

SGOT (DST)

ENZOPAK

Last update 04-2014

(IFCC METHOD)

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

PRODUCT HIGHLIGHTS:

- Long shelf life.
- International Standard 'IFCC' reagent.

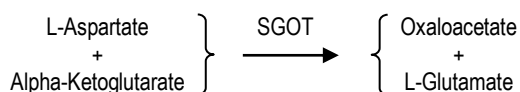
BACKGROUND & SYNOPSIS:

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.

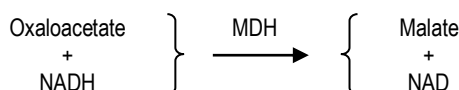
ENZOPAK SGOT is based on the procedure recommended by the IFCC.

PRINCIPLE:

- 1) In this reaction L-Aspartate and Alpha-Ketoglutarate react in the presence of GOT in 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 GOT 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	
• NADH Na ₂	0.1 mmol/L
• LDH	>1000 U/L
• MDH	> 1000 U/L
Reagent-2	
• Buffer	50 mmol/L
• L-Aspartic Acid	150 mmol/L
• α-KG	10 mmol/L

pH 8.0± 0.1 at 25°C

Also contains non-reactive fillers and Stabilizers.

PRESENTATION:

All Reagents: Store at 2-8°C

Pack Size	No. of Bottles/Vials	
	1 SGOT (Enzyme/Coenzyme) (2x10 Tablets)	2 SGOT (Buffer Substrate)
20 x 1.1 ml		2
5 x 10 ml	5	1
5 x 20 ml	5	5
4 x 50 ml	4	4

For 20x 1.1ml & 5x10 ml Reconstitution Vial Provided

PRECAUTION:

ENZOPAK SGOT is for *IN-VITRO* diagnostic use only.
Reagent contains Sodium Azide, DO NOT INGEST.

WORKING REAGENT PREPARATION:

FOR 20 x 1.1 ml.:

Add 1.1 ml. of 2 SGOT to one tablet of 1 SGOT. Mix well to dissolve and wait for 15 minutes prior to use. The working reagent is stable for 30 days at 2-8°C.

FOR 5 x 10 ml. :

Carefully transfer the content of 1 SGOT (Powder) into the bottle containing 10 ml of 2 SGOT (Buffer). Mix gently to dissolve completely. Wait for 5 minutes before use. The working reagent is stable for 4 months at 2-8°C.

FOR 5 x 20 ml. :

Carefully transfer the content of 1 SGOT (Powder) into the bottle containing 20 ml of 2 SGOT (Buffer). Mix gently to dissolve completely. Wait for 5 minutes before use. The working reagent is stable for 4 months at 2-8°C.

FOR 4 x 50 ml. :

Carefully transfer the content of 1 SGOT (Powder) into the bottle containing 50 ml of 2 SGOT (Buffer). Mix gently to dissolve completely. Wait for 5 minutes before use. The working reagent is stable for 4 months at 2-8°C.

REAGENT STORAGE AND STABILITY:

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

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 may be used.

PROCEDURE:

REACTION PARAMETERS

• Type of Reaction	:	Kinetic / Decreasing OD
• Wavelength	:	340 nm
• Flowcell Temperature	:	37°C
• Delay Time	:	60 seconds
• Interval	:	30 seconds
• No. of Readings	:	4
• Sample volume	:	100 µl (0.1ml)
• Working reagent volume	:	1.0 ml
• Factor	:	1746
• Light Path	:	1.0 cm.
• Zero setting with	:	Distilled water



PIPETTE INTO TEST TUBES		TEST
• WORKING REAGENT	(ml)	1.0
• SAMPLE	(ml)	0.1

Mix and after incubation at 37°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) = $\Delta A/\text{min.} \times F$
Where = 1746 (based on the millimolar Extinction coefficient of NADH at 340 nm).

NORMAL VALUES :

GOT : 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 and 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 infraction, Science 120, 497-499 (1954).
- IFCC Method for L-Aspartate aminotransferase. J. Clin. Chem. Clin.Biochem. 1986; 497-510.