EZ HDL Cholesterol (354L)

REF	354 LB	Max. 444 Tests
REF	354 LC	Max. 1000 Tests

Pour d'autres langues
Für andere Sprachen
Para otras lenguas
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Για τις άλλες λώσσες
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INTENDED USE

Trinity Biotech's EZ HDL™ Cholesterol test is an in vitro assay for the quantitative determination of high density lipoprotein cholesterol in serum or plasma.

SUMMARY

Blood total cholesterol levels have long been known to be related to coronary heart disease (CHD). In recent years, in addition to total cholesterol, high density lipoprotein cholesterol (HDL-C) has become an important tool used to assess an individual's risk of developing CHD since a strong negative relationship between HDL-C concentration and the incidence of CHD was reported.[1] Thus, there has been substantial interest in HDL-C measurements, and most clinical laboratories routinely perform HDL-C analysis. Selective chemical precipitation techniques are widely used for the determination of HDL-C such as heparin-manganese, dextran sulfate-magnesium, and phosphotungstate-magnesium.[2] However, these techniques require physical separation via centrifugation, which is not suited to large scale lab use. The Trinity Biotech EZ HDL Cholesterol test eliminates the precipitation procedure by employing a specific antibody, and thus, can be applied on automated analyzers.

PRINCIPLE

Anti-human ß-lipoprotein antibody in Reagent 1 binds to lipoproteins (LDL, VLDL, and chylomicrons) other than HDL. The antigen-antibody complexes formed block enzyme reactions when Reagent 2 is added. Cholesterol esterase (CHE) and cholesterol oxidase (CO) in Reagent 2 react only with HDL-C. Hydrogen peroxide produced by the enzyme reactions with HDL-C yields a blue color complex upon oxidase condensation with FDAOS [N-ethyl-N-(2-hydroxy-3-sulfopropyl)-3,5-dimethoxy-4 fluoro-analine, sodium salt] and 4-aminoantipyrine (4AA) in the presence of peroxidase (POD). By measuring the absorbance of the blue color complex produced, at approximately 600 nm, the HDL-C concentration in the sample can be calculated when compared with the absorbance of the EZ HDL Calibrator.

Anti-human

$$\text{LDL, VL chylomicrons} \xrightarrow{\text{ß-lipoprotein antibody}} \text{Antigen-antibody complex}$$

$$\text{HDL Cholesterol} + H_2O + O_2 \xrightarrow{\text{CHE \& CO}} \Delta^4 \text{ cholestenone} + \text{fatty acid} + H_2O_2$$

$$H_2O_2 + \text{FDAOS} + \text{4AA} \xrightarrow{\text{POD}} \text{Blue color Complex}$$

REAGENTS

EZ HDL Reagent 1, Catalog No. 354-1
Good's buffer (30 mmol/l), pH 7.0, containing 4AA (0.9 mmol/l), POD (from horseradish, 2400 U/l), anti-human ßlipoprotein antibody (sheep) and a preservative.

EZ HDL Reagent 2, Catalog No. 354-2
CHE (from Pseudomonas, 4000 U/l), CO (from Nocardia, 20,000 U/l), and FDAOS (0.8 mmol/l).

PRECAUTIONS

EZ HDL Cholesterol Reagents are for "in vitro Diagnostic Use". Normal precautions exercised in handling laboratory reagents should be followed. Dispose of waste observing all local, state and federal laws. Avoid contact and inhalation of EZ HDL Reagents. Refer to Material Safety Data Sheets for any updated risk, hazard or safety information.

EZ HDL Reagents are not to be used internally in humans or animals. Do not use reagents past the expiration date stated on each reagent container label. EZ HDL Reagents are not to be used for any purpose other than described within this insert.

PREPARATION

EZ HDL Reagents 1 and 2 are supplied ready for use.

STORAGE AND STABILITY

EZ HDL Reagents are stable until expiration date printed on the label when stored at 2–8 °C.

DETERIORATION

The presence of precipitates in the reagents or control sera results outside the manufacturer's acceptable range may be an indication of reagent instability.

SPECIMEN COLLECTION AND STORAGE

It is recommended that specimen collection be carried out in accordance with NCCLS document M29-T2. No known test method can offer complete assurance that human blood samples will not transmit infection. Therefore, all blood derivatives should be considered potentially infectious. Use serum, EDTA, sodium citrate, ammonium oxalate or heparin plasma as a specimen. A sample drawn from a fasting patient is preferred, although non-fasting samples are considered acceptable.[3] Store the specimen at 2–8 °C before analysis. HDL is stable for 4 days at 2–8 °C. Specimens frozen at -20 °C show statistically but not clinically significant decreases in HDL measured at 7–14 days.[2]

INTERFERING SUBSTANCES

Ascorbic acid, total and conjugated bilirubin, hemoglobin and RF cause less than 7% difference in recovery values with the assay at levels up to 100, 20, 40, 500 mg/dl, and 231 IU/ml, respectively. Triglyceride levels up to 1200 mg/dl will not affect the results of the HDL assay. If a specimen has a triglyceride concentration that exceeds 1200 mg/dl, dilute 1 part sample with 1 part saline and repeat the assay. Multiply the result by 2. Refer to the work of Young for drug effects on serum HDL levels.[4]

PROCEDURE

MATERIALS SUPPLIED

EZ HDL Cholesterol kit

INSTRUMENTS/MATERIALS REQUIRED BUT NOT PROVIDED:

Automated analyzer such as Hitachi® 717 analyzer. Refer to the operating manual for a description of instrument operation and specifications.

