PATHOZYME DIAGNOSTICS

LIQVIPATH HDL-CHOLESTEROL

(PTA - METHOD)

CAT NO.: PTA

Reagent kit for quantitative estimation of HDL Cholesterol in Serum or Plasma.

DIAGNOSTIC SIGNIFICANCE:

Cholesterol from liver is transported to various tissue cells by low density lipoproteins, very low density lipoproteins, and chylomicrons through blood (plasma) circulation. Cholesterol is also synthesized by the cells as needed. The excess cholesterol is transported back to liver by HDL. HDL therefore indicates excess cholesterol received by liver, where it is converted to bile salts and excreted in bile. Elevated levels of HDL-Cholesterol suggest a balanced status of cholesterol metabolism in tissues. Lower levels of HDL cholesterol are associated with higher risks of artherosclerosis (i.e. deposition of cholesterol in cells of blood vessels) and complications like hypertension, coronary heart disease (CHD) related to it.

PRINCIPLE:

High density lipoproteins (HDL) are separated from other lipoprotein fractions by treating serum with phosphotungstic acid and magnesium chloride. HDL remains in solution while all other lipoprotein fractions precipitated; cholesterol content of which is estimated by enzymatic method.

Serum + PTA Reagent → Supernatant (HDL) + Precipitates (other fractions)

SPECIMEN COLLECTION:

Fresh, clear serum under fasting condition with no hemolysis is the specimen of choice. However, plasma collected using heparin as an anticoagulant may also be used.

KIT PRESENTATION:

Pack Size	2 X 25 ml	2 X 50 ml
Precipiting (PTA) Reagent	2 X 25 ml	2 X 50 ml
HDL Cholesterol Std. (50mg/dl)	1 X 1 ml	1 X 1 ml

REAGENT STORAGE & STABILITY:

All reagents are stable at 2-8^{0}C until the expiry date indicated on the label.

ASSAY PARAMETERS:

Reaction	: End point	Sample Vol.	: 50 µl
Wavelength	: 505 (500-520)nm	Reagent Vol.	: 1.0 ml
Zero Setting	: Reagent Blank	Std Conc.	: 50 mg/dl
Incub. Temp.	: 37 ^{0}C	Linearity	: 250 mg/dl
Incub. Time	: 5 Minutes	Dilution Factor	: 2

PROCEDURE:

A. Separation Of HDL Cholesterol From Sample:

Addition Sequence	Test
Precipiting (PTA) Reagent	200 µl
Sample (Test)	200 µl

Mix well and centrifuge at 3500-4000 rpm for 10 minutes. Separate the clear supernatant immediately and determine cholesterol content as per the procedure given in B.

NOTE:

Do not subject HDL - Cholesterol Standard (50 mg/dl) provided in the kit to separation procedure of HDL.

B. HDL Cholesterol Determination:

Pipette into TT	Blank	Standard	Test
Cholesterol Reagent	1.0 ml	1.0 ml	1.0 ml
HDL Cholesterol Std	--	50 µl	--
Supernatant (Test)	--	--	50 µl

Mix & incubate at 37^{0}C for 5 minutes. Read absorbance of test (**T**) and standard (**S**) after 5 minutes against reagent blank at 505 nm (500-520 nm or Green filter).

Male: 35 - 55 mg/dl	**Female**: 45 - 65 mg/dl

CALCULATION:

HDL Cholesterol (mg/dl) = Abs T ÷ Abs S X 50 X **2**

Where 2 = Dilution Factor Of The Sample

NORMAL VALUES:

Each laboratory should establish its own reference range.

CALCULATION FOR LDL CHOLESTEROL

If the value of Triglycerides is known by using Friedewald's equation.

LDL Cholesterol = Total Chol -(HDL-Chol. + Triglycerides ÷ 5)

The formula is valid only if Triglyceride values are normal or not more than 400mg/dl.

REFERENCE:

Burstein M. Scholnick, H.P. and Mortin, R Cholesterol in High Density lipoprotein : Using Mg++/PTA; J. Lipid Res. 19. Pg- 583. (1970).

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Expiry Date | IVD In-Vitro Diagnostics Use | Storage | Mfg. Date | LOT Batch Number | REF Catalogue Number | See Package Insert

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