



TRIGLYCERIDES

GPO - PAP Method

Cat.No. 101-0241

Size 5 x 20 ml

Cat.No. 101-0016

Size 4 x 50 ml

Cat.No. 101-0268

Size 12 x 50 ml

Cat.No. 101-0052

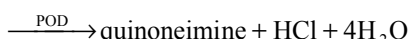
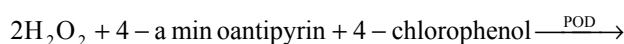
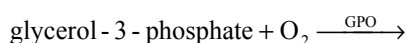
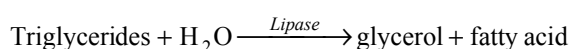
Size 6 x 100 ml

Cat.No. 101-0053

Size 4 x 250 ml

PRINCIPLE:

Glycerol produced by enzymatic hydrolysis of triglycerides is phosphorylated by ATP to produce glycerol-3-phosphate and ADP in the reaction catalyzed by glycerol kinase (GK). Glycerol-3-phosphate oxidase (GPO) catalyzes the oxidation of the glycerol-3-phosphate to produce dihydroxyacetone phosphate and H₂O₂. In further a reaction quinoneimine is formed from H₂O₂, 4-aminoantipyrine and 4-chlorophenol under the catalytic influence of peroxidase (POD). The intensity of the color formed is proportional to the concentration of glycerol in the reaction mixture and hence to the level of triglycerides in the sample.



SAMPLE:

Serum, heparinized or EDTA plasma.

Stable 2 weeks at +2 °C to +8 °C.

REAGENTS:

	Concentration in the test
1. Buffer	
Pipes buffer, pH 7.2	50.0 mmol/L
p-chlorophenol	2.0 mmol/L
2. Enzyme reagent	
4-aminoantipyrine	0.7 mmol/L
ATP	0.3 mmol/L
Glycerol-kinase (GK)	800 U/L
Glycerol-3-phosphate oxidase (GPO)	4000 U/L
Lipase	150000 U/L
Peroxidase (POD)	440 U/L
3. Standard	
Triglycerides	Standard concentration see on the vial label

PREPARATION OF REAGENTS:

Reconstitute the contents of vial 2 (enzyme reagent) with the corresponding volume of buffer. Stable for 60 days at +2 °C to +8 °C or 14 days at +15 °C to +25 °C stored in the dark bottle.

PROCEDURE:

Wavelength:	505 nm (490 - 550 nm)
Cuvette:	1 cm light path
Temperature:	25 °C, 37 °C
Color stability:	30 min
Zero:	reagent blank

Pipette into test tubes	Reagent blank	Standard	Sample
Standard	-	10 µl	-
Sample	-	-	10 µl
Working reagent	1000 µl	1000 µl	1000 µl

Mix and incubate for 10 min. at room temperature or 5 min at 37 °C. Avoid exposure to direct sunlight. Measure the absorbance of the standard and sample against the reagent blank within 30 minutes.

NOTE:

Volumes can be proportionally changed.

CALCULATION:

$$\frac{A_{\text{sample}}}{A_{\text{standard}}} \times \text{stand.conc.} = \text{mmol/L Triglycerides}$$

EXPECTED VALUES:

Normal values	up to 1.71 mmol/L (150 mg/dl)
Suspected above	> 1.71 mmol/L (> 150 mg/dl)
Increased above	> 2.28 mmol/L (> 200 mg/dl)

LINEARITY

up to 11.4 mmol/L (1000 mg/dl)

QUALITY CONTROL:

CONTRO-N	20 x 5 ml	Cat. No. 101-0083
CONTRO-P	20 x 5 ml	Cat. No. 101-0084

NOTE:

- If the triglycerides concentration >11.4 mmol/L (1000 mg/dl), dilute sample 1:2 with physiological solution and repeat the assay (result x 2).
- To correct for free glycerol, subtract 0.11 mmol/L from the triglycerides value (triglycerides mmol/L - 0.11mmol/L).
- Hemoglobin concentration >2 g/L and bilirubin concentration >340 µmol/L influence the results of the triglycerides.
- Reagent contain sodium azide as stabilizer. Do not swallow. Avoid contact with the skin and mucous membranes.

REFERENCES:

- Young, D., Pestaner, L. Clin. Chem. 21, 5 (1975).
- Fossati, P., Principe, L. Clin. Chem. 28, 2077 (1982).