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# TRIGLYCERIDES GPO Method

Reagent for quantitative determination of triglycerides in human serum and plasma

| | | | | | | |
|---|---|---|---|---|---|---|
| REF 80019 | **R1** | 2 x 50 mL | **R2** | 2 x 50 mL | **R3** | 1 x 5 mL |
| REF 87319 | **R1** | 10 x 100 mL | **R2** | 10 x 100 mL | **R3** | 1 x 5 mL |

**TECHNICAL SUPPORT AND ORDERS**
**Tel : (33) 03 23 25 15 50**
**support@biolabo.fr**
**Latest revision : www.biolabo.fr**

CE

IVD

Made In France

I: corresponds to significant modifications

## INTENDED USE (1)

This reagent is designated for professional use in laboratory (manual or automated method).
It allows the quantification of triglycerides in human serum and plasma to complete lipids profile.

## I GENERALITIES

The increase of triglycerides in blood can be of genetic origin or secondary to other metabolic disorders such as: diabetes mellitus, hyper and hypothyroidisms, hepatic diseases, acute and chronic pancreatitis, nephrosis. A rise in triglycerides also represents an atherogenic risk factor. It is responsible for the opalescence, or even the cloudiness of the serum. Corticoids and oestrogen/progestin treatments can also aggravate hypertriglyceridemia.

## PRINCIPLE (4) (5)

Fossati and Prencipe method associated with Trinder reaction.
Reaction scheme is as follows:

Triglycerides $\xrightarrow{\text{Lipase}}$ Glycerol + free fatty acids

Glycerol + ATP $\xrightarrow{\text{GK}}$ Glycerol 3 Phosphate + ADP

Glycerol 3 Phosphate + $O_2$ $\xrightarrow{\text{GPO}}$ DihydroxyacetonePhosphate + $H_2O_2$

$H_2O_2$ + 4-Chlorophenol + PAP $\xrightarrow{\text{POD}}$ Quinoneimine (pink) + $H_2O$

The absorbance measured at 500 (480 – 520) nm is proportional to the amount of triglycerides in the specimen.

## REAGENTS

**R1 TRIGLYCERIDES GPO** Buffer

| | | |
|---|---|---|
| PIPES | 100 | mmol/L |
| Magnesium chloride | 9.8 | mmol/L |
| Chloro-4-phenol | 3.5 | mmol/L |
| Preservative | | |

According to 1272/2008 regulation, this reagent is not classified as dangerous

**R2 TRIGLYCERIDES GPO** Enzymes

| | | |
|---|---|---|
| Lipase | ≥ 1000 | IU/L |
| Peroxidase (POD) | ≥ 1700 | IU/L |
| Glycerol 3 phosphate oxidase (GPO) | ≥ 3000 | IU/L |
| Glycerol Kinase (GK) | ≥ 660 | IU/L |
| 4 - Amino – antipyrine (PAP) | 0.5 | mmol/L |
| Adenosine triphosphate Na (ATP) | 1.3 | mmol/L |

Before reconstitution:

ATTENTION: Acute tox.4: H302 - Harmful if swallowed

P264 : Wash hands thoroughly after handling.
P301+312 : IF SWALLOWED: Call a POISON CENTER or doctor/physician if you feel unwell. P330 : Rinse mouth, P501: Dispose of contents/container in accordance with dangerous goods regulations. Classification due to : 4-Amino-antipyrine 1 - <2.5%. For more details refer to current Material Safety Data Sheet (MSDS)

Once reconstituted: Working Reagent is not classified as dangerous

**R3 TRIGLYCERIDES GPO** Standard 200 mg/dL (2.28 mmol/L)

According to 1272/2008 regulation, this reagent is not classified as dangerous

## SAFETY CAUTIONS

BIOLABO reagents are designated for professional, in vitro diagnostic use (do not pipette with mouth).

- Refer to current Material Safety Data Sheet available on request or on www.biolabo.fr
- Verify the integrity of the contents before use.
- Waste disposal: Respect legislation in force in the country.

All specimens or reagents of biological origin should be handled as potentially infectious. Respect legislation in force in the country.

## REAGENTS PREPARATION

Use a non-sharp instrument to remove aluminium cap.
Working reagent: Add promptly the contents of vial R2 into vial R1.
Mix gently and wait for complete dissolution.
Vial R3: Ready for use.

## STABILITY AND STORAGE

**Stored away from light, well caped in the original vial at 2-8°C, when stored and used as described, reagents are stable:**

Unopened:

- Until expiry date stated on the label of the kit.

Once opened:

- Reconstitute immediately contents of vial R2.
- Standard (vial R3) :
  Transfer requested quantity and store the vial at 2-8°C.

Once reconstituted:

- Transfer requested quantity and store in the original vial at 2-8°C.
- Working reagent is stable fo1 year when free from contamination.
- Discard reagent if cloudy or if absorbance at 500 nm > 0.200.
- Don't use working reagent after expiry date.

## SPECIMEN COLLECTION AND HANDLING (2)

Serum or plasma (Heparin or EDTA) fasting ≥ 12 hours.

