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Determination of serum triglycerides(TAG)

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Objective:

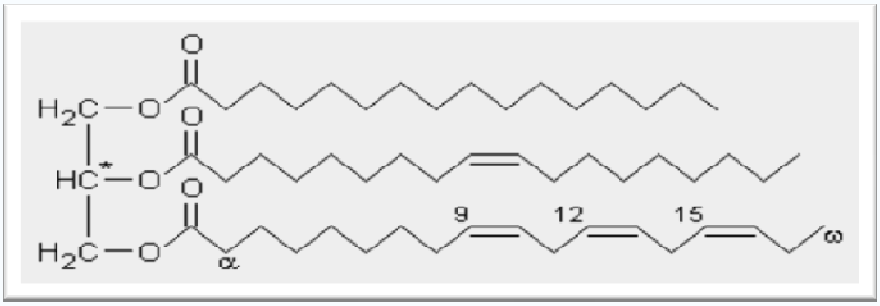
To determine the level of TAG in a serum or plasma sample.

¼ / ¼ Objective

1 ¼ / 1 ¼ introduction

Introduction:

TAG is a glyceride in which the glycerol is esterified with three fatty acids. It is the main constituent of vegetable oil and animal fats. Triacylglycerol is essentially confined to fat tissue.TAG is the most abundant lipid species, and the only one with an important role in energy metabolism.



Figer (1):General structure of a triglyceride

TAG is important in human metabolism in two points: 1- A significant fraction of our caloric intake is triacylglycerol.2-As an endogenous storage of metabolic energy. In the human body, high levels of triglycerides in the bloodstream have been linked to atherosclerosis, and, by extension, the risk of heart disease and stroke. However, the relative negative impact of raised levels of triglycerides compared to that of LDL: HDL ratios is as yet unknown. The risk can be partly accounted for by a strong inverse relationship between triglyceride level and HDL-cholesterol level. Another disease caused by high triglycerides is pancreatitis.

In the measurement of TAG concentration in blood we use a blank, standard, 2 samples. When TAG employed enzymatic reaction the (Quinoneimine Dye) appear and by measurement of the absorption at 505nm the concentration of TAG proportional.

Triglycerides are hydrolized by lipoproteinlipase to produce glycerol and free fatty acids. The glycerol participates in a series of coupled enzymatic reactions, in which glycerol kinase / glycerol phosphate oxidase are involved and H2O2 is generated. The H2O2 reacts with p-chlorophenol and 4-aminoantipyrine in the presence of peroxidase to form a quinoneimine dye. The intensity of color formed is proportional

to the triglycerides concentration and can be measured photometrically between 480 and 520 nm.

Material and method:

Material:

4 tubes.

Tg buffer reagent.

Tg enzyme reagent.

Tg standard (200mg/dl) .

Spectrophotometer.

Cuvette.

Pipettes.

Water bath.

Method:

4 tubs was labeled as the table

2.5 ml Reconstituted Reagent was added to all tubes, then these tubes were warmed at 37 C° for five minutes. After that standard, sample 1 and 2 were added as indicate in table 1.

After minutes red color appears, then the absorbance was measured at 505 nm.

Table 1: show the amount of reagent added to each test of the experiment.

	Blank	Standard	Test 1	Test 2
Reconstituted Reagent	2.5 ml	2.5ml	2.5ml	2.5ml
Standard	--------	0.025 ml	--------	------
Sample 1	--------	----------	0.025ml	---------
Sample 2	--------	----------	----------	0.025 ml

Result and calculations:

1 ½ / 1 ½ Result and calculations

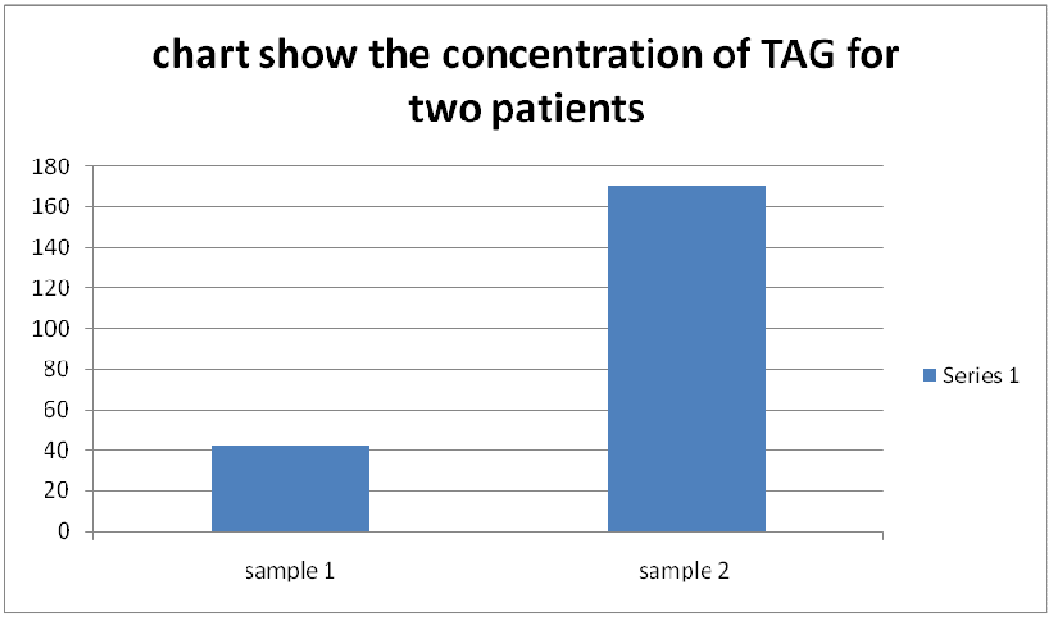
Table 2: show the absorbance reading for experiment test.

Tube	Absorbance
Blank	0.000
Standard	0.257
Test1	0.055
Test2	0.219

TAG concentration = (Absorbance of test/Absorbance of standard)× 200

Test 1= 42.80 mg/dl

Test2 = 170.42 mg/dl



2 / 2 discussion

Discussion:

The first test has a normal TAG concentration. But the second test has pretty high TAG concentration which may lead to: Cardiovascular Diseases, Atherosclerosis, Hypertriglyceridemia, Obesity, Heart Diseases, and Metabolic Syndrome X.

Question:

- Why is the triglyceride level higher in plasma than in serum?

Because plasma has the lipo proteins which transport TAG in body.

- How would the state of nutrition and general metabolic state of a person affect the serum triglyceride level?

When the diet has high calories than the body burn it will cause high level of TAG in serum.