ESTROM Cholesterol (Lypho)
(CHOD-POD Method)

ANGSTROM BIOTECH PVT. LTD

The reagent is for quantitative estimation of Cholesterol in serum/plasma.

PRINCIPLE:
The cholesterol esters present in the sample are acted upon by cholesterol estrase to release cholesterol and fatty acids. The cholesterol is oxidized by cholesterol oxidase to yield cholestenon 4 – en 3- one and hydrogen peroxide as by product. Hydrogen peroxide together with 4 -Aminoantipyrine and phenolic compound in the presence of peroxidase gives the coloured complex. The intensity of the colour is proportional to the total cholesterol in the sample and is measured at 505 nm (490-530 nm) or with green filter.

CHOESTEROLESTER+ H_2O **CHOLESTEROL ESTERASE** CHOLESTEROL + FATTY ACIDS

CHOLESTEROL +O_2 **CHOLESTEROL OXIDASE** CHOLESTENON – 4EN – 3ONE + H_2O_2

H_2O_2 + PHENOL + 4 AMINOANTIIPYRINE **PEROXIDASE** RED QUINIONE + $4H_2O$

CLINICAL SIGNIFICANCE:
Important factors affecting the normal cholesterol levels are age stress, pregenancy and hormonal imbalance. Low levels of cholesterol are observed incase of acute hepatitis, malnutrition, anemias, hyperthyroidism and Gaucher"s disease. Increased levels of cholesterol is also found in coronary artery disease, diabetic mellitus, hypothyroidism, nephritic syndrome and hepatic malfunction like obstructive jaundice and cirrohisis of liver.

SPECIMEN COLLECTION AND STORAGE:
- : Fresh, fasting, unhemolysed serum is preferred.
- Plasma collected with heparin or EDTA as anti coagulant may be used.
- Samples are stable for 2 days when stored at 2-8°C.

PRECAUTION:
Cholesterol reagent is for In Vitro diagnostic use only

REAGENTS:
All the reagents are to be stored at 2-8°C

	No of Bottles		
	2x25 ml	2x50 ml	4x50ml
Reagent 1 (Enzyme Chromogen)	2	2	4
Reagent 2 (Phenol)	2	2	4
Std. (200 mg/dl)	1	1	1

REAGENT RECONSTITUTION:
A) 2x25 ml (B) 2x50 ml (C) 4x50 ml :
Transfer the contents of 1 vial of Reagent 1 (Enzyme Chromogen) to the bottle containing Reagent 2 (Phenol). Mix gently. Allow to dissolve for 10 minutes. Store at 2-8°C, protected from light when not in use.

REAGENT STORAGE AND STABILITY:
All the reagents are stable up to expiry date indicated on the bottle label. Working Reagent is stable for 3 months when stored at 2-8°C. Protect from light. A pink colour of working reagent (up to 0.2) due to aging of reagent does not affect the test results.

PROCEDURE:
Allow the sample and reagent to attain room temperature prior to use.

For 1 ml.

Dispense	Blank	Std.	Test
Working Reagent	1.0 ml	1.0 ml	1.0 ml
Standard	-	10 µl	-
Sample	-	-	10 µl

Incubate at 37° C for 5 Minutes. Mix well and read at 505 nm (490-530 nm) or against green filter

STABILITY OF REACTION MIXTURE:
The colour of the reaction mixture is stable for 30 minutes when stored at room temperature, protected from light.

LINEARITY:
This method is linear for Cholesterol values up to 750 mg/dl. For sample with values higher than 750 mg/dl, dilute the sample using normal saline and repeat the assay. Apply proper dilution factor while calculation.

CALCULATIONS:
Concentration of cholesterol

$$\text{in sample (mg/dl)} = \frac{\text{Abs of Test}}{\text{Abs of Std.}} \times 200$$

REFERENCE VALUE:
Fasting Serum/Plasma : 140 – 250 mg/dl

It is recommended that each laboratory establish its own reference values.

BIBLIOGRAPHY:

- Trinder P. Ann. Clin. Biochen, 6 (1969) 24.
- Allain C.A., et. Al., Clin. Chem., 20, (1974)

GENERAL INSTRUMENT PARAMETERS:

Reaction Type	: End Point
Slope of Reaction	: Increasing
Wavelength	: 505nm (490-530nm)
Flowcell Temperature	: 37° C
Reagent Volume	: 1.0 ml
Sample Volume	: 10 µl (0.01 ml)
Standard Concentration	: 200 mg/dl
Units	: mg/dl
Incubation	: 5 Minutes
Zero Setting	: Reagent blank
Path length	: 1.0 cm

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⚠	Attention,see instructions for use		Consult Instructions For Use
IVD	For in vitro diagnostic use only	REF	Catalog #
2°C 8°C	Store between 2-8°C	LOT	Lot Number
	Do not use if package is damaged		Date of Manufacturing
	Manufacturer		Use by