INTRODUCTION

1. ***Infinite*** Liquid GOT (AST) is a reagent set for determination of GOT (AST) activity in serum and plasma based on **UV - Kinetic method.**
2. ***Infinite*** Liquid GOT (AST) is a **ready-to-use,** two liquid reagent system.
3. ***Infinite*** Liquid GOT (AST) estimates GOT (AST) activity in just **2½ minutes.**
4. ***Infinite*** Liquid GOT (AST) is **linear** upto 800 IU/l.
5. ***Infinite*** Liquid GOT (AST) can be used on any **Spectrophotometer, Discrete semiautomated and Automated analyzers.** Programme can be designed for any specific analyzer upon request.
6. ***Infinite*** Liquid GOT (AST) is **stable till expiry** at 2 - 8°C.

PRINCIPLE

α-ketoglutarate reacts with L-aspartate in presence of GOT (AST) to form oxaloacetate and L-glutamate. The increase in oxaloacetate is determined in an indicator reaction catalyzed by malate dehydrogenase. The conversion of NADH to NAD^+ at 340 nm. is proportional to the activity of GOT (AST) in serum/plasma and is determined kinetically as rate of decrease in absorbance.

$$\text{L-aspartate} + \alpha\text{-ketoglutarate} \xrightleftharpoons{\text{GOT (AST)}^*} \text{Oxaloacetate} + \text{L-glutamate}$$

$$\text{Oxaloacetate} + \text{NADH} + H^+ \xrightleftharpoons{\text{malate dehydrogenase}} \text{L-malate} + NAD^+$$

*Abbreviations

AST = Aspartate transaminase
GOT = Glutamate oxaloacetate transaminase

PREPARATION OF WORKING SOLUTION

Prepare working solution by mixing **Reagent R_1** and **Reagent R_2** in the ratio **4:1** as per requirement.

REAGENT STORAGE STABILITY

The reagent kit should be stored at 2 - 8°C and is stable till the expiry date indicated on the label.

R_1 and R_2 reagents are stable till expiry at 2 - 8°C.

The working solution (4 R_1 + 1 R_2) is stable for 30 days at 2 - 8°C.

COMPONENTS & CONCENTRATION OF WORKING SOLUTION

Component	Concentration
• Tris buffer, pH 7.8	80 mmol/l
• L-aspartate	240 mmol/l
• Lactate dehydrogenase	≥ 3000 IU/l
• Malate dehydrogenase	≥ 400 IU/l
• NADH	0.23 mmol/l
• α-ketoglutarate	10 mmol/l

SPECIMEN COLLECTION & PRESERVATION

Blood should be collected in a clean dry container. Although serum is preferred, plasma with heparin or EDTA can be used. Samples with any visible haemolysis are not acceptable since erythrocytes contain approximately ten times the normal activity of GOT (AST) found in serum. GOT (AST) activity in serum/plasma is stable for 1 week at 2 - 8°C and 1 month at -20°C. The samples should be brought to room temperature prior to use.

PROCEDURE

☐ Reaction type	UV - Kinetic
☐ Reaction direction	Decreasing
☐ Wavelength	340 nm.
☐ Flowcell temperature	37°C
☐ Zero setting with	Distilled water
☐ Delay time	60 seconds
☐ No. of readings	4
☐ Interval	30 seconds
☐ Blank absorbance limit	≥ 0.900 Abs.
☐ Sample volume	0.05 ml (50 µl)
☐ Working solution volume (4 R_1 : 1 R_2)	1.0 ml (1000 µl)
☐ Factor	3339
☐ Linearity	800 IU/l

MANUAL ASSAY PROCEDURE

Prewarm at 37°C the required amount of working solution before use. Perform the assay as given below:

1 ml procedure

Serum/plasma	**0.05 ml**
Working solution	**1.0 ml (800 µl R_1 + 200 µl R_2)**

Mix and aspirate. After the initial delay of 60 seconds, record the absorbance of the test at an interval of 30 seconds for the next 90 seconds at 340 nm. Determine the mean change in absorbance per minute and calculate test results.

Calculation:

Activity of GOT (AST) in IU/l = Δ Abs./min. x 3339

Conversion factors :

Following factors can be used for conversion of IU/l from one temperature to another :

Temperature Conversion

From 37°C to 30°C : 0.67
From 37°C to 25°C : 0.49

Note : Since temperature conversion factors are given only as an approximate conversion, it is suggested that values be reported at the temperature of measurement.

EXPECTED VALUES

Serum / Plasma

Temperature	at 25°C	at 30°C	at 37°C
MEN	≤ 18 IU/l	≤ 25 IU/l	≤ 37 IU/l
WOMEN	≤ 15 IU/l	≤ 21 IU/l	≤ 31 IU/l

Expected range varies from population to population. It is therefore recommended that each laboratory should establish its own normal range.

LO-2017-01-004

PROCEDURE LIMITATIONS

1. If the GOT (AST) activity exceeds 800 IU/l, dilute the specimen with normal saline and repeat the assay. The result obtained should then be multiplied with the dilution factor to obtain correct GOT (AST) activity.
2. The working solution is considered unsatisfactory and should not be used if the absorbance is less than 0.900 at 340 nm. against distilled water.

QUALITY CONTROL

To ensure adequate quality control, it is recommended that each batch should include normal and an abnormal commercial reference control serum. It should be realised 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 and accuracy of pipetting.

REFERENCES

1. Tietz, N.W., ***Clinical Guide to Laboratory Tests,*** 3rd ed. Philadelphia, Pa : W.B. Saunders, 1995 : 76 - 77.
2. Bergmeyer, H.U., Horder, M., Rej, R., Approved recommendation (1985) on IFCC methods for the measurement of catalytical concentration of enzymes, Part 3. IFCC method for L - aspartate aminotransferase. ***J. Clin. Chem. Clin. Biochem.*** 1986; 24 : 497 - 510.
3. Fischbach, F., Zawta, B., Age - dependent Reference Limits of Several Enzymes in Plasma at Different Measuring Temperatures. ***Clin. Lab.*** 1992; 38-555 - 561.
4. Penttila, I.M., et al, ***Scand. J. Clin. Lab. Invest.*** 35, 275 (1975).
5. Hafkensheild, J.C.M., et al. ***J. Clin. Chem. Clin. Biochem.*** 17, 219 (1979)
6. In-house test data. ***Accurex Biomedical Pvt. Ltd.,*** 2003.

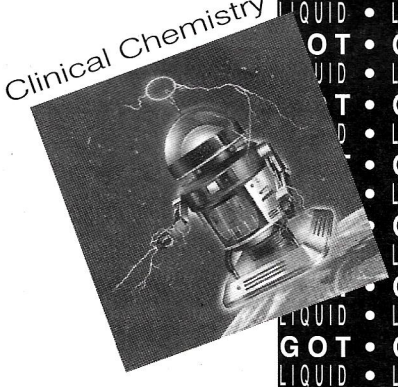
IVD	*In Vitro* Diagnostic Use		Date of Manufacturing
	Consult Instructions for use		Use by (YYYY-MM-DD)
REF	Catalogue Number		Temperature Limitation
LOT	Batch Code		Manufacturer

CE

AR. No.: I 57

LO-2017-01-004

Liquid dispensing facility



Infinite

GOT (AST)

UV-Kinetic