

## Triglycerides

## Order information

REF	CONTENT	System-ID	Analyzers on which <b>cobas c</b> pack can be used
20767107 322	Triglycerides (250 tests)	System-ID 07 6710 7	COBAS INTEGRA 400 plus COBAS INTEGRA 800
Materials required (but not provided):			
10759350 190	Calibrator f.a.s. (12 x 3 mL)	System-ID 07 3718 6	
10759350 360	Calibrator f.a.s. (12 x 3 mL, for USA)	System-ID 07 3718 6	
12149435 122	Precinorm U plus (10 x 3 mL)	System-ID 07 7999 7	
12149435 160	Precinorm U plus (10 x 3 mL, for USA)	System-ID 07 7999 7	
12149443 122	Precipath U plus (10 x 3 mL)	System-ID 07 8000 6	
12149443 160	Precipath U plus (10 x 3 mL, for USA)	System-ID 07 8000 6	
10171743 122	Precinorm U (20 x 5 mL)	System-ID 07 7997 0	
10171735 122	Precinorm U (4 x 5 mL)	System-ID 07 7997 0	
10171778 122	Precipath U (20 x 5 mL)	System-ID 07 7998 9	
10171760 122	Precipath U (4 x 5 mL)	System-ID 07 7998 9	
10781827 122	Precinorm L (4 x 3 mL)	System-ID 07 9026 5	
11285874 122	Precipath L (4 x 3 mL)	System-ID 07 9500 3	
05117003 190	PreciControl ClinChem Multi 1 (20 x 5 mL)	System-ID 07 7469 3	
05947626 190	PreciControl ClinChem Multi 1 (4 x 5 mL)	System-ID 07 7469 3	
05947626 160	PreciControl ClinChem Multi 1 (4 x 5 mL, for USA)	System-ID 07 7469 3	
05117216 190	PreciControl ClinChem Multi 2 (20 x 5 mL)	System-ID 07 7470 7	
05947774 190	PreciControl ClinChem Multi 2 (4 x 5 mL)	System-ID 07 7470 7	
05947774 160	PreciControl ClinChem Multi 2 (4 x 5 mL, for USA)	System-ID 07 7470 7	

## English

## System information

Test TRIGL, test ID 0-010

## Intended use

In vitro test for the quantitative determination of the triglycerides concentration in human serum and plasma on COBAS INTEGRA systems.

Summary<sup>1,2,3,4,5,6</sup>

Triglycerides are esters of the trihydric alcohol glycerol with 3 long-chain fatty acids. They are partly synthesized in the liver and partly ingested in food.

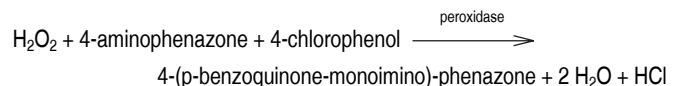
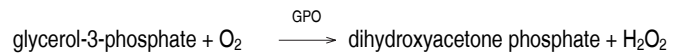
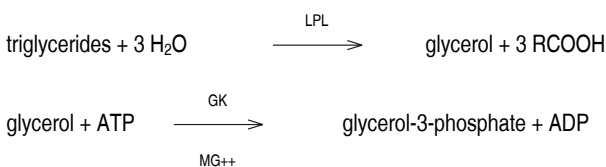
The determination of triglycerides is utilized in the diagnosis and treatment of patients having diabetes mellitus, nephrosis, liver obstruction, lipid metabolism disorders and numerous other endocrine diseases.

The enzymatic triglycerides assay as described by Eggstein and Kreutz still required saponification with potassium hydroxide. Numerous attempts were subsequently made to replace alkaline saponification by enzymatic hydrolysis with lipase. Bucolo and David tested a lipase/protease mixture; Wahlefeld used an esterase from the liver in combination with a particularly effective lipase from *Rhizopus arrhizus* for hydrolysis.

This method is based on the work by Wahlefeld using a lipoprotein lipase from microorganisms for the rapid and complete hydrolysis of triglycerides to glycerol followed by oxidation to dihydroxyacetone phosphate and hydrogen peroxide. The hydrogen peroxide produced then reacts with 4-aminophenazone and 4-chlorophenol under the catalytic action of peroxidase to form a red dyestuff (Trinder endpoint reaction). The color intensity of the red dyestuff formed is directly proportional to the triglyceride concentration and can be measured photometrically.

Test principle<sup>6</sup>

Enzymatic colorimetric test



## Reagents - working solutions

**R** PIPES buffer: 50 mmol/L, pH 6.8; Mg<sup>2+</sup>: 40 mmol/L; sodium cholate: 0.20 mmol/L; ATP: ≥ 1.4 mmol/L; 4-aminophenazone: ≥ 0.13 mmol/L; 4-chlorophenol: 4.7 mmol/L; LPL (microbial): ≥ 83 μkat/L; GK (microbial): ≥ 3 μkat/L; GPO (microbial): ≥ 41 μkat/L; POD (horseradish): ≥ 1.6 μkat/L; preservative; stabilizers

R is in position B.

## Precautions and warnings

For in vitro diagnostic use for health care professionals. Exercise the normal precautions required for handling all laboratory reagents.

