GGT-2

cobas®

γ-Glutamyltransferase ver.2 - Standardized against IFCC

Enzymes

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
03002721 122	γ-Glutamyltransferase ver.2 (400 tests)	System-ID 07 6598 8	COBAS INTEGRA 400 plus COBAS INTEGRA 800
10759350 190	Calibrator f.a.s. (12 × 3 mL)	System-ID 07 3718 6	
12149435 122	Precinorm U plus (10 × 3 mL)	System-ID 07 7999 7	
12149443 122	Precipath U plus (10 × 3 mL)	System-ID 07 8000 6	
10171743 122	Precinorm U (20 × 5 mL)	System-ID 07 7997 0	
10171735 122	Precinorm U (4 × 5 mL)	System-ID 07 7997 0	
10171778 122	Precipath U (20 × 5 mL)	System-ID 07 7998 9	
10171760 122	Precipath U (4 × 5 mL)	System-ID 07 7998 9	
05117003 190	PreciControl ClinChem Multi 1 (20 × 5 mL)	System-ID 07 7469 3	
05947626 190	PreciControl ClinChem Multi 1 (4 × 5 mL)	System-ID 07 7469 3	
05117216 190	PreciControl ClinChem Multi 2 (20 × 5 mL)	System-ID 07 7470 7	
05947774 190	PreciControl ClinChem Multi 2 (4 × 5 mL)	System-ID 07 7470 7	

English

System information

Test GGTI2, test ID 0-498, standardized against IFCC

Intended use

In vitro test for the quantitative determination of the catalytic activity of GGT (EC 2.3.2.2; γ-glutamyl peptide: amino acid γ-glutamyltransferase) in human serum and plasma on COBAS INTEGRA systems.

Summary[1,2,3,4,5]

Gamma-glutamyltransferase is used in the diagnosis and monitoring of hepatobiliary diseases. Enzymatic activity of GGT is often the only parameter with increased values when testing for such diseases, and is one of the most sensitive indicators known. Gamma-glutamyltransferase is also a sensitive screening test for occult alcoholism. Elevated GGT activities are found in the serum of patients requiring long-term medication with phenobarbital and phenytoin.

In 1969, Szasz published the first kinetic procedure for GGT in serum. In 1983, the International Federation of Clinical Chemistry (IFCC) recommended the standardized method for determining GGT including optimization of substrate concentrations, employment of NaOH, glycylglycine buffer and sample start.The IFCC confirmed the recommendation and extended it for 37 °C in 2002.[6]

Test principle[7]

Enzymatic colorimetric assay

Gamma-glutamyltransferase transfers the γ-glutamyl group of L-γ-glutamyl-3-carboxy-4-nitroanilide to glycylglycine.

L-γ-glutamyl-3-carboxy-4-nitroanilide + glycylglycine → L-γ-glutamyl-glycylglycine + 5-amino-2-nitrobenzoate

The amount of 5-amino-2-nitrobenzoate liberated is proportional to the GGT activity in the sample. It is determined by measuring the increase in absorbance at 409 nm.

Reagents - working solutions

R1 TRIS: 492 mmol/L, pH 8.25; glycylglycine: 492 mmol/L; preservative; additive

SR L-γ-glutamyl-3-carboxy-4-nitroanilide: 22.5 mmol/L; acetate: 10 mmol/L, pH 4.5; stabilizer; preservative

R1 is in position B and SR is in position C.

Precautions and warnings

Pay attention to all precautions and warnings listed in Section 1 / Introduction of this Method Manual.

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	12 weeks
COBAS INTEGRA 800 system	
On-board in use at 8 °C	12 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: Collect serum using standard sampling tubes.
Plasma: Heparin (Li-, Na-, NH_4^+-) or EDTA (K_2-, K_3-) plasma. K_3-EDTA plasma values are approximately 6 % lower than serum values.

