Spectrum For Diagnostic Industries

Triglycerides

IVD

REF.	Pack size
146 01 050	(1 x 50 ml) 50 tests
146 02 030	(2 x 30 ml) 60 tests
146 05 030	(5 x 30 ml) 150 tests

Intended Use

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

Introduction

Triglycerides are the most prevalent dietary glycerol esters encountered. They also constitute 95 % of the fat stored in tissue and are the predominant form glycerol ester found in plasma. Their fatty acids residues vary considerably but usually include combinations of the long-chain fatty acids.

Method

GPO-PAP-enzymatic colorimetric method.

Principle

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

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

Triglycerides $\xrightarrow{LPL}$ Glycerol + Fatty acids

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

Glycerol + ATP $\xrightarrow{GK}$ 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 (H_2O_2).

Glycerol-3-phosphate + O_2 $\xrightarrow{GPO}$ Dihydroxyacetone phosphate + H_2O_2

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 at 546 nm.

2 H_2O_2+4-APP + 4-chlorophenol $\xrightarrow{POD}$ Quinoneimine dye + $4H_2O$

Reagents

Reagent (R)

Pipes Buffer	50 mmol/L
4-chlorophenol	6.0 mmol/L
Magnesium aspartate	>0.5 mmol/L
LPL	>10 KU/L
POD	>2.0 KU/L
4-APP	1.0 mmol/L
GPO	>3.5 KU/L
GK	>750 U/L
ATP	1.0 mmol/L
Azide	8.0 mmol/L

Standard

200 mg/dL	2.29 mmol/L

Reagents preparation, storage and stability

Triglycerides reagents are supplied ready-to-use and stable till the expiration date stated on the bottles when properly stored refrigerated at 2 – 8 °C. Once opened, the reagent and the standard are stable for 3 months at the specified temperature.

Deterioration

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

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.

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 anticoagulants are EDTA or heparin at levels of 1 mg and 0.2 mg/dL whole blood, respectively.

Triglycerides in serum samples remain stable for 7 days at 4 °C, for 3 months at -20 °C, and for years at -70 °C.

Procedure

Wavelength	546 nm
Optical path	1 cm
Assay type	End-point
Direction	Increase
Sample : Reagent Ratio	1 : 100
Temperature	15 – 25 °C or 37 °C
Zero adjustment	Reagent blank
Reagent Blank Limits	Low 0.00 AU High 0.2 AU
Incubation time	10 minutes at 15 – 25 °C or 5 minutes at 37 °C

	Blank	Standard	Specimen
Reagent	1.0 ml	1.0 ml	1.0 ml
Standard	-------	10 µl	-------
Specimen	-------	-------	10 µl

Mix and incubate for 5 minutes at 37 °C or 10 minutes at 15 – 25°C. Measure absorbance of specimen ($A_{specimen}$) and standard ($A_{standard}$) against reagent blank .

Calculation

$$\text{Serum triglycerides conc. (mg/dL)} = \frac{A_{specimen}}{A_{standard}} \times 200$$

Quality control

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

Expected Values

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

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)

Performance characteristics

A comparison between Triglycerides reagent and a commercial reagent of the same methodology was performedon 20 human sera.
A correlation of 0.967 was obtained.

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%	1.4	0.87

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).

Interference

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

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

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

Others
Physiological ascorbic acid concentration doesn't interfere with the test. Ascorbic acid levels higher than 114 µmol /L (2 mg /dL) decrease the apparent triglycerides concentration significantly.

Waste Disposal

This product is made to be used in professional laboratories. Please consult local regulations for a correct waste disposal.
S56: dispose of this material and its container at hazardous or special waste collection point.
S57: use appropriate container to avoid environmental contamination.
S61: avoid release in environment. refer to special instructions/safety data sheets.

References

1. Young DS et al, Clin Chem
2. Chowdhury RF, Rodman H, Bleicher SJ : Glycerol like contamination of commerical blood sampling tubes. J Clin Pathol .
3. Tietz NW, Boden T, Stepleton JD : An improved method for the determination of lipase in serum

SYMBOLS IN PRODUCT LABELLING

IVD	For in-vitro diagnostic use
LOT	Batch Code/Lot number
REF	Catalogue Number
	Consult instructions for use
	Temperature Limitation
	Use by/Expiration Date
	CAUTION. Consult instructions for use
	Manufactured by

Spectrum For Diagnostics Industries - Free Zone
Ismailia Free Zone, Block 5 .
Cairo- Port said Avenue.
Ismailia,Egypt
Tel: +2 064 3488 013 - +2 064 3488 014 Fax: +2 064 3488 015
www.sdi-fz.com

EC REP **MDSS GmbH**
Schiffgraben 41
30175 Hannover, Germany