Pacific Biomarkers
The Value of Expertise

HDL-C Ultracentrifugation

Analyte: HDL Cholesterol

Specimen Type: Serum, Inquire for additional option(s)

Optimum Volume: 2.5 mL *

Stability:

2-8 Degrees C	-20 Degrees C	-70 Degrees C
5 days	2 months	2 years

Reporting Units: mg/dL

Method: Ultracentrifugation & Precipitation & Enzymatic

Biological or Clinical Significance:

HDL cholesterol (HDL-C) is a powerful inverse predictor of risk of coronary heart disease (CHD). Guidelines published by the American Heart Association (AHA) and the National Cholesterol Education Program (NCEP), which is sponsored by the National Heart, Lung and Blood Institute (NHLBI), recommend that physicians determine HDL-C levels together with other tests in a standard lipid profile prior to administering dietary or drug therapies for CHD. The NCEP guidelines state that patients with high cholesterol or borderline high cholesterol with risk factors (e.g. HDL-C less than 40 mg/dL, hypertension, smoking, family history, etc.) be tested for HDL-C. Subsequently, HDL-C and other lipid parameters should be measured 3-4 times per year to monitor the progress of therapy. Because of its protective effect, the NCEP has designated high HDL-C at or above 60 mg/dL as a negative risk factor.

Principle of Test Method:

The lipoprotein fractions (VLDL, IDL, LDL, and HDL) may be separated by preparative ultracentrifugation of plasma or serum. Isolation and quantification of the fractions are performed, along with measurement of total plasma cholesterol, triglycerides, or other analytes, such as apolipoprotein B, to provide a complete assessment of coronary disease risk. Using the ultracentrifuge, the lipoproteins may be separated on the basis of particle density. The most common procedure involves flotation of VLDL at the usual non-protein solvent density of plasma, d = 1.006, with LDL, IDL, and HDL remaining in the infranate or bottom fraction. The “bottom" containing IDL, LDL, and HDL is recovered for lipid quantification. The difference between the “bottom” cholesterol and the total cholesterol represents the VLDL and chylomicrons, if present. When LDL is removed from the “bottom” (i.e., by chemical precipitation), HDL can be quantified, and LDL lipid values may be determined by the difference, i.e., LDL cholesterol equals “bottom” cholesterol minus HDL cholesterol.

To obtain HDL-C by ultracentrifugation, the bottom fraction is precipitated (see HDL-C by dextran sulfate) and then cholesterol is measured on the supernate.

*2.5 mL allows for repeat analysis if needed.

References:

Hainline A, Karon J, and Lippel K., eds. Manual of Laboratory Operations. Lipid Research Clinics Program, Lipid and Lipoprotein Analysis. NIH - Dept. of Health and Human Services, Second Edition, 1982.

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