

### SAFETY PRECAUTIONS AND WARNINGS :

This reagent is for *In vitro* diagnostic use only.

### INTENDED USE :

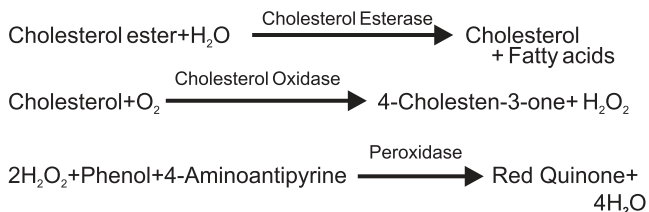
This reagent kit is intended for "*in vitro*" quantitative determination of Total Cholesterol concentration in serum. Enzymatic colorimetric method.

### CLINICAL SIGNIFICANCE :

The biosynthesis of Cholesterol predominantly takes place in the liver and in intestinal mucosa, but almost all cells synthesize it. It is a constituent of many membranes, it is also essential in the synthesis of bile acids and steroid hormones. It circulates in blood as cholesterol ester bound to beta lipoproteins. The measurement of the level of Cholesterol as well as Triglycerides and Lipoproteins is important in examining the metabolism of lipids. Changes in the level of Cholesterol mainly reflect disorders of liver function. Cholesterol level is increased in obstructive jaundice, diabetes mellitus and hypothyroidism. The level is decreased in some cases of hyperthyroidism and certain forms of anaemia. Identification of the different density fractions (HDL, LDL, VLDL) as well as total Cholesterol plays a role in the diagnosis.

### PRINCIPLE :

The Cholesterol esters of the sample are hydrolysed by Cholesterol esterhydrolase (ChEH). 4-Cholesten-3-one and H<sub>2</sub>O<sub>2</sub> are then formed from the released free Cholesterol by Cholesterol oxidase (ChOD). A measurable Red quinonimine derivative which absorbance light at 505 nm is formed from Hydrogenperoxide (H<sub>2</sub>O<sub>2</sub>) and 4-Aminoantipyrine in the presence of Phenol and peroxidase (POD).



### REAGENT COMPOSITION :

Reagent 1: Enzyme reagent  
 Cholesterol standard: 200 mg/dl

### MATERIALS REQUIRED BUT NOT PROVIDED :

- Clean & Dry Glassware.
- Micropipettes & Tips.
- Colorimeter or Bio-Chemistry Analyzer.

### SAMPLES :

Serum free of hemolysis, heparinised plasma or EDTA plasma.

### STABILITY OF REAGENT :

When Stored tightly closed at 2° to 8°C temperature protected from light and contaminations prevented during their use; reagents are stable up to the expiry date stated on the label.

### WORKING REAGENT :

The Reagent is ready for use.

### GENERAL SYSTEM PARAMETERS :

Reaction type	End Point (Increasing)
Wave length	505 nm (490 - 520) nm
Light Path	1 Cm
Reaction Temperature	37°C
Blank / Zero Setting	Reagent
Reagent Volume	1ml
Sample Volume	10 µl
Incubation Time	5 Minutes
Standard Concentration	200 mg/dl
Low Normal	109 mg/dl
High Normal	202 mg/dl
Linearity	1000 mg/dl

### ASSAY PROCEDURE :

	Blank	Standard	Sample
Reagent	1ml	1ml	1ml
Standard		10 µl	
Sample			10 µl

Mix and read the optical density (A) after a 5 - minute incubation.

### CALCULATION :

$$\text{Cholesterol Conc. (mg/dl)} = \frac{\text{OD of Sample}}{\text{OD of Standard}} \times \text{Conc. of Standard}$$

### LINEARITY :

Reagent is Linear up to 1000 mg/dl.

Dilute the sample appropriately and re-assay if Cholesterol concentration exceeds 1000 mg/dl. Multiply result with dilution factor.

### REFERENCE NORMAL VALUE :

109 - 202 mg/dl

### QUALITY CONTROL :

For accuracy it is necessary to run known controls with every assay.

### LIMITATION & PRECAUTIONS :

1. Storage conditions as mentioned on the kit to be adhered.
2. Do not freeze or expose the reagents to higher temperature as it may affect the performance of the kit.
3. Before the assay bring all the reagents to room temperature.
4. Avoid contamination of the reagent during assay process.
5. Use clean glassware free from dust or debris.
6. Do not use the reagent if it is hazy or cloudy.

### BIBLIOGRAPHY :

Tietz N.W Fundamentals of clin. Chem, Young D.S, Naito, HK.et.al. (1973), 10.79.

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