EZ HDL/LDL™ Combination Calibrator, Catalog No. E0277, or equivalent. If using an equivalent, the value of the calibrator used should be assigned by procedures traceable to the National Reference System for Cholesterol (NRS/CHOL) and the calibrator value should be near the medical decision levels. Reconstitute and store according to product instructions.

Quality control material.

TEST PROCEDURE

The EZ HDL Cholesterol procedure is recommended only for use with automated instruments such as the Hitachi 717. All analyzer applications should be validated in accordance with NCEP and CLIA recommendations. Please contact Trinity Biotech Technical Services Department for availability of application procedures for various automated instruments.

RESULTS

The final results are automatically calculated and printed in concentration when using an automated analyzer.

SI UNITS

To convert results into SI units (mmol/L), multiply HDL cholesterol value (mg/dL) by 0.0259. For example, to convert an HDL concentration of 46 mg/dL: 46 x 0.0259 = 1.19 mmol/L.

QUALITY CONTROL

The reliability of test results should be monitored with routine use of control material with known HDL Cholesterol levels in both the normal and abnormal ranges for each run of the assay.

Quality control materials are intended for use only as monitors of accuracy and precision. The NCEP Lipid Standardization Program recommends two levels of control: one in the normal range (35–65 mg/dl) and one in the abnormal range (< 35 mg/dl) for decision making. If values are to be established for unassayed control material, the laboratory should assay each level of control material a sufficient number of times to generate a valid mean and acceptable range. If the problems cannot be resolved, contact Trinity Biotech Technical Services.

LIMITATIONS

The linearity range of the EZ HDL Cholesterol test is 1–180 mg/dl. If the HDL Cholesterol value exceeds 180 mg/dl, dilute 1 part sample with 1 part saline, repeat assay and multiply result by 2.

EXPECTED VALUES

According to the National Cholesterol Education Program a level of 35 mg/dL is the recommended ust-off point. A level of less than 35 mg/dL is considered a risk factor for coronary artery disease.(CAD). [3]
The second report of the NCEP ATP (APTT), published in June 1993, identified a high HDL-cholesterol (≥ 60 mg/dL) as a "negative" risk factor, one that reduced CHD risk.[5]

PERFORMANCE CHARACTERISTICS

IMPRECISION

Total and within-run Imprecision was determined using three levels of control material following the NCCLS EP5-T2 Guideline.

WITHIN RUN

	Level 1	Level 2	Level 3
Overall Mean (mg/dl)	29.00	50.68	80.38
Standard Deviation (mg/dl)	0.55	0.79	0.79
Coefficient of Variation (%)	1.89	1.56	0.98

TOTAL

	Level 1	Level 2	Level 3
Overall Mean (mg/dl)	29.00	50.68	80.38
Standard Deviation (mg/dl)	0.93	1.73	2.13
Coefficient of Variation (%)	3.22	3.41	2.65

SENSITIVITY

The minimum detectable level of this method is estimated to be 1 mg/dl.

CORRELATION

Ninety samples with HDL cholesterol levels ranging from 21 to 105 mg/dl were assayed by the all liquid EZ HDL method (y) and a commercially available phosphotungstic acid (PTA) method (x). The mean value for y was 49 mg/dl and for x was 43 mg/dl. The correlation coefficient (r) was 0.9924. The linear regression equation was y = 0.98x + 5.51.

Additionally, ninety samples with HDL cholesterol levels ranging from 21 to 105 mg/dl were assayed by this all liquid EZ HDL Procedure No. 354L (y) and by the liquid-lyophilized EZ HDL Procedure No. 354 (x). The mean values for both y and x were 48 mg/dl. The correlation coefficient (r) was 0.9933. The linear regression equation was y = 0.98x – 0.03.

A Document of Comparison of the EZ HDL method (y) to the Designated Comparison Method for HDL cholesterol (x) was received from the Cholesterol Reference Method Laboratory Network for the Hitachi 911. This study showed a regression equation of y = 0.87x + 7.00 and a correlation coefficient (r) of 0.999 with 40 samples.

REFERENCES

1. Rifai N, Warnick GR, Ed. Laboratory Measurement of Lipids, Lipoproteins and Apolipoproteins. *AACC Press*, Washington, DC, USA, 1994
2. Burtis CA, Ashwood ER, Ed. Tietz *Textbook of Clinical Chemistry*, 2nd ed., Saunders, Philadelphia, 1994
3. Folsom AR, Kuba K, Luepker RV, et al. Lipid concentrations in serum and EDTA-treated plasma from fasting and non-fasting normal persons, with particular regard to high density lipoprotein cholesterol. *Clin Chem* 1983; 29:505–8
4. Young DS: Effects of Drugs on Clinical Laboratory Tests. *AACC Press*, 4th ed. Washington, DC 1995
5. The Expert Panel. Summary of the second report of the National Cholesterol Education Program (NCEP) expert panel on detection, evaluation, and treatment of high blood cholesterol in adults (Adult Treatment Panel II). *JAMA* 1993; 269:3015–23

ORDERING INFORMATION

KIT		EZ HDL™ Cholesterol
Catalog No.	Item	Quantity
354LB	EZ HDL™ Reagent 1	2 x 60 mL
	EZ HDL™ Reagent 2	2 x 20 mL
354LC	EZ HDL™ Reagent 1	270 mL
	EZ HDL™ Reagent 2	90 mL

REAGENTS REQUIRED BUT NOT PROVIDED

Catalog No.	Item	Quantity
E0277	EZ HDL/LDL™ Combination Calibrator	4x3 mL

OPTIONAL REAGENTS

Catalog No.	Item	Quantity
354-2	EZ HDL™ Reagent 2	20 mL