# CHOLESTEROL-L (Single Liquid)

**ENZOPAK** 

(CHOD-PAP, Enzymatic)

Last update 09-2020

Ref.

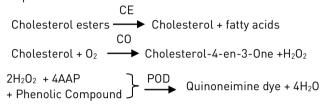
CC3-CLE.05M, 2x60 ml CC3-CLE.5MU, 5x60 ml CC3-CLE.5MV, 5x120 ml

#### INTENDED USE

Liquid Reagent for quantitative estimation of cholesterol in serum or plasma.

#### **PRINCIPLE**

The cholesterol esters are hydrolysed to free cholesterol by cholesterol esterase (CE). The free cholesterol is then oxidised by cholesterol oxidase (CO) to cholesten 4-en-3-one with the simultaneous production of hydrogen peroxide. The hydrogen peroxide reacts with 4-aminoantipyrine and phenolic compound in the presence of peroxidase to yield a coloured complex which is read at 505 nm.



The intensity of colour produced is directly proportional to the concentration of total cholesterol in the sample.

#### **PRESENTATION**

All reagents to be stored at 2-8°C	No. of	No. of bottles		
	2x60ml	5x60ml	5x120ml	
<ul> <li>Cholesterol-L (Ready to use)</li> </ul>	2	5	5	
<ul> <li>Cholesterol-Standard (200 mg/dl)</li> </ul>	1	1	1	

# FINAL REAGENT COMPOSITION

Active Ingredients Concentration • Cholesterol oxidase ≥500 U/L ≥600 U/L • Cholesterol Esterase • Peroxidase ≥6000 U/L • 4-Amino Antipyrine 0.5 mmol/L • Phenolic compound 20 mmol/L 10 mmol/L Surfactant • Buffer 100 mmol/L

pH 7.00+ 0.1 at 25°C

Cholesterol Standard (200 mg/dl)

Also contains non-reactive fillers and Stabilizers.

#### **PRECAUTION**

Cholesterol -L is for *in-vitro* diagnostic use. Reagent Contains Sodium Azide, DO NOT INGEST.

# PREPARATION OF WORKING REAGENT

Cholesterol-L reagent is Ready-to-use.

# REAGENT STORAGE AND STABILITY

Cholesterol-L reagents are stable at  $2-8^{\circ}\text{C}$  until the expiry date stated on the label.

# SPECIMEN COLLECTION

Fresh, clear serum with no hemolysis under fasting condition is specimen of choice. Plasma collected with Heparin or EDTA as anticoagulants can also be used.

#### REACTION PARAMETERS

• Type of Reaction : End Point

• Wavelength : 505 nm (505-530 nm)

• Flowcell Temperature :  $30 \, ^{\circ}\text{C} / 37 \, ^{\circ}\text{C}$ • Sample Volume :  $10 \, \mu\text{l}$ 

Incubation time : 30 min. R.T. / 10 min. 37° C

: 1.0 ml.

Std. Concentration : 200 mg/dl
 Light Path : 1.0 cm.
 Zero setting with : Reagent Blank

#### **TEST PROCEDURE**

Reagent Volume

Pipette In To Tubes	BLANK	STANDARD	TEST
Cholesterol Reagent (ml)	1.0	1.0	1.0
Standard (ml)	1	0.01	1
Sample (ml)	-	-	0.01

Mix well and incubate for 10 minutes at  $37^{\circ}$ C or 30 minutes at R.T. (25 ± 5°C) Read absorbance of standard and test at 505 nm (505-530) (Green filter) against reagent blank.

#### **TEST RESULTS**

Cholesterol Concentration (mg/dl) =  $\frac{\text{Absorbance of test}}{\text{Absorbance of Std.}} \times 200$ 

#### LIMITATIONS FOR INTERFERENCE

As per studies carried out for interference. Following results were obtained.

- No Interference from Hemoglobin upto 375 mg/dl.
- No Interference from free Bilirubin upto 7.5 mg/dl.
- No Interference from Lipemic (Measured as Triglycerides) upto 1000 mg/dl.

## **NORMAL VALUES**

140 to 250 mg/dl.

# **LINEARITY**

This procedure is linear upto 1000 mg/dl. For sample values higher than 1000 mg/dl, dilute the sample suitably with 0.9 % saline and repeat the assay. Apply dilution factor to obtain test results.

### NOTE

A special surfactant, Lipid Clearing Factor (L.C.F.) is added to the Reagent to solubilise the lipemic sera (causing turbidity or opalascence) which adds to the accuracy of results.

# **REFERENCES**

- 1. Allain, CC, Poon, L, Chan, S.G., Richmond, W., Fu, P. Enzymatic determination of total serum cholesterol, Clin. Chem. 20, 470 (1974)
- 2. Rochalu, B.Bernt, E. Gruber, W., Enzymatishe Bestimmung des Gesamt-Cholesterins in serum, Z. Klin. Chem. Klin Biochem 12,226 (1974)
- 3. Pescse, M.A. Bodourian, S.H. Interference with the enzymatic measurement of cholestrol in serum by use of five reagent kits. Clin. Chem. 23,757 (1977).





