GGT

ENZOPAK

Last update 04-2014

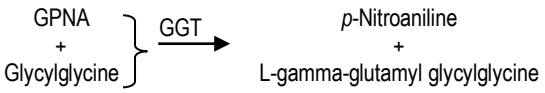
(SZASZ METHOD)
Reagent kit for quantitative estimation of Gamma Glutamyl Transferase (transpeptidase) activity in serum or plasma.

BACKGROUND & SYNOPSIS:
Gamma glutamyl transferase (GGT) was purified and characterized by Szewczuk and Baranowski. It catalyzes the transfer of the gamma glutamyl group from a gamma glutamyl peptide to an amino acid of another peptide.

In earlier methods, various substances such as glutathione, L-gamma-glutamyl-naphthylamide and L-gamma-amylanilide were used. Current methods use L-gamma-glutamyl-p-nitroanilide (GPNA) as a substrate since end product is coloured and it permits a direct reaction rate measurement without deproteinization.

ENZOPAK GGT is formulated on the Szasz method recommended by Scandinavian Society for Clinical Chemistry and Physiology. The method is kinetic, specific and sensitive.

PRINCIPLE :
Gamma - glutamyl - *p* - nitroanilide (GPNA) and glycylglycine are converted by the action of GGT to *p* - nitroaniline and L - gamma glutamyl glycylglycine. The rate of increase in absorbance at 405 nm due to the release of *p*-nitroaniline is directly proportional to the GGT activity.



DIAGNOSTIC SIGNIFICANCE :
GGT is one of the most sensitive enzyme of the hepatobiliary system. Elevated serum gamma-GT levels are indicative of the disease of the liver, biliary tract and pancreas. In cases of metastatic carcinoma, viral hepatitis, chronic hepatitis, cholelithiasis, cholangitis and cholecystitis, gamma-GT levels are found to be elevated. Since GGT activity is not elevated in any bone disorders, the assay is considered as a valuable diagnostic aid for differentiation between bone and liver disease in conjuction with alkaline phosphatase.

REAGENT COMPOSITION

Active Ingredients	Concentration
Reagent-1	
• Soluble GPNA	$\geq$ 3 mmol/L
Reagent-2	
• Buffer	100 mmol/L
• Glycyl glycine	30 mmol/L

pH 8.5$\pm$ 0.1 at 25^{0}C
Also contains non-reactive fillers and Stabilizers.

PRESENTATION :

	No. of Bottles
All reagents to be stored at 2-8^{0}C	20 x 1.1 ml
• 1 GGT (Substrate)	2 (10 Tablet)
• 2 GGT (Buffer)	2
• Reconstitution vial	1

PRECAUTION:
ENZOPAK GGT is for *IN-VITRO* diagnostic use only.
Reagent contains Sodium Azide. DO NOT INGEST.

WORKING REAGENT PREPARATION:
For 20 x 1.1 ml:
Reconstitute one tablet of 1 GGT with 1.1 ml of 2-GGT. Mix gently to disolve the contents. Use after 5 minutes.

REAGENT STORAGE AND STABILITY:
ENZOPAK GGT reagent is stable until the expiry date printed on the label at 2-8^0 C.

Working reagent is stable for 14 days at 2-8^0 C. Discard the working reagent if the absorbance reads 0.8 at 405 nm against distilled water.

SPECIMEN COLLECTION:
Fresh, clear serum with no hemolysis is the specimen of choice. Plasma, collected with the use of heparin as an anticoagulant may also be used.

REACTION PARAMETERS :

• Type of Reaction	:	Kinetic/Increasing O.D.
• Wavelength	:	405 nm
• Flowcell Temperature		37^{0}C / 30^{0}C
• Delay Time	:	60 Seconds
• Interval Time	:	30 Seconds
• No. of reading	:	4
• Sample Volume	:	100 µl (0.1 ml)
• Factor	:	1158
• Light Path	:	1.0 cm.
• Zero setting with	:	Distilled Water

PROCEDURE:
For laboratories using instruments with 1.0 ml. cuvette capacity.

PIPETTE IN TO TEST TUBES		TEST	REACTION TEMP.
• WORKING REAGENT	(ml)	1.0	37 ^{0} C
• SAMPLE	(ml)	0.1	

Mix immediately and read first absorbance of test exactly at 60 seconds and then, second, third and fourth at an interval of 30 seconds at 405 nm. Determine the mean change in absorbance per minute (ΔA/min) and use this for calculating test results.

NOTE :
For laboratories using instruments with cuvette capacity more or less than 1.0 ml. sample volume and working reagent volume may be proportionately increased or decreased.

TEST RESULTS :
Serum GGT Activity (IU/L) =ΔA/min X F
Where F = 1158 (Calculated on the basis of molar extinction coefficient for paranitroaniline and sample to total volume ratio.)

NORMAL VALUES :

	30^{0}C.	37^{0}C.
MEN (IU/L)	7-40	9-52
WOMEN (IU/L)	4-25	5-32

LINEARITY :
The method is linear upto 500 IU/L. For sample values higher than linear limit, dilute the sample suitably with 0.9 % saline and repeat the assay. Apply dilution factor to calculate test results.

REFERENCE :

- SZASZ G. Reaction rate method for gamma-glutamyl transferase activity in serum. Clin. Chem. 22.2051-2055 (1976).

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3/7,B.I.D.C., Gorwa, Vadodara 390 016 (INDIA)
Web: www.reckondiagnostics.com
Phone: +91·265·2281631
Email: mail@reckondiagnostics.com
RECKON DIAGNOSTICS P. LTD.