Infectious or microbial waste:

Warning: handle waste as potentially biohazardous material. Dispose of waste according to accepted laboratory instructions and procedures.

Environmental hazards:

Apply all relevant local disposal regulations to determine the safe disposal.

Safety data sheet available for professional user on request.

For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

This kit contains components classified as follows in accordance with the Regulation (EC) No. 1272/2008:

H412 Harmful to aquatic life with long lasting effects.

## Prevention:

P273 Avoid release to the environment.

## Disposal:

**Triglycerides**

P501 Dispose of contents/container to an approved waste disposal plant.

Product safety labeling follows EU GHS guidance.

Contact phone: all countries: +49-621-7590, USA: 1-800-428-2336

**Reagent handling**

Ready for use

**Storage and stability**

Shelf life at 2-8 °C See expiration date on **cobas c** pack label

**COBAS INTEGRA 400 plus system**

On-board in use at 10-15 °C 8 weeks

**COBAS INTEGRA 800 system**

On-board in use at 8 °C 8 weeks

**Specimen collection and preparation**

For specimen collection and preparation only use suitable tubes or collection containers.

Only the specimens listed below were tested and found acceptable.

Serum

Plasma: Li-heparin and EDTA plasma

EDTA tubes that are less than 1/2 full may cause a negative bias for triglycerides results.

The sample types listed were tested with a selection of sample collection tubes that were commercially available at the time of testing, i.e. not all available tubes of all manufacturers were tested. Sample collection systems from various manufacturers may contain differing materials which could affect the test results in some cases. When processing samples in primary tubes (sample collection systems), follow the instructions of the tube manufacturer.

Centrifuge samples containing precipitates before performing the assay.

Stability in serum: 2 days at 20-25 °C<sup>7</sup>  
10 days at 4 °C<sup>8</sup>  
3 months at -20 °C<sup>9</sup>  
several years at -70 °C<sup>9</sup>

Stability in plasma: 2 days at 20-25 °C<sup>7</sup>  
15 days at 4 °C<sup>10</sup>  
3 months at -20 °C<sup>9</sup>  
several years at -70 °C<sup>9</sup>

**Materials provided**

See "Reagents – working solutions" section for reagents.

**Assay**

For optimum performance of the assay follow the directions given in this document for the analyzer concerned. Refer to the appropriate operator's manual for analyzer-specific assay instructions.

**Application for serum and plasma****COBAS INTEGRA 400 plus test definition**

Measuring mode	Absorbance
Abs. calculation mode	Endpoint
Reaction mode	R-S
Reaction direction	Increase
Wavelength A/B	512/659 nm
Calc. first/last	17/42
Unit	mmol/L

**Pipetting parameters**

Diluent (H<sub>2</sub>O)

R	120 µL	
Sample	2 µL	28 µL
Total volume	150 µL	

**COBAS INTEGRA 800 test definition**

Measuring mode	Absorbance
Abs. calculation mode	Endpoint
Reaction mode	R-S
Reaction direction	Increase
Wavelength A/B	512/659 nm
Calc. first/last	17/60
Unit	mmol/L

**Pipetting parameters**

		Diluent (H <sub>2</sub> O)
R	120 µL	
Sample	2 µL	28 µL
Total volume	150 µL	

**Calibration**

Calibrator	Calibrator f.a.s. Use deionized water as zero calibrator.
Calibration mode	Linear regression
Calibration replicate	Duplicate recommended
Calibration interval	Each lot and as required following quality control procedures

Calibration interval may be extended based on acceptable verification of calibration by the laboratory.

Traceability: This method has been standardized against the ID-MS<sup>a)</sup> method.

a) Isotope Dilution Mass Spectrometry

**Quality control**

Reference range	Precinorm U, Precinorm U plus, Precinorm L or PreciControl ClinChem Multi 1
Pathological range	Precipath U, Precipath U plus, Precipath L or PreciControl ClinChem Multi 2
Control interval	24 hours recommended
Control sequence	User defined
Control after calibration	Recommended

For quality control, use control materials as listed in the "Order information" section. In addition, other suitable control material can be used.

The control intervals and limits should be adapted to each laboratory's individual requirements. Values obtained should fall within the defined limits. Each laboratory should establish corrective measures to be taken if values fall outside the defined limits.

Follow the applicable government regulations and local guidelines for quality control.

**Calculation**

COBAS INTEGRA analyzers automatically calculate the analyte concentration of each sample. For more details, please refer to Data Analysis in the Online Help (COBAS INTEGRA 400 plus/800 analyzers).

Conversion factor: mmol/L × 88.5 = mg/dL

**Note**

If the free glycerol is to be taken into account, then 0.11 mmol/L (10 mg/dL) must be subtracted from the patient's triglycerides value obtained.<sup>9</sup>

**Limitations - interference**

Endogenous unesterified glycerol in the sample will falsely elevate serum triglycerides.

Criterion: Recovery within  $\pm 10\%$  of initial value.

Icterus:<sup>11</sup> No significant interference up to an I index of 5 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 86  $\mu\text{mol/L}$  or 5 mg/dL).