Separate from cells within 2 hours.

Do not use oxalate, fluoride or citrate.

Triglycerides are stable in specimen for:

- 5-7 days at 2-8°C.
- 3 months at –20°C.
- many years at –70°C.

Avoid repeated freezing and thawing.

## LIMITS (1) (2) (3)

For a more comprehensive review of factors affecting this assay refer to the publication of Young D.S.

## MATERIAL REQUIRED BUT NOT PROVIDED

1. Basic medical analysis laboratory equipment.
2. Spectrophotometer or Biochemistry Clinical Analyzer

## QUALITY CONTROL

- REF 95010 EXATROL-N Level I
- REF 95011 EXATROL-P Level II
- External quality control program.

It is recommended to control in the following cases:

- At least once a run.
- At least once within 24 hours.
- When changing vial of reagent.
- After maintenance operations on the instrument.

If control is out of range, apply following actions:

1. Prepare a fresh control and repeat the test.
2. If control is still out of range, use a new vial of calibrator or a fresh calibrator and repeat the test.
3. If control is still out of range, repeat the tests with a new vial of reagent.

If control is still out of range, please contact BIOLABO technical support or your local Agent.

## REFERENCE INTERVAL (6)

| Serum or Plasma | mg/dL | [ mmol/L ] |
|---|---|---|
| Reference range | 35-160 | [ 0.40-1.82 ] |

Each laboratory should establish its own normal ranges for the population that it serves.

## PERFORMANCES

On Kenza 240TX, 37°C, 505nm

Detection limit: approx. 1 mg/dL

Precision:

| *Within-run N = 20* | *Low level* | *Normal level* | *High level* |
|---|---|---|---|
| Mean mg/dL | 41 | 102 | 206 |
| S.D. mg/dL | 1.0 | 3.0 | 0.5 |
| C.V. % | 2.5 | 2.7 | 2.5 |

| *Between run N = 20* | *Low level* | *Normal level* | *High level* |
|---|---|---|---|
| Mean mg/dL | 59 | 123 | 223 |
| S.D. mg/dL | 0.31 | 5.2 | 8.4 |
| C.V. % | 5.4 | 4.3 | 3.8 |

Analytical Sensitivity: approx. 0.125 abs at 500 nm for 100 mg/dL (manual procedure, 500 m, 1cm pathlength)

On Cobas Mira, 505 nm, 37°C:

Measuring Range: between 10 and 700 mg/dL

Comparison studies with commercially available reagent:
Realised with human specimens (n=75) between 25 and 350 mg/dL

$y = 1.0182 x - 0.0302$ $r = 0.9958$

Interferences:

| | |
|---|---|
| Ascorbic acid | Negative interference from 1 mg/dL |
| Total bilirubin | Positive interference from 100 µmol/L |
| Haemoglobin | No interference up to 300 µmol/L |
| Glucose | No interference up to 1260 mg/dL |

Other substances may interfere (see § Limits)

## CALIBRATION (7)

- REF 95015 Multicalibrator traceable *to SRM 909b2*
- Standard (vial R3): With manual procedure.

The calibration frequency depends on proper instrument functions and on the preservation of reagent.

Make a new calibration when changing reagent batch, if quality control results are found out of the range and after maintenance operations

## PROCEDURE

Manual method

Let stand reagent and specimens at room temperature.

| Reagent | 1000 µL |
|---|---|
| Blank, Calibrator, control or specimen | 10 µL |

Mix well. Let stand for 10 minutes at room temperature or 5 minutes at 37°C.
Record absorbance at 500 nm (480-520) against reagent blank.
Color is stable for 1 h.

1- Performances with manual procedure should be validated by user.
2- Kenza applications and other applications proposal are available on request.

## CALCULATION

Serum or plasma:

$$\text{Result} = \frac{\text{Abs (Assay)}}{\text{Abs (Calibrator)}} \times \text{Calibrator concentration}$$

## REFERENCES


(1) *TIETZ N.W. Text book of clinical chemistry, 3rd Ed. C.A. Burtis, E.R. Ashwood, W.B. Saunders (1999) p. 809-857.*
(2) *Clinical Guide to Laboratory Test, 4thEd., N.W. TIETZ (2006) p. 1074-1077.*
(3) *YOUNG D.S., Effect of Drugs on Clinical laboratory Tests, 4th Ed. (1995) p.3-573 to 3-589*
(4) *Fossati P., Prencipe L., Clin. Chem. (1982), 28, p.2077-2080.*
(5) *Trinder P. Ann. Clin. Biochem. (1969), 6, p.27-29.*
(6) *TIETZ N.W. Text book of clinical chemistry, 2nd Ed. C.A. Burtis, E.R. Ashwood, W.B. Saunders (1994)p. 1030-1058 et p. 1073-1080.*
(7) *SRM: Standard Reference Material ®*


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|---|---|---|---|---|---|
| Manufacturer | Expiry date | In vitro diagnostic | Storage temperature | Dematerialized water | Biological risk |
| Product Reference | See Insert | Batch number | Store away from light | Sufficient for | Dilute with |