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:	7 days at 15-25 °C[8]
	7 days at 2-8 °C[8]
	1 year at (-15)-(-25) °C[9]

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	Kinetic
Reaction mode	R1-S-SR
Reaction direction	Increase
Wavelength A/B	409/659 nm
Calc. first/last	50/69

GGT-2

γ-Glutamyltransferase ver.2 - Standardized against IFCC

Unit	U/L

Pipetting parameters

		Diluent (H_2O)
R1	25 µL	35 µL
Sample	3 µL	20 µL
SR	20 µL	20 µL
Total volume	123 µL	

COBAS INTEGRA 800 test definition

Measuring mode	Absorbance
Abs. calculation mode	Kinetic
Reaction mode	R1-S-SR
Reaction direction	Increase
Wavelength A/B	409/659 nm
Calc. first/last	73/98
Unit	U/L

Pipetting parameters

		Diluent (H_2O)
R1	25 µL	35 µL
Sample	3 µL	20 µL
SR	20 µL	20 µL
Total volume	123 µL	

Calibration

Calibrator	Calibrator f.a.s. COBAS INTEGRA 400 plus system: STD-2 is defined by a fixed values. COBAS INTEGRA 800 system: STD-2 is defined by a 1:100 dilution of STD-1, performed automatically by the instrument.
Calibration mode	Linear regression
Calibration replicate	Duplicate recommended
Calibration interval	Each lot

Traceability: This method has been standardized against the original formulation and procedures recommended by the IFCC.[6] Use the set point value assigned as GGT liquid standardized against IFCC.

Quality control

Reference range	Precinorm U, Precinorm U plus or PreciControl ClinChem Multi 1
Pathological range	Precipath U, Precipath U plus 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 activity of each sample. For more details, please refer to Data Analysis in the Online Help (COBAS INTEGRA 400 plus/800 analyzers).

Conversion factor: U/L × 0.0167 = µkat/L

Limitations - interference

Criterion: Recovery within ± 10 % of initial value.

Icterus:[10] No significant interference up to an I index of 48 for unconjugated bilirubin (approximate unconjugated bilirubin concentration: 821 µmol/L or 48 mg/dL). No significant interference with conjugated bilirubin.

Hemolysis:[10] No significant interference up to an H index of 550 (approximate hemoglobin concentration: 0.34 mmol/L or 550 mg/dL).

Lipemia (Intralipid):[10] No significant interference.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[11,12]

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[13]

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

3-1200 U/L (0.05-20 µkat/L)

Determine samples having higher activities 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:
3 U/L (0.05 µkat/L)

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 = 21).

Expected values

Reference Interval Study at 37 °C (corrected in 2005)[14,15]

Men (n = 216)	10-71 U/L	(0.17-1.19 µkat/L)
Women (n = 228)	6-42 U/L	(0.10-0.70 µkat/L)

Consensus values[16]

Men	< 60 U/L	(< 1.00 µkat/L)
Women	< 40 U/L	(< 0.67 µkat/L)

Conversion factors to other temperatures have been published[17] but these have not been checked by Roche for the present reagent.

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 COBAS INTEGRA 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 (n = 21) and intermediate precision (1 aliquot per run, 1 run per day, 21 days). The following results were obtained:

GGT-2

γ-Glutamyltransferase ver.2 - Standardized against IFCC

cobas®

Enzymes

	Level 1	Level 2
Mean	42.0 U/L (0.701 µkat/L)	230 U/L (3.84 µkat/L)
CV repeatability	1.8 %	1.0 %

	Level 1	Level 2
Mean	42.6 U/L (0.711 µkat/L)	221 U/L (3.69 µkat/L)
CV intermediate precision	1.8 %	1.3 %

Method comparison

GGT values for human serum and plasma samples obtained on a COBAS INTEGRA 800 analyzer with the COBAS INTEGRA γ-Glutamyltransferase ver.2 (GGT-2) reagent and the application GGTI2 (y) were compared with those determined using the corresponding reagent on a Roche/Hitachi 917 analyzer (x).