Hemolysis:<sup>11</sup> No significant interference up to an H index of 600 (approximate hemoglobin concentration: 373  $\mu\text{mol/L}$  or 600 mg/dL).

Drugs: No interference was found at therapeutic concentrations using common drug panels.<sup>12,13</sup> Exceptions: Ca-Dobesilate, L- $\alpha$ -Methyldopa, Levodopa, and Phenylbutazone cause artificially low triglycerides values at the tested drug level. Dicyclic (Etamsylate) at therapeutic concentrations may lead to false-low results.<sup>14</sup>

No significant interference by physiological ascorbic acid concentrations. Ascorbic acid levels higher than 114  $\mu\text{mol/L}$  (2 mg/dL) decrease the apparent triglycerides concentration significantly.

Acetaminophen intoxications are frequently treated with N-Acetylcysteine. N-Acetylcysteine at a plasma concentration above 333 mg/L and the Acetaminophen metabolite N-acetyl-p-benzoquinone imine (NAPQI) independently may cause falsely low results.

Venipuncture should be performed prior to the administration of Metamizole. Venipuncture immediately after or during the administration of Metamizole may lead to falsely low results. A significant interference may occur at plasma Metamizole concentrations above 0.05 mg/mL.

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.<sup>15</sup>

For diagnostic purposes, the results should always be assessed in conjunction with the patient's medical history, clinical examination and other findings.

**ACTION REQUIRED**

**Special Wash Programming:** The use of special wash steps is mandatory when certain test combinations are run together on COBAS INTEGRA analyzers. Refer to the CLEAN Method Sheet for further instructions and for the latest version of the Extra wash cycle list.

**Where required, special wash/carry-over evasion programming must be implemented prior to reporting results with this test.**

**Limits and ranges****Measuring range**

0.1-10 mmol/L (8.85-885 mg/dL)

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:10 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 10.

**Lower limits of measurement**

Lower detection limit of the test:

0.1 mmol/L (8.85 mg/dL)

The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying 3 standard deviations above that of a zero sample (zero sample + 3 SD, repeatability, n = 30).

**Expected values**

According to NCEP<sup>16</sup>

Normal range: < 1.70 mmol/L (< 150 mg/dL)

**Clinical interpretation** according to the recommendations of the European Atherosclerosis Society:<sup>17</sup>

	mmol/L	mg/dL	Lipid metabolism disorder
Cholesterol	< 5.18	< 200	No
Triglycerides	< 2.26	< 200	
Cholesterol	5.22-7.77	200-300	Yes if HDL-cholesterol < 0.9 mmol/L (< 35 mg/dL)

	mmol/L	mg/dL	Lipid metabolism disorder
Cholesterol	> 7.77	> 300	Yes
Triglycerides	> 2.26	> 200	

Each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference ranges.

**Specific performance data**

Representative performance data on the analyzers are given below. Results obtained in individual laboratories may differ.

**Precision**

Precision was determined using human samples and controls in an internal protocol with repeatability and intermediate precision (2 aliquots per run, 2 runs per day, 20 days). The following results were obtained:

	Level 1	Level 2
Mean	0.97 mmol/L (85.9 mg/dL)	1.63 mmol/L (144 mg/dL)
CV repeatability	1.6 %	1.6 %
CV intermediate precision	1.9 %	1.9 %

**Method comparison**

Triglycerides values for human serum and plasma samples obtained on a COBAS INTEGRA 700 analyzer using the COBAS INTEGRA Triglycerides (TRIGL) reagent (y) were compared with those determined using commercially available reagents for triglycerides on a COBAS INTEGRA 700 analyzer (COBAS INTEGRA TRIG reagent) (x) and an alternative manufacturer's clinical chemistry system (x). Samples were measured in duplicate. Sample size (n) represents all replicates.

The sample concentrations were between 0.53 and 7.0 mmol/L (46.9 and 620 mg/dL).

**COBAS INTEGRA 700 analyzer**

Sample size	(n)	222
Correlation coefficient	(r)	0.998
	(r <sub>s</sub> )	0.994
Linear regression		y = 1.038x - 0.065 mmol/L
Passing/Bablok <sup>18</sup>		y = 1.013x - 0.030 mmol/L

**Alternative system**

Sample size	(n)	200
Correlation coefficient	(r)	0.998
	(r <sub>s</sub> )	0.996
Linear regression		y = 1.002x + 0.039 mmol/L
Passing/Bablok <sup>18</sup>		y = 1.012x + 0.007 mmol/L

**References**

- Greiling H, Gressner AM, eds. Lehrbuch der Klinischen Chemie und Pathobiochemie, 3rd ed. Stuttgart/New York: Schattauer Verlag 1995.
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**Triglycerides**




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A point (period/stop) is always used in this Method Sheet as the decimal separator to mark the border between the integral and the fractional parts of a decimal numeral. Separators for thousands are not used.

Any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the Member State in which the user and/or the patient is established.

**Symbols**

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard (for USA: see [dialog.roche.com](http://dialog.roche.com) for definition of symbols used):

	Contents of kit
	Volume after reconstitution or mixing
	Global Trade Item Number

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