

PRODUCT CODE

CS016

INTENDED USE

For the quantitative determination of Triglyceride in serum or plasma

CLINICAL SIGNIFICANCE

Triglycerides are simple lipids, formed in the liver by glycerol & fatty acids. They are transported by VLDL, LDL & constitute about 95% of fat, stored as source of energy in the tissue & plasma. Increased levels are found in hyperlipidemias, diabetes, nephrotic syndrome & hypothyroidism. Increased levels are risk factor for arteriosclerotic coronary disease, peripheral vascular disease, acute pancreatitis & hyperlipoproteinaemia. Decreased levels are found in malnutrition & hyperthyroidism.

PRINCIPLE

Lipases catalyze hydrolysis of triglycerides to yield glycerol and free fatty acids. The glycerol concentration is determined enzymatically with the Trinder reaction using glycerol kinase (GK), glycerol-3-phosphate oxidase (GPO) and peroxidase (POD). The end product is a Quinonimine dye, the concentration of which at 546 nm is directly proportional to the concentration of triglyceride in the sample.

Triglyceride +H₂O ----LPL-----> Glycerol + Free fatty acids

Glycerol+ATP ----GK-----> Glycerol-3 phosphate+ADP

Glycerol-3 phosphate +O₂ ----GPO-----> H₂O₂+Dihydroxyacetone phosphate

H₂O₂+Aminoantipyrine+ chlorophenol ----POD-----> Quinonimine dye

REAGENT COMPOSITION

- 1- Enzyme concentrate (Reagent-1)
- 2- Buffer (Reagent-2)
- 3- Triglyceride standard 200mg/dL or 2.26 mmol/L

When combined as instructed, the working reagent contains the following,

Pipes buffer (pH 7.50)	40 mmol/L
Lipases	150 KU/L
Glycerol Kinase (GK)	0.4 KU/L
Glycerol-3-phosphate oxidase (GPO)	1.5 KU/L
Peroxidase (POD)	0.5 KU/L
Magnesium	5.0 mmol/L
Adenosine Tri Phosphate (ATP)	1.0 mmol/L
Aminoantipyrine	5.0 mmol/L
Stabilizers and preservatives	0.4 mmol/L

REAGENT PREPARATION

To prepare working reagent, dilute 1 part of Reagent 1 (Enzyme concentrate) with 100 parts of Reagent 2 (buffer), e.g.: 1 mL / 100mL, 100µL/10mL. Mix gently and allow equilibrating to room temperature before use.

REAGENT STORAGE AND STABILITY

Both reagent and standard are stable until the expiry date when stored at 2-8°C.

The working reagent is stable for 3 weeks at 2-8°C.

SPECIMEN

Serum, Plasma: Fresh, clear & non haemolysed, from patients fasting at least 12 hours, Triglycerides in serum are stable for 3 days at 2-8° C, with prolonged storage at room temperature, glyceride-containing compounds hydrolyze to yield free glycerol with an apparent increase in triglyceride levels.

PRECAUTION

To avoid contamination, use clean laboratory wares. Avoid direct exposure of reagent to light.

ASSAY

Wavelength	546nm
Cuvette	1 cm light path
Temperature	25°C or 37°C
Measurement	Against reagent blank

PROCEDURE

	Blank	Standard	Sample
Working reagent	1000 µL	1000 µL	1000 µL
Standard	--	10 µL	--
Sample	--	--	10 µL

Mix and incubate for 10 minutes at 25°C or 5 minutes at 37°C. Measure the absorbance of the sample (As) and the standard (Astd) against the reagent blank within an hour.

CALCULATION

$$\text{Triglyceride Conc. (mg/dL)} = \frac{\Delta A \text{ sample}}{\Delta A \text{ standard}} \times 200 \text{ (Std.conc.)}$$

To convert mg/dL to mmol/L, divide by 88.50

Linearity:

The test is linear up to triglyceride concentration of 1000 mg/dl (11.3mmol/l). Sample with higher values should be diluted 1+5 with physiological saline (0.9 % NaCl), multiply the values by 6.

NORMAL RANGE

36 – 165 mg/dl; 0.4 – 1.86 mmol/l; > 200 mg/dl elevated. It is strongly recommending each laboratory establish its own normal range.

QUALITY CONTROL

All control sera with Triglyceride values estimated by this method may be used.

NOTES

- 1- The buffer contains 0.1% sodium azide so avoid contact with skin.
- 2- It is suggested that 10 mg/dl be subtracted from the triglyceride result to compensate for any free glycerol in the sample.
- 3- Keep reagents and samples out of direct sunlight.

SYMBOL ON LABELS

Symbols	Signify	Symbols	Signify
	Catalogue Number		Pack Size
	Expiry Date		Volume
	Storage Condition		Lot Number
	Instruction for Use		In Vitro Diagnostics
	Manufacturing Date		Manufacturer
	Number of Tests		For Single Use Only
	EC Representative		European conformity

BIBLIOGRAPHY

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