

# LDL CHOLESTEROL

Direct Enzymatic colorimetric, Liquid

REF: 280 001 100 Test      REF: 280 002 200 Test  
R1 1 x 30 ml   R2 1 x 10 ml   R1 1 x 60 ml   R2 1 x 20 ml

## Intended Use

The SPECTRUM LDL-Cholesterol Assay is intended for the *in vitro* quantitative determination of Low Density Lipoprotein Cholesterol in human serum or plasma. The reagents can assist in the diagnosis and treatment of patients at risk of developing coronary heart disease. Elevated LDL cholesterol is the primary target of cholesterol-lowering therapy.

## Clinical Significance

Low Density Lipoproteins (LDL) are synthesized in the liver by the action of various lipolytic enzymes on triglyceride-rich Very Low Density Lipoproteins (VLDLs). Specific LDL receptors exist to facilitate the elimination of LDL from plasma by liver parenchymal cells. It has been shown that most of the cholesterol stored in atherosclerotic plaques originates from LDL. For this reason the LDL-Cholesterol concentration is considered to be the most important clinical predictor, of all single parameters, with respect to Coronary atherosclerosis.

Accurate measurement of LDL-Cholesterol is of vital importance in therapies which focus on lipid reduction to prevent atherosclerosis or reduce its progress and to avoid plaque rupture.

## Assay Principle

The assay is based on a modified polyvinyl sulfonic acid (PVS) and polyethylene-glycol methyl ether (PEGME) coupled classic precipitation method with the improvements in using optimized quantities of PVS/PEGME and selected detergents. LDL, VLDL, and chylomicron (CM) react with PVS and PEGME and the reaction results in inaccessibility of LDL, VLDL and CM by cholesterol oxidase (CHOD) and cholesterol esterase (CHER), whereas HDL reacts with the enzymes. Addition of R2 containing a specific detergent releases LDL from the PVS/PEGME complex. The released LDL reacts with the enzymes to produce H<sub>2</sub>O<sub>2</sub> which is quantified by the Tindler reaction.

## Reagent Composition

**Reagent 1 (R1):** MES buffer (pH 6.5) Polyvinylsulfonic Acid Polyethyleneglycolmethylester, MgCl<sub>2</sub> Detergent, EDTA, 4aminoantipyrine, Cholesterol esterase, Cholesterol oxidase Peroxidase.

**Reagent 2 (R2):** MES buffer (pH 6.5), EDTA, Detergent TODB, N, N-Bis (4-sulfobutyl)-3-methylaniline)

## LDL Calibrator

Standard, Lyophilized Human Serum  
LDL actual concentration is stated on the vial label.

## Reagent Preparation

SPECTRUM LDL-Cholesterol Assay Reagents (R1, R2) are liquid stable, ready to use reagents.

## Reagent Stability and Storage

Unopened reagents are stable until the expiration date printed on the outer box when stored at 2–8° C. Reagent on-board stability is at least 60 days. The reagent solutions should be clear. If turbid, the reagents may have deteriorated.

## Specimen Collection and Preparation

Use fresh patient serum or plasma samples (EDTA, Citrate). If samples contain LDL cholesterol greater than 250 mg/dL, they should be diluted with saline.

**LDL Calibrator:** Dissolve the contents with the amount of distilled water indicated on Label. Cap vial and mix gently to dissolve contents, and wait for 30 minutes. Stability is 2 weeks at - 20°C.

## REAGENT STABILITY

All the components of the kit are stable until the expiration date on the label when stored tightly closed at 2-8°C and contaminations are prevented during their use. Do not freeze the reagents.

## LINEARITY

250 mg/dl

## PROCEDURE

***This reagent can be used manually (see method below) and on most analyzers. Applications are available on request.***

Wavelength	600 nm (580 nm is an option)
Cuvette	1 cm
Temperature	37 °C
Measure	Against distilled water

	Blank	Calibrator	Sample
R1 (µL)	300	300	300
Calibrator(µL)	-	4	-
Sample (µL)	-	-	4
Mix and Incubate for 5 min at 37°C. Then add :			
R2(µL)	100	100	100

Mix Read ***immediately*** the absorbance (A<sub>1</sub>) of the samples and calibrator, against the Blank, then Read the absorbance (A<sub>2</sub>) of the Samples and calibrator after 5 mins, against the Blank.  
**Calculate the Increase of the absorbance  $\Delta A = A_2 - A_1$ .**

## CALCULATION

$\Delta A$  Sample  
—————→ X Calibrator conc. = mg/dL of LDL-C in the sample  
 $\Delta A$  Calibrator

Conversion factor: mg/dL x 0.0259 = mmol/L

## QUALITY CONTROL

Control sera are recommended to monitor the performance of assay procedures.

If control values are found outside the defined range, check the instrument reagents and calibrator for problems.

Each laboratory should establish its own Quality Control scheme and corrective actions if controls do not meet the acceptable tolerances.

## Expected Values

Levels of the risk

Desirable	<100 mg/dL
Medium	100 – 160 mg/dl
High	>160mg/dL

These values are for orientation purpose; each laboratory should establish its own reference range.

## PERFORMANCE CHARACTERISTICS

### Dynamic range:

The measuring range is from 1.0 mg/dl to linearity limit of 250 mg/dl. If the results obtained were greater than linearity limit, dilute the sample 2 times with NaCl 9 g/L and multiply the result by 2.

### Sensitivity:

The sensitivity of the test is 1 mg/dl.

**Accuracy:** Results obtained using Spectrum reagents (y) did not show systematic differences when compared with other commercial reagents. (x).

The results obtained using 92 samples were the following. Correction coefficient (r): 0.996.  
Regression equation:  $y = 4.6 + 0.940x$ .

The results of the performance characteristics depend on the analyzer used.

## INTERFERENCES

No Interferences were observed with ascorbic acid up to 50 mg/dL, haemoglobin up to 500 mg/dL or bilirubin up to 30 mg/dL.

A list of drugs and other interfering substances with LDL cholesterol determination has been reported by Young et al <sup>8,4</sup>

## NOTES

Spectrum has Instrument application sheets for several automatic analyzers. Instructions for many of them are available on request.

## BIBLIOGRAPHY

1. Kaplan A et al. Lipoprotein Clin Chem the C. V. Masby Co. St Louis.
2. Okada M. et al Low- density lipoprotein can be chemically measured  
J. Lab, Clin. Med., 1996; 132, 195-201.
3. Young DS. Effects of Drugs on Clinical Lab. Tests, 4th ad AACC Press, 1995.
4. Young DS. Effects of diseases on Clinical Lab. Tests 4th ad AACC 2001.
5. Burlis A et al. Teitz Texbook of Clinical Chemistry, 3rd ed AACC 1999.
6. Tietz N W et al, Clinical Guide to Laboratory Tests, 3rd ed AACC 1995.

## ORDERING INFORMATION

CATALOG NO.	QUANTITY
280 001	100 tests
280 002	200 tests

**Egyptian Company for Biotechnology (S.A.E.)** Obour city  
Industrial area, block 20008 piece 19 A, Cairo, Egypt.  
Tel: 02 4665 1848 Fax: 02 4665 1847  
www.Spectrum-diagnostics.com  
E-mail: info@Spectrum-diagnostics.com



EC	REP
----	-----

MDSS GmbH Schiffgraben 41  
30175 Hannover, Germany

IFUFCC71

Rev.(3), 26/4/2014