is made to be used in professional laboratories and by professional operators. Perform the test according to the general GLP guidelines.
R32: Contact with acids liberates very toxic gas.
EUH032: Contact with acids liberates very toxic gas.
H300: Fatal if swallowed
H400: Very toxic to aquatic life
H410: Very toxic to aquatic life with long lasting effects.
Refer to special instructions/safety data sheets.
Please consult local regulations for a correct waste disposal.

***ABBREVIATIONS**

CLSI : Clinical and Laboratory Standards Institute
CV% : Coefficient of Variation Percentage
EP : Evaluation Protocols
GLP : Good Laboratory Practice
NCEP : National Cholesterol Education Program
IU : International Unit
mL : milliliter
QC : Quality Control
mg : miligram
L : liter
g : gram
dL : deciliter

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SYMBOLS

IVD	Only for in vitro diagnostic use
LOT	Lot of manufacturing
R1	Reagent 1
R2	Reagent 2
CONC	Concentration
INGRED	Reagent Ingredients
SN	Serial Number
REF	Reference Number (Catalog No)
	Expiration date
	Storage temperature interval
	Read the directions
	Biological risk

CE

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