

HDL CHOLESTEROL - Precipitant

REF: 266 001 (1 X 50 ml) **100 test**
 REF: 266 002 (4 X 100 ml) **800 test**
 REF: 266 003 (2 X 50 ml) **200 test**
 REF: 266 004 (5 X 50 ml) **500 test**

Intended Use

Spectrum Diagnostics HDL cholesterol reagent is intended for the in-vitro quantitative, diagnostic determination of HDL cholesterol in human serum, heparinized or EDTA plasma.

Background

High density lipoprotein measurement, in conjunction with other lipid determination, has been shown to be useful in assessing the risk of coronary heart disease. HDL is responsible for carrying cholesterol back from peripheral cells to the liver, therefore the risk of coronary heart disease is lowered with increased levels of HDL. usually, very low density lipoprotein (VLDL) and low density lipoprotein (LDL) are selectively precipitated from serum or plasma samples followed by determination of cholesterol in the HDL-containing supernatant.

Method

Precipitation Method.

Assay Principle

low density lipoproteins (LDL) and very low density lipoproteins (VLDL) in sample precipitate with phosphotungstate and magnesium ions. After centrifugation, the cholesterol concentration in the HDL fraction, which remains in the supernatant, is determined.

Cholesterol esters + H₂O $\xrightarrow{\text{chol. esterase}}$ Cholesterol + Fatty acid

Cholesterol + 1/2 O₂ + H₂O $\xrightarrow{\text{chol. oxidase}}$ Cholestenone + H₂O₂

2H₂O + 4-Aminoantipyrine + phenol $\xrightarrow{\text{peroxidase}}$ Quinoneimine + 4H₂O

Reagents

Reagent (R)

Phosphotungstate 0.52 mmol/L
 Magnesium chloride 30 mmol/L

Reagents also contain non-reactive stabilizers and surfactants.

* Supplementary reagents : Apack for Spectrum liquizyme cholesterol reagent is required










Precautions and Warnings

Pay attention to all precautions and warnings listed in Spectrum Diagnostics catalogue available upon request.

Reagent Preparation, Storage and Stability

Spectrum HDL cholesterol reagent is supplied ready-to-use and stable up to the expiry date labeled on the bottles when properly stored at 2 - 8 °C.

SYMBOLS IN PRODUCT LABELLING

	Authorised Representative		Temperature Limitation
	For in-vitro diagnostic use		Use by/Expiration Date
	Batch Code/Lot number		CAUTION. Consult instructions for use
	Catalogue Number		Manufactured by
	Consult instructions for use		

Deterioration

Do not use The HDL cholesterol reagents if precipitate forms. Failure to recover control values within the assigned range may be an indication of reagent deterioration.

Specimen Collection and Preservation

Serum or plasma

EDTA and Heparin may be used as anticoagulants.

Stability : 7 days at 2 - 8 °C
 4 days at 20 - 25 °C

Procedure

1 - Precipitation

Pipette into centrifuge tubes :

Reagent	0.5 ml
Specimen	0.2 ml

Mix and incubate for 10 minutes at room temperature, then centrifuge for 10 minutes at 4000 rpm.

Carefully collect the supernatant.

Stability : the supernatant may be stored up to five days at 2 - 8 °C

2 - Cholesterol - Liquizyme

Pipette into test tubes :

	Blank	Specimen
Distilled water	50 µl	-----
Specimen supernatant	-----	50µl
Cholesterol Reagent	1ml	1ml

Mix, incubate for 10 minutes at 20 - 25 °C or 5 minutes at 37°C. Measure the absorbance of the specimen (A_{specimen}) against reagent blank at 546 nm (500 - 550 nm) within 60 minutes.

Calculation

HDL cholesterol conc. (mg/dL) = A_{sample} x 570

Expected Values

Females	48.6 - 75 mg/dL	1.26 - 1.94 mmol/L
Males	41.0 - 58.7 mg/dL	1.06 - 1.52 mmol/L
Children	51.8 - 71.9 mg/dL	1.34 - 1.86 mmol/L

**To calculate LDL cholesterol
in mg/dL**

$$\text{LDL Cholesterol} = \text{Total Cholesterol} - \frac{\text{Triglycerides}}{5} - \text{HDL Cholesterol}$$

in mmol/L

$$\text{LDL Cholesterol} = \text{Total Cholesterol} - \frac{\text{Triglycerides}}{2.2} - \text{HDL Cholesterol}$$

ORDERING INFORMATION	
REAGENTS	
CATALOG NO.	QUANTITY
266 001	1 x 50 ml
266 002	4 x 100 ml
266 003	2 x 50 ml
266 004	5 x 50 ml


Clinical Interpretation

	Desirable	Standard Risk Level	Increased Risk Level
HDL Cholesterol			
Females (mg/dL)	>65	45 - 65	<45
(mmol/L)	>1.68	1.16 - 1.68	<1.16
Males (mg/dL)	>55	35 - 55	<35
(mmol/L)	>1.42	0.90 - 1.42	<0.90
LDL Cholesterol			
(mg/dL)	<150	150 - 190	>190
(mmol/L)	<3.38	3.88 - 4.91	>4.91
Total Cholesterol			
(mg/dL)	<200	200 - 300	>300
(mmol/L)	<5.17	5.17 - 7.76	>7.76

Spectrum Diagnostics does not interpret the results of a clinical laboratory procedure ; interpretation of the results is considered the responsibility of qualified medical personnel . All indications of clinical significance are supported by literature references .

References

1. National Cholesterol Education Program Recommendation for Measurement of High-Density Lipoprotein Cholesterol: Executive Summary. Clin Chem. 1995;41:1427 - 1433.
2. Friedewald , W.T. et al. Clin. Chem. 1972; 18: 499.
3. Lopes- Virella, M.F. et al. Clin. Chem. 1977; 23: 882.

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