# GOT ASAT IFCC LS 4+1

IVD

Quantitative determination of ASAT (GOT) in Serum or Plasma. UV kinetic method, IFCC optimized.

REF **CC1214** R1: 3x20 mL R2: 1x15 mL REF **CC1212** R1: 3x40 mL R2: 1x30mL REF **CC1210** R1: 3x80 mL R2: 1x60 mL

## METHOD AND PRINCIPLE

Optimized UV-test according to IFCC (International Federation of Clinical Chemistry).

GOT / ASAT catalyzes the transformation of 2-Oxiglutarate and L-Aspartate into L-Glutamate and Oxaloacetate. Subsequently, the Malate-Dehydrogenase (MDH) catalyzes the transformation of the formed Oxalacetate and of the NADH into L-Malate and $NAD^+$ causing a decrease in Absorbance (Abs) of the reaction which is measured at 340 nm. The rate of decrease of the Abs is proportional to the activity of the GOT / ASAT enzyme in the sample.

$$\text{L-Aspartate + 2-Oxoglutarate} \xrightarrow{\text{ASAT (GOT)}} \text{L-Glutamate + Oxalacetate}$$

$$\text{Oxalacetate + NADH + H}^+ \xrightarrow{\text{MDH}} \text{L-Malate + NAD}^+ \text{ + H}_2\text{O}$$

## CLINICAL SIGNIFICANCE

The group of enzymes called transaminase exist in tissues of many organs. Necrotic activity in these organs causes a release of abnormal quantitaties of enzyme into the blood where they are measured. Since heart tissue is rich in GOT/ASAT increased serum levels appear in patients after myocardial infarction, as well as in patients with muscle disease, muscular dystrophy and dermatomyositis. The liver is specially rich in GPT/ALAT, being this enzyme measurement used primarily as a test for infectious and toxic hepatitis, although high levels of both GPT/ALAT and GOT/ASAT may also be found in cases of liver cell damage and acute pancreatitis, suggesting that the obstruction of the biliary tree by the edematous pancreas and the presence of associate hepatic disease may contribute to elevated GOT/ASAT levels in these patients. Slight or moderate elevations of GOT/ASAT and GPT/ALAT activities may be observed after intake of alcohol and after administration of various drugs, such as salicylates, opiates and ampicillin.

## REAGENT COMPOSITION

**Reagent (R1)**

| | |
|---|---|
| TRIS, pH 7.65 | 80 mmol/L |
| L-Aspartate | 240 mmol/L |
| MDH (malate dehydrogenase) | ≥ 600 U/L |
| LDH (lactate dehydrogenase) | ≥ 900 U/L |

**Reagent (R2)**

| | |
|---|---|
| 2-Oxoglutarate | 12 mmol/L |
| NADH | 0.18 mmol/L |

## REAGENT PREPARATION AND STABILITY

Liquid and ready to use reagents, stable until the expiry date shown, if stored as indicated on the label and avoid contamination, evaporation and prolonged exposure to direct light.

Do not freeze the reagents.

Discard the reagents if signs of deterioration appear, such as the presence of particles and turbidity or failure to recover the values of certified control sera or Blank Absorbance (Abs) of Working Reagent at 340 nm < 1.000 in 1 cm cuvette.

After opening the bottles, it is advisable to withdraw the necessary volume, to immediately close the bottles and store them in the fridge in order to avoid contamination, degradation from direct light and evaporation.

For the Sample Starter Procedure, prepare a Work Solution by mixing 4 parts of R1 and 1 part of R2 (E.g. 20 mL of R1 + 5 mL of R2). Stability: Stability: 5 days at 15-25 ° C, 30 days at 2-8 ° C.

For the Substrate Starter Procedure, reagents R1 and R2 are ready to use and stable until the expiry date if stored at the temperature shown on the label and avoid contamination, prolonged exposure to direct light and evaporation.

## SPECIMEN

Serum, heparin plasma or EDTA plasma.

Avoid hemolysate or lipemic samples. Separate the serum from the clot quickly.

Loss of activity within 3 days:at 2° - 8 °C < 8 %; at 15° - 25 °C < 10 %.

Stability: 8 hours at 20-25° C, 5 days at 2-8° C, 3 mounths at -20 °C.

Discard contaminated specimens.

## PROCEDURE

| | |
|---|---|
| Wavelength: | 340 nm |
| Temperature: | 37°C |
| Measurement: | against distilled water |

Sample Start procedure:

| | |
|---|---|
| Working Reagent | 1000 µL |
| Sample | 100 µL |

Mix, read Absorbance (Abs) after 1 minute and start stopwatch. Read Absorbance again after 1, 2 and 3 minutes. Calculate ΔAbs/min (average).

Substrate Start procedure:

| | |
|---|---|
| Reagent (R1) | 800 µL |
| Sample | 100 µL |

Mix and after 1 minute add reagent (R2):

| | |
|---|---|
| Reagent (R2) | 200 µL |

Mix, read Absorbance (Abs) after 1 minute and start stopwatch. Read Absorbance again after 1, 2 and 3 minutes.Calculate ΔAbs/min (average).

