

# Triglycerides - Liquizyme GPO-PAP (Single Reagent)

#### Intended Use

Spectrum Diagnostics liquizyme Triglycerides reagent is intended for the in-vitro quantitative, diagnostic determination of triglycerides in human serum on both automated and manual systems.

### **Background**

Triglycerides are the main lipids present in the human plasma; the others are the cholesterol, phospholipids and nonesterified fatty acids. They are formed in the intestinal mucosa by the esterification of glycerol and fatty acids. Triglycerides measurements are used in the diagnosis and treatment of patients with diabetes mellitus, liver obstruction, nephrosis and other diseases involving lipid metabolism. The measurement of serum triglycerides is important in the diagnosis of hyperlipoproteinemia and in the prediction, detection and monitoring of atherosclerosis.

#### Method

GPO-PAP-enzymatic colorimetric method.

# **Assay Principle**

The series of the reaction involved in the assay system is as follows:

1. Triglycerides are hemolyzed by lipoprotein lipase (LPL) to glycerol

Glycerol + Fatty acids Triglycerides LPL

2. Glycerol is then phosphorylated to glycerol-3-phosphate by ATP in a reaction catalyzed by glycerol kinase (GK).

Glycerol + ATP GK Glycerol-3-phosphate + ADP

3. The oxidation of glycerol-3-phosphate is catalyzed by glycerol phosphate oxidase (GPO) to form dihydroxyacetone phosphate and hydrogen peroxiode (H2O2).

Glycerol-3-phosphate GPO Dihydroxyacetone phosphate

4. In the presence of peroxidase (POD), hydrogen peroxide effects the oxidative coupling of 4-chlorophenol and 4-aminoantipyrine (4AAP) to form a red color quinoneimine dye which is measured

# Reagents

Standard Triglycerides (ST) 200 mg/dL 2.29 mmol/L

# Reagent (R)

safety data sheet.

Pipes Buffer pH 7.0	50 mmol/L
4-chlorophenol	6.0 mmol/L
Magnesium aspartate	>0.5 mmol/L
Lipase	>10 K U/L
Peroxidase	>2.0 KU/L
4-Aminoantipyrine	1.0 mmol/L
Glycerol-3-phosphate oxidase	>3.5 K U/L
Glycerol kinase	>750 U/L
ATP	1.0 mmol/L
Sodium Azide	8.0 mmol/L
For further information, refer to the Triglycerides	s reagent material

SYMBOLS IN PRODUCT LABELLING

ECREP Authorised Representative 

Use by/Expiration Date Batch Code/Lot number Catalogue Number Consult instructions for use X (Xi) - Irritant Temperature Limitation

Manufactured by

# **Precautions and Warnings**

Do not ingest or inhalate. In case of contact with eyes or skin; rinse immediately with plenty of soap and water. In case of severe injuries; seek medical advice immediately.

Reagent (R) contains sodium azide which may react with copper or lead plumbing.

# Reagent Preparation, Storage and Stability

Spectrum triglycerides reagents are supplied ready-to-use and stable up to the expiry date labeled on the bottles when properly stored refrigerated at ( 2 – 8  $^{\rm O}$ C). Once opened, the reagent and the standard are stable for 3 months at the specified temperature if contamination is avoided.

#### Deterioration

The reagent is normally clear or pale pink. Do not use liquizyme triglyceride reagent if it is turbid or if the absorbance is greater than 0.2 at 546 nm.

#### Specimen Collection and Preservation

Patients should be fasting for 10 to 14 hours before blood is drawn. Samples must be drawn in a soap and glycerol free collection device. Recommended anticoagulats are EDTA or heparin at levels of 1mg and 0.2 mg/dl whole blood, respectively.

Triglycerides in serum samples remain stable for 7 days at 4  $^{\rm oC}$ , for 3 months at -20  $^{\rm oC}$ , and for years at -70  $^{\rm oC}$ .

# **System Parameters**

Wavelength Hg 546 nm (500 - 550 nm) Optical path Assay type End-point Increase 1:100 1 ml Direction Sample : Reagent Ratio e.g.: Reagent volume Sample volume 10 μl 15 – 25 °C or 37 °C Temperature Zero adjustment Reagent blank 10 minutes at 15 – 25 °C or 5 minutes at 37 °C Incubation time Reagent Blank Limits Low 0.00 AU High 0.2 AU

5 mg/dL (0.057 mmol/L) 1000 mg/dL (11.45 mmol/L) Sensitivity Linearity

### **Procedure**

	Blank	Standard	Sample
Reagent Standard	1.0 ml	1.0 ml 10 นl	1.0 ml
Sample			10 μl

Mix and incubate for 5 minutes at 37  $^{\circ}$ C or 10 minutes at 15 – 25 $^{\circ}$ C. Measure absorbance of specimen (Aspecimen) and standard (<sup>A</sup>standard) against reagent blank within 30 minutés

### Calculation

Serum Triglycerides conc. (mg/dL) =

A<sub>specimen</sub> x 200 A<sub>standard</sub>

# **Quality Control**

Normal and abnormal commercial control serum of known concentrations should be analyzed with each run.

