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ISO 13485:2016 ISO 9001:2015

Ver.25111

Total Cholesterol Assay Kit

BC9908-02 (60Tests)

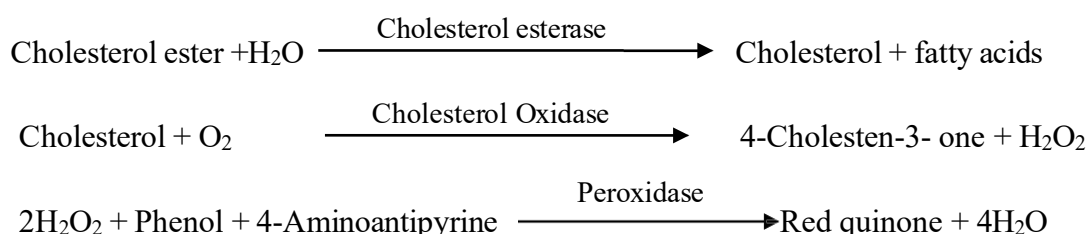
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Product Description

It is the main lipid found in the blood, bile and brain tissues. It is also one of the most important steroids of the body and is a precursor of many steroid hormones. Two thirds of cholesterol present in the blood is esterified. The liver metabolizes the cholesterol and it is transported in the blood stream by lipoproteins. Wavelength of absorbance is 630 nm.

Increased levels are found in hypercholesterolaemia, hyperlipidaemia, hypothyroidism, uncontrolled diabetes, nephritic syndrome and cirrhosis. Decreased levels are found in malabsorption, malnutrition, hyperthyroidism, anaemia & liver diseases.

Enzymatic colorimetric determination of total cholesterol according to the following reactions.



Kit components

Reagent	Volume	Storage
Extraction Reagent	1 × 60 mL	2-8°C
Cholesterol Reagent	1 × 60 mL	2-8°C
Cholesterol Standard	1 × 2 mL	2-8°C

Open Vial Stability

Once opened, the reagent is stable up to 4 weeks at 2-8°C, if contamination is avoided.

Reagent Deterioration

Turbidity or precipitation in any kit component indicates deterioration and the component must be discarded.

Reagent Preparation

Cholesterol Reagent, Standard and Extraction reagent are ready to use.

Precaution

- To avoid contamination, use clean laboratory wares use clean, dry disposable pipette tips for dispensing. Close reagent bottles immediately after use.
- Avoid direct exposure of reagent to light. Do not blow into the reagent bottles.

Reagents and Equipment Required but Not Provided

Analytical balance, mortar/homogenizer, low temperature centrifuge, water bath, adjustable pipette, spectrophotometer, 1 mL glass cuvette and distilled water

Operation Procedures

Sample Preparation

1. Bacteria or cells

Harvest the cells and wash twice with PBS. Ideal to use 5 million cells for the assay. Add 1mL Extraction Reagent to 5 million cells and ultrasonicate (200W, work time 3 second / interval 10 second repeat for 30 times) for complete lysis. Perform ultrasonication while keeping the cells in ice bath. Centrifuge at 8000 rpm, 4°C for 10 minutes and collect the supernatant. The supernatant should be kept on ice.

Note: Ideal proportion of Cells/Bacteria to Extraction Reagent is 1:5-10.

2. Tissue

Prepare 10% tissue homogenate by adding 1mL Extraction Reagent to 0.1g tissue. Grind completely to make a homogenate. Centrifuge at 8000 rpm, 4°C for 10 minutes and collect the supernatant.

3. Serum or Plasma

Directly use for the assay.

Interferences

No interference for

Bilirubin up to 10 mg/dL

Ascorbic acid up to 50 mg/dL

Haemoglobin up to 1000 mg/dL

Unit Conversion

Traditional Unit	SI Unit	Conversion from Traditional to SI
mg / dL	mmol/L	× 0.026

Procedure Notes

Reagent	Blank tube	Standard tube	Sample tube
Working Reagent	1000 µL	1000 µL	1000 µL
Standard	-	10 µL	-
Sample	-	-	10 µL

Mix & incubate for 5 minutes at 37° C. Read the absorbance of standard and sample against reagent blank. Wavelength of absorbance is 630 nm

Calculation

$$\text{Cholesterol Concentration (mg/dL)} = \frac{\text{Absorbance of sample}}{\text{Absorbance of standard}} \times 200$$

Performance

Linearity: Upto 600 mg/dL.

If the concentration is greater than linearity (600 mg/dL) dilute the sample with normal saline and repeat the assay. Multiply the result with dilution factor.

Sensitivity: Lower detection Limit is 3 mg/dL