## CALCULATION

Calculation Factor (reading at 340 nm in 1 cm cuvette):

GOT-ASAT (U/L) = ΔAbs/min x 1746

Multiparametric Calibrator:

Calculate a specific factor using a certificate multiparametric calibrator:

$$\text{Factor} = \frac{\text{Calibrator Concentration}}{\Delta\text{Abs/min (average).}}$$

GOT-ASAT (U/L) = ΔAbs/min x Factor

Conversion Factor: U/L x 0.0167 = µKat/L= µmol/sec/L

## CALIBRATION

The results depend on the accuracy of the calibration, on the correct setting of the test on the instrument, on the correct volumetric ratio of reagent / sample and on the correct analysis temperature.
Use **MTD Diagnostics Calibrator:**

**Chemistry Multicalibrator - REF CAL1010 (10 x 3 mL)**

## QUALITY CONTROL

Normal and abnormal control sera of known enzymatic activity should be analysed routinely with each group of unknown samples utilizing **MTD Diagnostics** Quality Control Material:

**Chemistry Control N - REF CNN1010** 10 x 5 mL (Level 1)
**Chemistry Control P - REF CNP1020** 10 x 5 mL (Level 2)

## EXPECTED VALUES

Women < 31 U/L
Men < 35 U/L

Each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference range. For diagnostic purposes, the results should always be assessed in conjunction with the patient's medical history, clinical examination and other findings.

## PERFORMANCE

PRECISION:
Low Level: Samples= 20; Average = 47; S.D. = 0.98; CV = 2.08%
High Level: Samples = 20; Average = 196; S.D. = 2.41; CV = 1.23%

ACCURACY (CORRELATION): A comparison between this method (x) and a certified method of trade (y) gave the following correlation:

**y=1.04x + 0.069 ; r=0.998**

SENSITIVITY: 2 U/L

LINEARITY: 2 - 300 U/L

## SPECIFICITY / INTERFERENCES

No interference was observed by Bilirubin up to 20 mg/dL, Haemoglobin up to 500 mg/dL and Lipemia up to 1000 mg/dL Triglycerides.

## NOTES

1. This method may be used with different instruments. Any application to an instrument should be validated to demonstrate that results meet the performance characteristics of the method. It is recommended to validate periodically the instrument. Contact your distributor for any question on the application method.
2. Clinical diagnosis should not be made on findings of a single test result, but should integrate both clinical and laboratory data.

## PRECAUTIONS

The product does not contain any other hazardous substances or mixtures according to EC Regulation No. 1272/2008 (CLP) or their concentrations are such that they are not considered to be persistent, bioaccumulative or toxic (PBT). Therefore, it is not subject to the special labeling required by the aforementioned regulation. The product is labeled according to the directive for CE marking (98/79 / EC). Sodium Azide less than 0.1%.

However, in compliance with the normal prudential rules that everyone has to keep managing any chemical or laboratory reagent, in case of contact of reagents with the operator, you must apply the following first aid:
S26 (P305 – P351 – P338): In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.
S28 (P302 – P352): After contact with skin, wash immediately with plenty of water.
S36/37/39 (P280): Wear suitable protective clothing, gloves and eye/face protection.
S46 (P301 – P310): If swallowed, seek medical advice immediately and show container or label. If victim is conscious and alert, wash out mouth with water.
S56 (P273): Dispose of this material and its container at hazardous or special waste collection point.
S63 (P304 – P340): In case of accident by inhalation: remove casualty to fresh air and keep at rest. If breathing is difficult, give oxygen and get medical aid.

All samples, calibrators and controls should be treated as potentially infectious material capable of transmitting HIV and hepatitis

FOR MORE INFORMATION, REQUEST SAFETY DATA SHEET FOR REAGENT (MSDS) AT THE MANUFACTURER.

## SIMBOLOGY

| | | | |
|---|---|---|---|
| CE | CE Mark (EC Directive 98/79) | | |
| IVD | In Vitro Diagnostic | | Temperature Limitation |
| | Consult instructions for use | | Contains sufficient for <n> test |
| REF | Catalog Number | | Use By |
| LOT | Batch Code | | Manufacturer |

## BIBLIOGRAPHY

Moss DW, Henderson AR. Clinical enzymology. In: Burtis CA, Ashwood ER, editors. Tietz Textbook of Clinical Chemistry. 3rd ed. Philadelphia: W.B Saunders Company; 1999. p. 617-721.

Thomas L. Alanine aminotransferase (ALT), Aspartate aminotransferase (AST). In: Thomas L, editor. Clinical Laboratory Diagnostics. 1st ed. Frankfurt: TH-Books Verlagsgesellschaft; 1998. p. 55-65.

Schumann G, Bonora R, Ceriotti F, Férard G et al. IFCC primary reference procedure for the measurement of catalytic activity concentrations of enzymes at 37 °C. Part 5: Reference procedure for the measurement of catalytic concentration of aspartate aminotransferase. Clin Chem Lab Med 2002;40:725-33.