High-Density Lipoprotein Cholesterol (HDL-C) Content Assay Kit

Spectrophotometric Method

Product Code: 112773

Product Introduction

High-density lipoprotein (HDL) is one of the serum proteins, mainly synthesized by the liver. It transports cholesterol from peripheral tissues, where cholesterol is converted into bile acids or directly excreted through bile in the intestine. HDL is an anti-atherosclerotic plasma lipoprotein and an important reference indicator for the clinical diagnosis of coronary heart disease.

This kit uses a precipitant to separate HDL cholesterol in serum. Cholesteryl esters are hydrolyzed by esterase to produce free cholesterol (FC) and free fatty acids. Cholesterol oxidase then catalyzes the oxidation of FC to generate Δ4-cholestenone and H2O2. Finally, peroxidase catalyzes the reaction of H2O2 with 4-aminoantipyrine and phenol to form a red quinone compound with a characteristic absorption peak at 500 nm.

Package Contents

Pack Size	50T

Item Code	Description	Quantity
112773.1	Reagent I	1 bottle
112773.2	Reagent II	1 bottle
112773.3	Reagent III	1 bottle
112773.4	Reagent IV	1 bottle
112773.5	Standard	1 vial
112773.m	Instructions	1 copy

Quality Standards and Safety

Raw Material and Packaging Name	Quality Standard	Main Toxicity
Reagent I	—	—
Reagent II	—	—
Reagent III	—	—
Reagent IV	—	—
Standard	—	—

Handle all reagents using standard laboratory safety procedures.

Transportation and Storage

Transportation: Transport with ice packs.

Storage: Store Reagent III at -20°C. Store all other reagents at 2-8°C.

Shelf life: 180 days.

Instructions for Use

1. Sample Processing

1. After blood collection, separate the serum within 3 hours.
2. Mix the serum sample with Reagent I at a 1:1 ratio.
3. Mix thoroughly and incubate at 25°C for 15 min.
4. Centrifuge at 2000g for 15 min.
5. Place the supernatant on ice for testing.

2. Reagent Preparation

Immediately before use, add 15 mL of Reagent IV to Reagent III and dissolve completely.

3. Assay Procedure

Set up the assay as follows:

Component	Standard Tube	Test Tube
Standard (μL)	20	—
Reagent II (μL)	750	750
Reagent III (μL)	250	250
Sample (μL)	—	20

1. Mix well and incubate at 37°C for 30 min.
2. Use distilled water to zero the instrument.
3. Measure the absorbance at 500 nm in a 1 mL glass cuvette.
4. Record the absorbance of the standard tube as Astandard and the absorbance of the test tube as Asample.

4. Calculation

HDL-C content (mmol/L) = Asample ÷ Astandard × Cstandard

HDL-C content (mmol/L) = 5 × Asample ÷ Astandard

Cstandard = 5 mmol/L

Notes

1. Samples must not be repeatedly frozen and thawed. The assay is best completed within 12 hours after blood collection.
2. The linear range is 0.17 mmol/L-4.2 mmol/L.
3. Prepared Reagent III can be stored at 4°C and used for up to two weeks.
4. Before the formal assay, select 2-3 samples with large expected differences for pretesting.
5. Required instruments and supplies: centrifuge, constant-temperature water bath, visible spectrophotometer, 1 mL glass cuvette, and distilled water.
6. This 50T kit can assay 48 samples.