**ENZOPAK**

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# SGOT (AST) LIQUID

**(IFCC METHOD)**

**Reagent kit for quantitative estimation of glutamate oxaloacetate transaminase activity in serum or plasma.**

- Long shelf life.
- Liquid stable reagents available.
- International Standard 'IFCC' reagent.

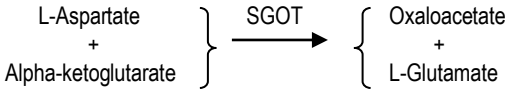
**BACKGROUND & SYNOPSIS:**

In 1955, Karmen published a method for the determination of glutamate oxaloacetate transaminase activity (also called aspartate transaminase AST). The primary transaminase reaction was coupled with malate dehydrogenase (MDH) and reduced nicotinamide adenine dinucleotide (NADH). This method was further improved upon by many workers and reviewed by professional societies like SCE, IFCC, GSCC etc.

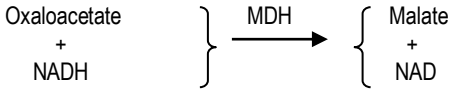
ENZOPAK SGOT-L is based on the procedure recommended by the IFCC.

**PRINCIPLE :**

1) In this reaction, L-Aspartate and alpha-ketoglutarate react in the presence of GOT with the sample to yield oxaloacetate and L-glutamate.




2) The oxaloacetate is reduced by malate dehydrogenase (MDH) to yield L-malate with the oxidation of NADH to NAD. The reaction is monitored by measurement of the decrease in absorbance of NADH at 340 nm.




The rate of reduction in absorbance is proportional to SGOT activity in sample.

**DIAGNOSTIC SIGNIFICANCE:**

Aspartate transaminase is present in all human tissues of the body. It is also present in large amounts in liver, kidneys, heart and skeletal muscles. When any of these organs is damaged or diseased, serum GOT level rises. The rise is proportional to the extent of damage or disease. Elevated levels are associated with liver disease or damage, myocardial infarction, muscular dystrophy and cholecystitis. In myocardial infarction GOT/AST levels increase after 3 to 8 hours of onset of attack and returns to normal in 4 to 6 weeks. The duration and extent of increase in level is proportional to the severity of attack. The change in levels over a period of time is useful to the physician in evaluating myocardial infarction, following chronic heart disease or resolving hepatitis.

**REAGENT COMPOSITION**

| **Active Ingredients** | **Concentration** |
|---|---|
| **Reagent-1** | |
| • Buffer pH 7.6 ± 0.1 | 100 mmol/L |
| • MDH (Microbial) | ≥ 500 U/L |
| • LDH (Microbial) | ≥ 900 U/L |
| • L-Aspartate | 210 mmol/L |
| **Reagent-2** | |
| • NADH | ≥ 0.12 mmol/L |
| • 2-oxoglutarate | 13 mmol/L |

Also Contains non-reactive fillers and stabilizers.

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**PRESENTATION:**

| **Pack size** | **1x25ml** | **2x25ml** |
|---|---|---|
| All reagents to be stored at 2-8$^0$C | | |
| • SGOT – L (R1) (Enzyme reagent) | 1x20ml | 2x20ml |
| • SGOT – L (R2) (Substrate reagent) | 1x5ml | 2x5ml |

**PRECAUTIONS:**

ENZOPAK SGOT-L is for *IN-VITRO* diagnostic use only.

**Reagent contains Sodium Azide. DO NOT INGEST.**

**WORKING REAGENT PREPARATION:**

**For 1x10ml, 1x25ml & 2x25 ml:**

Carefully transfer the contents of 1 bottle of R2 into the bottle of R1. Mix well. Wait for 2 minutes before use.

Alternatively for flexibility as much of working reagent may be made as and when desired by mixing 4 parts of R1 & 1 part of R2.

Alternatively 0.8 ml of R1 and 0.2 ml of R2 may also be used instead of 1 ml of working reagent directly during the assay.

**REAGENT STORAGE AND STABILITY:**

ENZOPAK SGOT-L reagents are stable at 2-8$^0$C until the expiry date stated on the label.

The working reagent is stable for 30 days at 2-8$^0$C

**SPECIMEN COLLECTION:**

Fresh, clear serum under fasting condition with no hemolysis is the specimen of choice. Plasma collected with anticoagulants such as heparin or EDTA can also be used.

**PROCEDURE :**

**REACTION PARAMETERS**

- Type of reaction : Kinetic/Decreasing OD
- Wavelength : 340 nm
- Flow cell Temperature : 37$^0$C
- Delay Time : 60 Seconds
- Interval : 30 seconds
- No. of Intervals : 4
- Sample Volume : 50µl (0.05 ml)
- Working Reagent Volume : 1.0 ml
- Factor : 3376
- Light Path : 1 cm
- Zero setting with : Distilled Water

**One Reagent Procedure**

| **PIPETTE INTO TEST TUBES** | **TEST** |
|---|---|
| • Working Reagent (ml) | 1.0 |
| • Sample (ml) | 0.05 |

**Two Reagent Procedure**

| **PIPETTE INTO TEST TUBES** | **TEST** |
|---|---|
| • SGOT-L R1 (ml) | 0.8 |
| • SGOT-L R2 (ml) | 0.2 |
| • Sample (ml) | 0.05 |

Mix well and after incubation at 37$^0$C for 60 seconds, measure the absorbance at an interval of 30 seconds for 2 minutes at 340 nm.

**NOTE:**

For laboratories using instruments with cuvette capacity less than 1.0 ml, sample and working reagent volumes should be proportionately decreased.

**TEST RESULTS:**
Serum GOT activity (IU/L) = ΔA/min. x F
Where F = 3376 (based on the milimolar extinction coefficient of NADH at 340 nm).

**NORMAL VALUES:**
SGOT : 0-55 IU/L

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

**REFERENCES:**

- The Committee on Enzymes of the Scandinavian Society for Clinical Chemistry & Clinical Physiology. Recommended methods for determination of four enzymes in blood. Scand. J. Clin. Lab. Invest.33.291 (1974).
- LADUE.J.S. WROBLEWSKI. F. AND KARMEN. A. Serum glutamate oxaloacetate transaminase in human acute transmural myocardial infarction. Science 120.497-499 (1954).



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