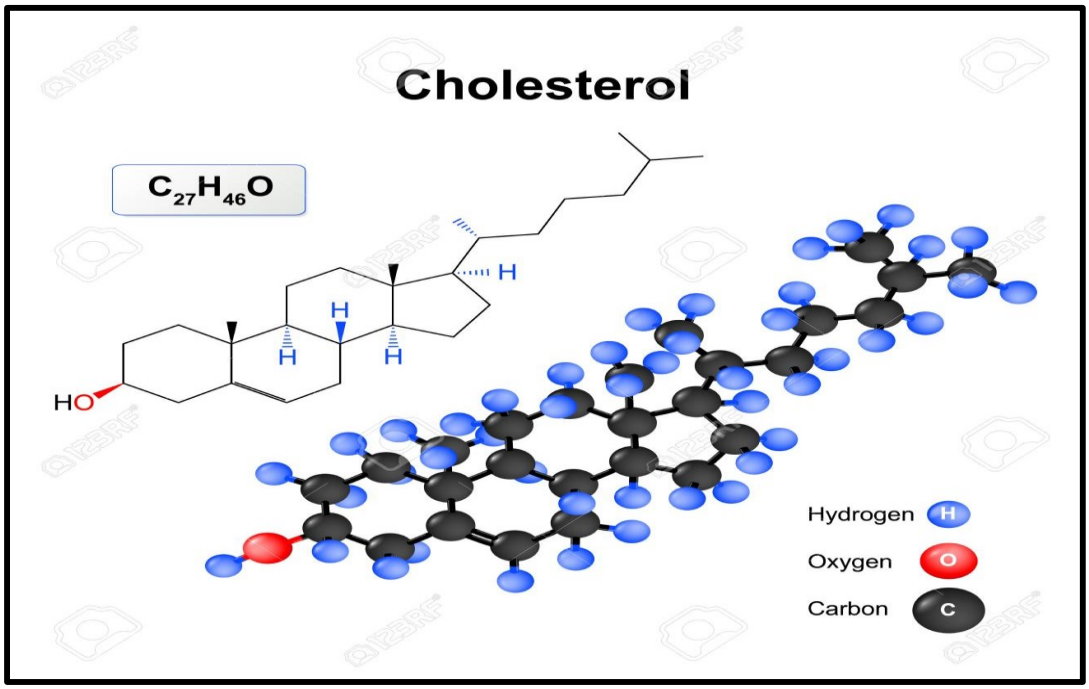
Measurement of serum cholesterol level

Cholesterol is a sterol that plays a central role in many biochemical processes and is vital for life and good health. In language: Stero- (rigid), -ol: (alcohol), chole- (yellow or pertaining to bile).

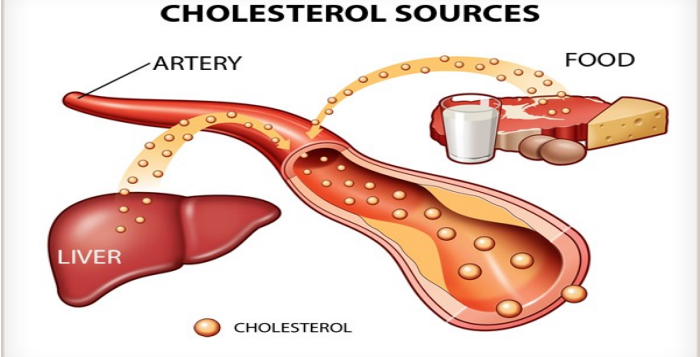


Sources of cholesterol:

Cholesterol comes from two sources:

1- Human body can make cholesterol. Liver is the major internal source but cholesterol can be made by all nucleated cells.

2- The remainder of the cholesterol in the body comes from foods derived from animals. For example, meat , full-fat dairy products , and egg yolk.



Functions of cholesterol:

- Cell membrane synthesis – Cholesterol helps to regulate membrane fluidity over the range of physiological temperatures. Lipid rafts are important components of cell membranes and are enriched in cholesterol, sphingomyelin and receptors. They are involved in various essential cellular processes, including endocytosis, exocytosis and cellular signaling.
- Cholesterol in the myelin sheaths – The nerve cells are covered with a protective layer or myelin sheath. The myelin sheath is rich in cholesterol.
- In the liver, cholesterol is converted to bile acids or salts which are then stored in the gallbladder. Bile is rich in bile salts that help in making the fats more soluble and so help in their absorption. Bile salts also aid in absorption of fat soluble vitamins like Vitamins A, D, E and K.
- Cholesterol is an important precursor molecule for the synthesis of vitamin D under the skin on exposure to UV ray.
- Cholesterol is an important precursor molecule for the synthesis of steroid hormones like corticosteroids, sex hormones like estrogen, progesterone and testosterone.