Sample size (n) = 65

Roche/Hitachi 917 analyzer

Passing/Bablok[18]	Linear regression
y = 1.008x - 1.26 U/L	y = 1.007x - 0.939 U/L
τ = 0.992	r = 1.00
SD (md 95) = 6.33	Sy.x = 2.83

The sample activities were between 40.7 and 919 U/L (0.680 and 15.4 µkat/L).

References

1 Thomas L, ed. Labor und Diagnose, 4th ed. Marburg: Die Medizinische Verlagsgesellschaft 1992.

2 Shaw LM. Keeping pace with a popular enzyme GGT. Diagnostic Medicine May/June 1982;1–8.

3 Szasz G. A kinetic photometric method for serum γ-glutamyl-transferase. J Clin Chem 1969;15:124-136.

4 Shaw LM, Stromme JH, London JL, et al. Approved recommendation (1983) on IFCC methods for the measurement of catalytic concentrations of enzymes. Part 4. IFCC method for gamma-glutamyl transferase. J Clin Chem Clin Biochem 1983;21:633-646.

5 Klauke R, Schmidt E, Lorentz K. Recommendations for carrying out standard ECCLS procedures (1988) for the catalytic concentrations of creatine kinase, aspartate aminotransferase, alanine aminotransferase and γ-glutamyltransferase at 37 °C. Eur J Clin Chem Clin Biochem 1993;31:907-909.

6 Schumann G, Bonora R, Ceriottiet F et al. IFCC Primary Reference Procedures for the Measurement of Catalytic Activity Concentrations of Enzymes at 37 °C – Part 6. Reference Procedure for the Measurement of Catalytic Activity Concentrations of gamma-glutamyltransferase. Clin Chem Lab Med 2002;40(7):734-738.

7 Szasz G, Weimann G, Stähler F, et al. New Substrates for measuring gamma-glutamyl-transpeptidase activity. Z Klin Chem Klin Biochem 1974;12:228-233.

8 Szasz G. Methods of Enzymatic Analysis. 2nd English ed. New York. Academic Press, Inc 1974;717.

9 Tietz NW, ed. Clinical Guide to Laboratory Tests, 3rd ed. Philadelphia PA: WB Saunders Company 1995;286.

10 Glick MR, Ryder KW, Jackson SA. Graphical Comparisons of Interferences in Clinical Chemistry Instrumentation. Clin Chem 1986;32:470-475.

11 Breuer J. Report on the Symposium “Drug effects in Clinical Chemistry Methods”. Eur J Clin Chem Clin Biochem 1996;34:385-386.

12 Sonntag O, Scholer A. Drug interference in clinical chemistry: recommendation of drugs and their concentrations to be used in drug interference studies. Ann Clin Biochem 2001;38:376-385.

13 Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. Clin Chem Lab Med 2007;45(9):1240-1243.

14 Abicht K, El-Samalouti V, Junge W, et al. Multicenter evaluation of new GGT and ALP reagents with new reference standardization and determination of 37 °C reference intervals. Clin Chem Lab Med 2001;39:Special Supplement pp S 346.

15 Kytzia H-J. Reference intervals for GGT according to the new IFCC 37°C reference procedure. Clin Chem Lab Med 2005;43:A69 [abstract].

16 Thomas L, Müller M, Schumann G, et al Consensus of DGKL and VDGH for interim reference intervals on enzymes in serum. J Lab Med 2005; 29(5):301-308.

17 Zawta B, Klein G, Bablok W. Temperature Conversion in Clinical Enzymology? Klin Lab 1994;40:33-42.

18 Bablok W, Passing H, Bender R, et al. A general regression procedure for method transformation. Application of linear regression procedures for method comparison studies in clinical chemistry, Part III. J Clin Chem Clin Biochem 1988 Nov;26(11):783-790.

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.

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard.

CONTENT	Contents of kit
→	Volume after reconstitution or mixing

CE

Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com

Roche