



γ -GT

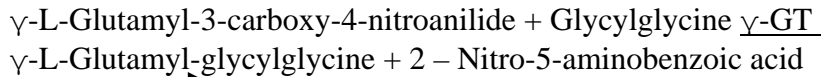
Carboxy substrate kinetic.

Quantitative determination of gamma-glutamyl transferase (γ -GT)

Store at 2-8 °C

PRINCIPLE OF THE METHOD

Gamma-glutamyl transferase (γ -GT) catalyses the transfer of γ -glutamyl group from γ -glutamyl-p-nitroanilide to acceptor glycylglycine, according to the following reaction:



The rate of 2-nitro-5-aminobenzoic acid formation, measured photometrically, is proportional to the catalytic concentration of γ -GT present in the sample ^{1,2}.

CLINICAL SIGNIFICANCE

Gamma-glutamyl transferase (γ -GT) is a cellular enzyme with wide tissue distribution in the body, primarily in the kidney, pancreas, liver and prostate.

Measurements of gamma-glutamyl transferase (γ -GT) activity are used in the diagnosis and treatment of hepatobiliary diseases such biliary obstruction, cirrhosis or liver tumours ^{1,2,5,6}.

Clinical diagnosis should not be made on a single test result; it should integrate clinical and other laboratory data.

REAGENTS

R1 Buffer	TRIS pH 8.6 Glycylglycine	100 mmol/L 100 mmol/L
R2 Substrate	L- γ -glutamyl-3-carboxy-4-nitroanilide	

PREPARATION

Working reagent (WR)

Dissolve one tablet of R2 Substrate in 2ml of R1 buffer

Cap and mix gently to dissolve contents.

Stability: 21 days at 2-8 °C

or 5 days at room temperature 15-25 °C

STORAGE AND STABILITY

All the components of the kit are stable until the expiration date on the label when stored tightly closed at 2-8 °C, protected from light and contaminations prevented during their use.

Do not use reagents over the expiration date

Signs of reagent deterioration:

- Presence of particles and turbidity.
- Blank absorbance (A) at 405 nm \geq 1.20.

ADDITIONA EQUIPMENT

- Spectrophotometer or colorimeter measuring at 405 nm.
- Thermostatic bath at 25°C, 30°C o 37°C (\pm 0.1°C)
- Matched cuvettes 1.0 cm light path.
- General laboratory equipment.

SAMPLES

Serum ¹. γ -GT is stable for at least 3 days at 2-8 °C, 8 hours at 15-25 °C and 1 month at – 20°C.

PROCEDURE

1. Assay conditions:
Wavelength405nm
Cuvette.....1 cm light path
Constant temperature25°C / 30°C/ 37°C.
2. Adjust the instrument to zero with distilled water or air.
3. Pipette into a cuvette

WR (ml)	1.0
Sample (μ L)	100

4. Mix, wait for 1 minute.
5. Read initial absorbance (A) of the sample, start the stopwatch and read absorbances at 1 minute intervals thereafter for 3 minutes.
6. Calculate the difference between absorbances and the average absorbance differences per minute (Δ A/min).

CALCULATIONS

$(\Delta A/Min) \times 1190 = U/L$ of γ -GT

Units: One international unit (IU) is the amount of enzyme that transforms 1 μ mol of substrate per minute. In standsard conditions. The concentration is expressed in units per liter of sample (U/L).

TEMPERATURE CONVERSION FACTORS

To correct results to other temperatures multiply by:

Assay temperature	Conversion factor to		
	25°C	30°C	37°C
25°C	1.00	1.37	1.79
30°C	0.73	1.00	1.30
37°C	0.56	0.77	1.00

QUALITY CONTROL

If control values are found outside the defined range, check the instrument, reagents and technique for problems.

Each laboratory should establish its own Quality Control scheme and corrective actions if controls do not meet the acceptable tolerances.

REFERENCE VALUES ¹

	25°C	30°C	37°C
Men	4-18 U/L	5-25 U/L	7-32 U/L
Women	6-28 U/L	8-38 U/L	11-50 U/L

These values are for orientation purpose; each laboratory should establish its own reference range.

PERFORMANCE CHARACTERISTICS

Measuring range: Form detection limit of 2U/L to linearity limit of 250 U/L.

If the results obtained were greater than linearity limit, dilute the sample 1/2 with NaCl 9 g/L and multiply the result by 2.

Precision:

	Intra-assay (n=20)		Inter-assay (n=20)	
Mean (U/L)	45.7	203	45.4	203
SD	0.66	1.72	1.04	3.43
CV (%)	1.44	0.85	2.28	1.69

Sensitivity: 1 U/L = 0.0008 ΔA/min.

Accuracy: Results obtained using Atlas reagents (y) did not show systematic differences when compared with other commercial reagents (x).

The results obtained using 50 samples were the following:

Correlation coefficient (r): 0.99

Regression equation: $y=0.9919x-0.1042$

The results of the performance characteristics depend on the analyzer used.

INTERFERENCES

Plasma should not be used ,anticoagulants inhibit the enzyme. Gross haemolysis interferes in the assay¹.

A list of drugs and other interfering substances with γ -GT determination has been reported by Young et. Al 3,4.

BIBLIOGRAPHY.

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ATLAS MEDICAL

William James House, Cowley Road, Cambridge

Tel: ++44 (0) 1223 858 910

Fax: ++44 (0) 1223 858 524

PPI493A01

Revision A (25.10.2007)