#### **Performance Characteristics**

#### Precision

Within run (Repeatability)

	Level 1	Level 2
n	20	20
Mean (mg/dL)	155.1	245.8
SD	2.03	1.85
CV%	1.31	0.75

#### Run to run (Reproducibility)

	Level 1	Level 2
n	20	20
Mean (mg/dL)	156	246.5
SD	2.2	1.9
CV%	1.41	0.77

# **Methods Comparison**

A comparison between Spectrum Diagnostics Triglycerides reagent and a commercial reagent of the same methodology was performed on 20 human sera. A correlation of 0.976 was obtained.

#### Sensitivity

When run as recommended, the minimum detection limit of the assay is 5 mg/dL (0.057 mmol/L).

#### Linearity

The reaction is linear up to triglycerides concentration of 1000 mg/dL; specimens showing higher concentration should be diluted 1+1 using physiological saline and repeat the assay (result × 2).

# **Interfering Substances**

### Haemolysis

No significant interference up to a haemoglobin level of 6.0 g/L (0.36 mmol/L).

Bilirubin levels higher than 171 µmol/L (10 mg/dL) decrease the apparent Triglycerides concentration significantly.

Of the drugs tested in-vitro, methyldopa and levodopa cause artificially low Triglycerides values at the tested drug Level.

Physiological ascorbic acid concentration does not interfere with the test. Ascorbic acid levels higher than 114  $\mu$ mol /I (2 mg /dL) decrease the apparent Triglycerides concentration significantly.

# **Expected Values**

(0.4 – 1.54 mmol/L) (0.45 – 1.82 mmo/L) 35 -135 mg/dL **Females** 40 -160 mg/dL

The following limits are recommended for the recognition of the risk factor hypertriglyceridemia:

Suspicious above 150 mg/dL (1.71 mmol/L) Elevated above 200 mg/dL (2.28 mmol/L)

Spectrum Diagnostics does not interpret the results of a clinical laboratory procedure; interpretation of the results is considered the responsibility of qualified medical personnel. All indications of clinical significance are supported by literature references.

### **Dynamic Range**

5 - 1000 mg/dL (0.057 - 11.45 mmol/L)

# **Waste Disposal**

This product is made to be used in professional laboratories. Please consult local regulations for a correct waste disposal. **\$56**: dispose of this material and its container at hazardous or special waste collection point.

\$57: use appropriate container to avoid environmental contamination. S61: avoid release in environment. refer to special instructions/safety data sheets.

#### References

- Bucolo G, David H: Quantitative determination of serum triglycerides by the use of the enzymes. Clin Chem 19: 475,
- Chowdhury RF, Rodman H, Bleicher SJ: Glycerol like contamination of commerical blood sampling tubes. J Clin Pathol 12: 116. 1971
- 3. MGowan MW, Artiss JD, Standbergh DR, Zak B. A peroxidasecoupled method for colorimetric determination of serum triglycerides. Clin Chem;29:538-452 ;1983.
- Stein EA; Lipids , lipoproteins, and apolipoproteins. In : NW Tietz, ed. Fundamentals of clinical chemistry, 3 rd ed. Philadelphia : WB Saunders; 448 ; 1987.
- Tietz NW, Boden T, Stepleton JD : An improved method for the determination of lipase in serum. Am J Clin Pathol 31: 148, 1959 6. Young DS et al, Clin Chem. 21; 1975

ORDERING INFORMATION			
CATALOG NO.	QUANTITY		
314 001 314 002 314 003 314 004 314 005 314 006 314 007 314 008 314 009 314 010	2 x 25 ml 4 x 25 ml 4 x 30 ml 10 x 15 ml 4 x 50 ml 4 x 60 ml 4 x100 ml 5 x100 ml 8 x100 ml 4 x250 ml		



Egyptian Company for Biotechnology (S.A.E)
Obour city industrial area. block 20008 piece 19 A. Cairo. Egypt.
Tel: +202 4489 2248 - Fax: +202 4489 2247

www.spectrum-diagnostics.com

E-mail:info@spectrum-diagnostics.com





