



HDL Cholesterol

Version: 1

Edited by: John Stack, Gary Cline: Yale MMPC Analytical Core

Summary: Procedure used to determine the concentration of HDL cholesterol in blood, serum, and plasma. HDL Cholesterol is determined in a two-step procedure. First chylomicrons, VLDL, and LDL are selectively reacted with cholesterol esterase and eliminated from the reaction. In the second step, the remaining HDL-cholesterol is assayed as described for total cholesterol.

Reagents and Materials:

Reagent/Material	Vendor	Stock Number
Lipid Calibrator	Prolabs	R85528
HDL Cholesterol Direct Reagent 1	Prolabs	R85549
HDL Cholesterol Direct Reagent 2	Prolabs	R85549

Protocol: Analysis by automated system Cobas Mira Plus.

- 1) Calibrate Cobas for HDL analysis by running a lipid calibrator, HDL Direct Reagent Reagent 1 and HDL Direct Reagent 2.
- 2) Sample handling as performed by the Cobas Mira Plus.
 - a) Pipette 3µL of sample into cuvette.
 - b) Add 180 µL of Direct Reagent 1.
 - c) Add 60 µL of Direct Reagent 2.
 - d) Mixture is incubated at 37°C for 10 minutes.
 - e) Absorbance is measured at 600 nm.

Reagent Preparation:

Lipid Calibrator: As supplied by vendor.

HDL Cholesterol Direct Reagent 1: As supplied by vendor.

HDL Cholesterol Direct Reagent 2 As supplied by vendor.