ANA BIO ISP SGPT (ALT)

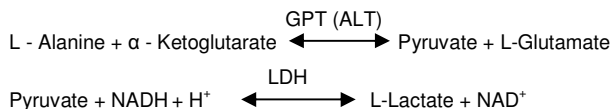## (IFCC without P5P activation Method)

For Miura Instruments

Intended Use
GPT (ALT) is a reagent kit used for the determination of GPT (ALT) activity in serum or plasma based on enzymatic UV-Kinetic method.

Principle
α - Ketoglutarate reacts with L-alanine in presence of GPT (ALT) to form pyruvate and L-glutamate. The increase in pyruvate is determined in an indicator reaction catalyzed by lactate dehydrogenase. The conversion of NADH to NAD$^+$ is proportional to the activity of GPT (ALT) in serum/plasma and is determined kinetically.

$$L - Alanine + \alpha - Ketoglutarate \xrightleftharpoons{GPT\ (ALT)} Pyruvate + L\text{-}Glutamate$$

$$Pyruvate + NADH + H^+ \xrightleftharpoons{LDH} L\text{-}Lactate + NAD^+$$

Components & Concentration of Reagents

Reagent	Component	Concentration
Reagent 1	TRIS buffer pH 7.8	100 mmol/L
	LDH	≥1000 U/L
	2-Oxoglutarate	15 mmol/L
	L-Alanine	≥500 mmol/L
	Stabilizers, excipients & surface active agents	
Reagent 2	TRIS buffer pH 9.8	20 mmol/L
	NADH	2.60 mmol/L
	Stabilizers, excipients & surface active agents	

Reagent storage and stability
The reagent kit should be stored at 2° - 8℃ and is stable till the expiry date indicated on the label.
A slight variation in the composition of the components may occur between batches, but this has no effect on the test results.
After opening, the vial R1 and R2 are stable 30 days if recapped immediately and protect from contamination, evaporation, direct light and stored at correct temperature.

Specimen collection and preservation
Blood should be collected in a clean dry container. Although serum is prefered, plasma with heparin or EDTA can also be used. Samples with any visible haemolysis are not acceptable. GPT (ALT) activity in serum/plasma is stable for 1 week at 2° - 8℃ and one month when stored at -20℃. The samples should be brought to room temperature prior to use.

Automation
This kit, though developed and manufactured to be used as manual assay and with I.S.E. Miura Analyzer, can be used also with other analyzers able to meet the specifications indicated in section "Reaction conditions – Test procedure" Application sheets are available for automatic instruments.
All applications not explicitly approved by KDPL. Cannot be guaranteed in terms of performance, and must there be established by the operator.

Calibration
For Calibration use the "Multicalibrator"

Calibration Stability
For the instrumentation series Miura, the calibration is recommended to be done every 10 days.

Materials required but not supplied in the kit
Calibrators and controls

Assay guidelines for Analyzer I.S.E. Miura

Analyte Name	ALT(GPT)
Method Code	GPT
Type	Kinetic
Unit	IU/L

Filter F1	340 nm
Blank in	Not Use

Step	Reaction Volume	U.M.
Volume reagent R1	200	µl
Volume reagent R2	50	µl
Sample Volume	25	µl
First Incubation	60	Sec
Final Incubation	192	Sec.

Normal Range
Guidance value : Up to 49 IU/L
Note: Expected range varies from population to population and each laboratory should establish its own normal range.

Limitations
This method is linear up to 600 IU / L. If the activity exceeds 600 IU/L, dilute the sample suitably with normal saline and repeat the assay. Apply proper dilution factor to calculate the final results.

Quality Control
To ensure adequate quality control measures, it is recommended that each batch should include a normal and an abnormal commercial reference control serum. It should be realized that the use of quality control material checks both instrument and reagent functions together. Factors which might affect the performance of this test include proper instrument function, temperature control, cleanliness of glassware, Wavelength setting, Expiration date of reagents and accuracy of prob aspiration.

Accuracy-Recovery
ALT/GPT added to a serum matrix containing known amounts of ALT gave an average recovery of 95%.

Interference
Triglycerides is below 2000 mg/dl does not interfere in the reaction. Bilirubin below 5.8 mg/dl does not interfere in the reaction. Haemoglobin interferes at concentrations above 10.0 g/L. Ascorbic Acid influences the reaction at concentrations over 30 mg/dl.

Precision of the Method

Within-run					
Range	U.M	Mean	S.D.	C.V. (%)	No. run
Low	IU/L	20.60	0.91	4.43	20
High	IU/L	96	1.46	1.52	20
Between run					
Range	U.M	Mean	S.D.	C.V. (%)	No. run
Low	IU/L	20.6	0.84	4.07	60
High	IU/L	96	1.73	1.81	60

Sensitivity
At 340 nm, the activity of ALT/GPT of 5 IU/L can estimate.

References
1. Tietz, N.W, ed. Clinical Guide to Laboratory tests, 3 ed. Philadelphia, pa : W.B. Saunders, 1995:20-21.
2. Bergmmeyer, HU,Horder M,Rej R. Approved recommandation(1985) on IFCC methods for the measurment of catalytic concentration of enzymes, Part 3. IFCC method for alanine aminotransferase. J.Clin. Chem. Clin. Biochem. 1986 ; 24 :481-489.
3. Fischbach F, Zawta B. Age - dependant reference limits of several enzymes in plasma at different measuring temperatures. Clin. Lab. 1992 ; 38 :555 - 561.
4. Hafkensheild. J.C.M., *et. al.*, *J. Clin. Chem. Clin. Biochem*. 17, 219 (1979).

Symbols

IVD	In Vitro Diagnostics	⚠	Caution
LOT	Batch No.	⧖	Product Expiry Date
CONT	Content	▲	Manufactured By
📖	Read Instructions	⋀	Date of Manufacture
🌡	Storage Temperature	☂	Keep Dry
REF	Catalogue No.	⚱	Fragile
		☀	Keep away from sun light

▲

KEE DIAGNOSTICS PVT LTD
(Formerly known as Kee GAD Biogen Pvt. Ltd.) .
CIN: U24231DL2004PTC128343 .
A subsidiary of KEE PHARMA LTD
A-8 . 3RD FLOOR. NARAINA INDUSTRIAL
AREA, PHASE 2 . NEW DELHI 110028.
W. www.keediagnostics.in.
E. info@keediagnostics.in, T. 011 43136000.