Carriers of cholesterol :

- **Low-density lipoprotein (LDL):** This carries cholesterol and transports it throughout the body and so cholesterol may accumulate up in the walls of the arteries, making them hard and narrow(atherosclerosis). This is why LDL-C is sometimes is called the “bad” cholesterol..

- **High-density lipoprotein (HDL):** This picks up excess cholesterol from tissues and takes it back to the liver. This is why HDL-C is sometimes called the carrier of “good” cholesterol.

Causes of Hypercholesterolemia :

Primary hypercholesterolemia :

- Familial hypercholesterolemia

Secondary hypercholesterolemia :

This may be associated with the following conditions:
1. Primary hypothyroidism
2. Nephrotic syndrome
3. Intake of certain drugs such as thiazide diuretics and β – blockers.
4. Long standing cholestasis

Hypocholesterolemia :

- **Liver disease**

The liver is a key organ for synthesis of cholesterol . A damage to the liver can cause a significant decrease in blood cholesterol level.

- **Hyperthyroidism**

Hyperthyroidism can lead to low cholesterol levels. Hyperthyroid patients show an enhanced excretion of cholesterol and an increased turnover of LDL resulting in a lower total and LDL cholesterol.

- **Malabsorption**

Some conditions that are characterized by malabsorption such as Celiac disease show a lower cholesterol levels in their blood.

Normal range of cholesterol :

Level mg/dL	Interpretation
< 200	Desirable level and correspond to a lower risk for heart disease
200–240	Borderline high risk
> 240	High risk

Measurement of serum cholesterol

Principle:

Cholesterol is measured enzymatically in serum through a series of coupled reactions that hydrolyze cholesteryl esters and oxidize the 3-OH group of cholesterol. One of the reaction byproducts, H_2O_2 is measured quantitatively in a peroxidase-catalyzed reaction that produces a purple color. Absorbance is measured at 500 nm. The color intensity is proportional to cholesterol concentration. The reaction sequence is as follows:

$$\text{Esters of cholesterol} + H_2O \xrightarrow{\text{Cholesterol esterase}} \text{Cholesterol} + \text{Fatty acid}$$

$$\text{Cholesterol} + O_2 \xrightarrow{\text{Cholesterol oxidase}} \text{Cholest -4-enone} + H_2O_2$$

$$H_2O_2 + \text{Phenol} + \text{4- aminophenazone} \xrightarrow{\text{Peroxidase}} \text{Quinonimine}$$

- **Procedure:**

1- Pipette into test tube :

	Blank	Standard	Sample
Working reagents (ml)	**2.0**	**2.0**	**2.0**
Standard (µL)	**------**	**20**	**------**
Sample (µL)	**------**	**------**	**20**

2- Mix and incubate for at 10 min in 37 ºC.

3- Read the absorbance of the sample and standard against blank at a wave length of 500 nm.

- **Calculations:**

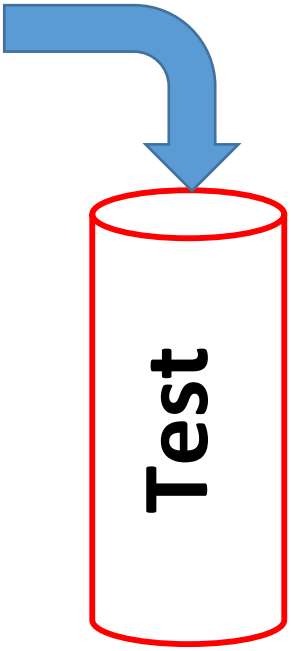
Cholesterol conc. mg/dl = (Absorbance of sample/absorbance of standard) × conc. Of standard

➢ Conc. of standard = 200 mg/dl

2 ml Working Reagent

+

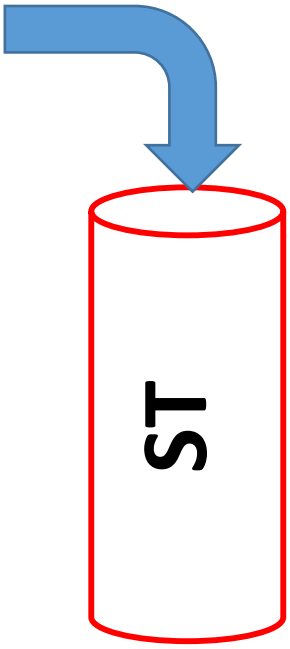
20 µl Serum



2 ml Working Reagent

+

20 µl Standard



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