(DNPH METHOD)



Reagent Kit for quantitative estimation of glutamate pyruvate transaminase activity in serum or plasma.

BACKGROUND & SYNOPSIS:

Glutamate pyruvate transaminase (alanine aminotransferase, ALT) is one of the two transaminases used for diagnostic purpose. Transaminase or aminotransferrase carry out a reaction between an amino acid by transferring the amino group to a keto acid. The enzyme is found in most of the tissues, but liver is the richest in its content. It is also present in heart, kidneys, pancreas, muscles etc. Estimation of GPT is useful in the diagnosis of liver damage, obstructive jaundice etc.

The DNPH method was first used by Tonhazey and Cabaud et.al. it was modified by Reitman and Frankel.

SGPT is formulated on the methodology as modified by Reitman and Frankel.

PRINCIPLE:

For estimation of GPT sample is allowed to react with alphaketoglutarate and L-alanine. Pyruvate thus generated is coupled with 2,4-dinitrophenyl hydrazine to give corresponding hydrazone, a brown/red coloured complex, in an alkaline medium. This is measured photometrically at 505nm (500-550 nm or with Green filter).

Alpha-Ketoglutarate	L-Glutamate	
: + :	GPT	. +
L-Alanine	5W 8	Pyruvate

DIAGNOSTIC SIGNIFICANCE:

Even though glutamate pyruvate transaminase is widely distributed in various tissues of the body, it is a useful parameter in evaluating liver function. The serum levels are found elevated in case of hepatitis, obstructive jaundice, metastatic carcinoma, hepatic congestion and myocardial infarction or in kidney diseases.

SPECIMEN COLLECTION:

Fasting, fresh, unhemolysed, clear serum is the specimen of choice. However, plasma collected with heparin, oxalate or citrate as anticoagulants may be used.

PRECAUTIONS:

SGPT is for In Vitro diagnostic use only. Do not use hemolysed sample. Contact of reagent with skin, eyes, & clothes should be avoided.

PRESENTATION:

 1 SGPT (Substrate)
 2 SGPT (color reagent)
 3 SGPT (alkali Reagent)
 PYRUVATE STANDARD (STORE ALL AT 2-8° C)
 Ready for use Ready for use Dilute 1 + 9 ml
 170 U/L

NOTE:

It is suggested to plot a calibration graph to interpret test results.

Procedure with graph plotting available on request

WORKING REAGENT PREPARATION:

Dilute the 3 SGPT in the proportion 1:10 (1 ml of 3 SGPT + 9 ml of distilled water)

REAGENT STORAGE AND STABILITY:

SGPT reagents are stable at 2-8° C until expiry date stated on the label.

TEST PROCEDURE:

Pipette into Test Tubes	Reagent Blank	Calibrator	Test
1 SGPT	0.25 ml	ml 0.25 ml	
Sample	4	왕복의	0.05 ml
Calibrator / Std. 170U/L	i i	0.05 ml	9
Mix and incub	ate at 37°C	for 30 minu	tes.
2 SGPT	0.25 ml	0.25 ml	0.25 ml
Mix well and in	cubate for	20 minutes 3	7°C.
Sample	i s u		
3 SGPT (Diluted)	1.5 ml	1.5 ml	1.5 ml

Mix and read absorbance of all tubes against distilled water at 505 nm or with Green filter.

CALCULATION:

SGPT (ALT) Activity in U/L.

=(Abs. Test - Abs.Reagent Blank) x Conc. of standard (Abs. Calibrator - Abs.Reagent Blank) (170 U/L)

NORMAL VALUES: 5-50 UNITS/L.

PROCEDURE LIMIT:

The method is readable up to 300 U/liter for GPT activity higher than 300 U/liter dilute it suitably with 0.9% saline and repeat the assay. Apply proper dilution factor to obtain test results.

BIBLIOGRAPHY:

- REITMAN. S. AND FRANKEL, S., America J. of Clinical Pathology, 28; 56 (1957)
- TIETZ. N., Fundamentals of Clinical Chemistry. W.B. Saunders Co., U.S.A., (1970), P 447.

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ı.	$\overline{\mathbb{A}}$	Attention,see instructions for use	i	Consult Instructions For Use
а	IVD	For in vitro diagnostic use only	REF	Catalog #
n	2°C / 8°C	Store between 2-8°C	LOT	Lot Number
1	8	Do not use if package is damaged	M	Date of Manufacturing
	3	Manufacturer		Use by