SGOT (ASAT)

(Mod. IFCC method)

INTRODUCTION

Aspartate aminotransferase (ASAT, AST, GOT) is an enzyme participated in amino acids metabolism. ASAT is found in all tissues but particularly high level of ASAT is observed in heart muscle, skeletal muscle, liver and kidney. Elevated ASAT serum level is a marker of myocardial infarction and kidney, liver or skeletal muscle in jury.

METHOD PRINCIPLE

Optimized, modified method according to International Federation of Clinical Chemistry (IFCC), without pyridoxal phosphate.

L-aspartate + 2-oxoglutarate < ASAT> oxalacetate + L-glutamate oxalacetate + NADH + H+ < MDH> malate + NAD+

The rate of absorbance changing at I=340 nm is directly proportional to aspartate aminotransferase activity.

KIT CONTENTS

| Reagent Name | Pack Size |
|-----------------|-----------|
| R1 SGOT reagent | 2 x 40 ml |
| R2 SGOT reagent | 2 x 10 ml |

The reagents when stored at 2-8°C are stable up to expiry date printed on the package. The reagents are stable for 4 weeks on board the analyser at 2-10°C. Protect from light and avoid contamination.

WORKING REAGENT PREPARATION AND STABILITY

Assay can be performed with use of separate R1-ASAT and R2-ASAT reagents or with use of working reagent. For working reagent preparation mix gently 4 parts of R1-ASAT with 1 part of R2-ASAT. Avoid foaming.

Stability of working reagent: 4 weeks at 2-8 °C

5 days at 15-25°C

Protect from light and avoid contamination.

CONCENTRATIONS IN THE TEST

WARNINGS AND NOTES

- Product for in vitro diagnostic use only.
- The reagents contain 0.09% sodium azide as a preservative. avoid contact with skin and mucous membranes.
- The reagents are usable when the absorbance of the working reagent is higher than 1.00 (read against distilled water, wavelength = 340 nm, cuvettel=1 cm, attemp. 25°C).

ADDITIONAL EQUIPMEN

- Automatic analyzer or photometer able to read at 340 nm (Hg 334nm, 365nm)
- Thermostat at 25°C, 30°C or 37°C
- General laboratory equipment



SPECIMEN

Serum, heparinized or EDTA plasma free from hemolysis. Do not use heparine ammonium salt.

Hemolysis should be avoided, since ASAT activity in erythrocytes is 10 times higher than in normal serum.

ASAT activity remains stable in specimen up to 1 day at 15-25°C or up to 7 days at 2-8°C. Although samples frozen at -20°C can be stored up to 1 month, it is recommended to perform the assay with freshly collected samples.

PROCEDURE

These reagents may be used both for manual assay and in several automatic analysers. Programme Sheets are available on request.

Wavelength 340 nm
Temperature 37°C
Cuvette 1 cm

Pipette into the cuvette:

| Reagent | Test (T) |
|--------------------------------------|----------|
| R1 SGOT reagent | 800 µ1 |
| R2 SGOT reagent | 200 μ1 |
| Bring to assay temperature, then add | |
| Sample | 100 μ1 |

Mix and incubate at adequate temperature. After about 1 min. read the absorbance against air or water. Repeat the reading after exactly 1, 2 and 3 minutes. Calculate the mean absorbance change per minute ($\Delta A/min.$).

CALCULATION

ASAT activity $[U/I] = \Delta A/min. x 1746$

REFERENCE VALUES

| Female | upto 31 U/L |
|--------|-------------|
| Male | upto 37 U/L |

It is recommended for each laboratory to establish its own referenceranges for local population.

OUALITY CONTROL

To ensure adequate quality control, each run should include assayed normal and abnormal controls. If commercial controls are not available it is recommended that known value samples be aliquoted, frozen and used as controls.

PERFORMANCE CHARACTERISTICS

- Sensitivity / Limit of Quantitation: 5 U/L.
- · Linearity: up to 500 U/L.
- Specificity / Interferences: Haemoglobin up to 2.5 g/dl, ascorbate up to 7.75 mg/l, bilirubin up to 20.0 mg/dl and triglycerides up to 500 mg/dl do not interere with the test.
- The working reagent should not be used, if the absorbance is less than 0.900 at 340 nm. against distilled water

WASTE MANAGEMENT

Please refer to local legal requirements.

LITERATURE

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- 4. Bergmeyer H.U., Horder M., Rej R.: J. Clin. Chem. Clin. Biochem. 24, 497(1986).
- Tietz N.W., ed. Clinical Guide to Laboratory Tests, 3rd ed. Philadelphia, PA: W.B. Saunders, 76 (1995).

SYSTEM PARAMETERS

| Method | Kinetic |
|------------------------|------------------|
| Wavelength | 340 nm |
| Zero Setting | distilled water |
| Temperature Setting | 37° C |
| Incubation Temperature | 37° C |
| Incubation Time | |
| Delay Time | 60 sec |
| Read Time | 180 sec |
| No. of Reading | 3 |
| Interval Time | 60 sec |
| Sample Volume | 0.1 ml (100 ul) |
| Reagent Volume | 1.0 ml (1000 ul) |
| Units | U/L |
| Factor | 1746 |
| Reaction Slope | Decreasing |
| Linearity | 500 U/L |



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