

# Determination of serum cholesterol level

NARJES ALI AL-RASHE

Supervisor Amani Alghamdi

King Saud University

**Objective:**

TO determine serum cholesterol level.

## **Introduction:**

Cholesterol is a member of a large group of substances called steroid, which include vitamin D. Cholesterol is an essential component of cell membrane, brain and nerve cells, and bile, which helps the body absorb fats and fat soluble vitamins. The body uses cholesterol to make vit.D and various hormones, such as estrogen, testosterone, and cortisol. The body can produce all the cholesterol that it needs, but it also obtains cholesterol from food.

Lieberman-burchard test: The Lieberman-Burchard or acetic anhydride test is used for the determination of cholesterol. The formation of a green or green-blue color after a few minutes is positive. Lieberman-Burchard is a reagent used in a colorimetric test to detect cholesterol, which gives a deep green color. This color begins as a purplish, pink color and progresses through to a light green then very dark green color. The color is due to the hydroxyl group (-OH) of cholesterol reacting with the reagents and increasing the conjugation of the un-saturation in the adjacent fused ring. Because this test uses acetic anhydride and sulfuric acid as reagents caution must be exercised so as not to receive severe burns.

## **Material:**

1- cholesterol reagent (Acetic anhydride + acetic acid )

2- Sulphuric acid 95- 97 %

3-Standard cholesterol (300 mg/dl)

4-Samples

5-Test tubes

6-Pipettes

7-Cuvettes

8-Spectrophotometer

9-Water bath

## Method:

7 test tubes were labeled, (A, B) for test 1, (C, D) for test 2, (E, F) for the standard, and (G) for the blank, the following was added:

	(A,B)	(C,D)	(E,F)	(G)
Sample 1	0.1 ml			
Sample 2		0.1 ml		
Standard Cholesterol			0.1 ml	
Distilled water				0.1 ml
Cholesterol Reagent	4 ml	4 ml	4 ml	4 ml

The tubes were incubated at room temperature for 20 minutes. Then 1.0 ml of sulphuric acid was added to each tube. Incubation in a water bath at room temperature for 5 minutes. Removed from the water bath and shake vigorously. Then after 10 minutes the absorbance was measured for the samples against the blank at 610 nm.

## Result:

Result sample	Absorbance at 610 nm
Sample	
A	0.222
B	0.189
Sample	
C	0.322
D	0.386
Standard	
E	0.237
F	0.439
G	0

## CALCULATION:

(Absorbance of sample \ Absorbance of standard)  $\times$  300

1-  $\frac{A+B}{2}$  ,  $\frac{0.222+0.189}{2} = \frac{0.411}{2} = 0.2055$  SAMPLE 1.

2-  $\frac{C+D}{2}$  ,  $\frac{0.322+0.386}{2} = \frac{0.708}{2} = 0.354$  SAMPLE 2 .

3-  $\frac{E+F}{2}$  ,  $\frac{0.237+0.439}{2} = \frac{0.676}{2} = 0.338$  STANDARD.

(Sample 1 \ Standard)  $\times$  300 =  $(\frac{0.2055}{0.338}) \times 300 = 182.39$   
mg\dl

(Sample 2 \ Standard)  $\times$  300 =  $(\frac{0.354}{0.338}) \times 300 = 314.2$  mg\dl

The normal level of cholesterol is 150 -250 mg\dl .

## **DISSCSION:**

In these experiment we study the measure of serum cholesterol level and compare with the normal level.

We have good and bad cholesterol, the good cholesterol called high density lipoprotein (HDL), a high level of HDL in blood is associated with decreased risk of cardio vascular disease.

The bad cholesterol called low density liopprotein (LDL), a high level of cholesterol are related to CHD which has high concentration of cholesterol.

In these experiment we have two sample and I would to compare with the normal range, the normal level various with age and diet, the average is 150 -250 mg \dl .

Sample 1 =182.39 these is normal cholesterol, and the anther sample (sample 2) =314.2 when we compare these value with the normal level the patient have high cholesterol.

The patient may have: diabetes mellitus (low insulin level), hypothyroidism or obstructive jaundice.

If the value is low compared with the normal the patient may have: sever infection, massive cell damage, severe anemia or genetic hypolipoproteinemia.

## QUESTION:

1) You must make sure that all glass ware used in the experiment is dry. Why ?

-To avoid dilution of solution or if the material uses in the experiment is conc. May react with water.

2) Yellow serum is very dangerous. Why?

Yellow serum detect to the patient may have diabetes mellitus (high cholesterol).