

ASPARTATE AMINOTRANSFERASE (AST-GOT)

Determination of aspartate aminotransferase in serum and plasma on IFCC recommendation

TEST SUMMARY

The enzyme aspartate aminotransferase catalyzes the transaminase between L-Aspartate and alphachetoglutarate. The 2-Oxaloacetate formed, is reduced to malate in presence of MDH. As the reaction proceed, NADH is oxidized to NAD. The disappearance of NADH per unit time is followed by measuring the decrease of absorbance at 340 nm.

SAMPLES

Serum, plasma. Avoid hemostasis during collection. Stability: until 4 days at 2-8°C or 1 month at -20°C.

REAGENTS

Reagent A: Tris buffer 80 mM pH 7.65; preservatives and stabilizers.

Reagent B: L-aspartate 240 mM, alphachetoglutarate 12 mM, NADH 0.18 mM, MDH ≥ 600 U/l, LDH ≥ 900 U/l.

MATERIAL REQUIRED BUT NOT SUPPLIED

Normal laboratory equipment. Spectrophotometer UV/VIS with thermostatisation. Automatic Micropipette. Cuvette in optical glass or monouse in optical polystyrene. Physiologic solution.

PRECAUTIONS

Reagent may contain not reactive and conservative components. It is opportune to avoid contacts with the skin and do not swallow.

Perform the test according to the general "Good Laboratory Practice" (GLP) guidelines.

REAGENTS PREPARATION

PROCEDURE STARTER SAMPLE

Add 10 ml of Reagent B to a vial of Reagent A.

Stability: ≥ 30 days at 2-8°C away from light source.

PROCEDURE STARTER REAGENT

Use reagents separately. Stability: until expiration date on label, away from light source.

Stability after first opening: ≥ 60 days.

PROCEDURE (STARTER SAMPLE)

Kind of analysis: Kinetics (decreasing)
Reading time: 90,150,210 seconds
Delay: 90 sec.
Wavelength: 340 nm
Temperature: 37°C
Lightpath: 1 cm
Zero: Distilled Water

REAGENTS	CUVETTE
Work Reagent	1 ml
Preincubate at 37 °C at least for 5 minutes	
Samples	100 µl

PROCEDURE (STARTER REAGENT)

Kind of analysis: Kinetics (decreasing)
Reading time: 90,150,210 seconds
Delay: 90 sec.
Wavelength: 340 nm
Temperature: 37°C
Lightpath: 1 cm
Zero: Distilled Water

REAGENTS	CUVETTE
Reagent A	1 ml
Sample	125 µl
Preincubate at 37 °C at least for 5 minutes	
Reagent B	250 µl

CALCULATION

Activity in U/l: $\Delta A/\text{min} \times 1746$

Activity in µkat/l: $U/l \times 0.0167$

EXPECTED VALUES

Men <35 U/l (<0.58 µkat/l)
Women <31 U/l (<0.52 µkat/l)

Every laboratory should establish own reference intervals in accordance with own population.

NOTES

- If the results are incompatible with clinical presentation, they have to be evaluated within a total clinical study.
- Only for IVD use.

CALIBRATION/QUALITY CONTROL

It is suggested to perform an internal quality control. For this purpose the following control sera on human base are available on request:

QN 0050 CH 10 x 5 ml
Control Sera normal values

QP 0050 CH 10 x 5 ml
Control Sera pathological values

TEST PERFORMANCE

Precision

Intra-assay (n = 10)	Mean (U/l)	SD (U/l)	CV%
Sample 1	38.88	0.56	1.40
Sample 2	132.50	0.96	0.70

Inter-assay (n = 20)	Mean (U/l)	SD (U/l)	CV%
Sample 1	37.87	0.46	1.20
Sample 2	134.48	1.48	1.10

Sensitivity/limit of detection

The method is able to discriminate until 0.47 U/l.

Linearity

The method is linear up to 440 U/l.

If $\Delta A/\text{min}$ of 0.200, it is suggested to dilute the sample 1+9 with saline and to repeat the test, multiplying the results by 10.

Methods comparison

A comparison with a commercial available product gave the following results in a comparison on 112 samples:

AST/GOT LTA = x
AST/GOT competitor = y
n = 112

$y = 0,986x + 1,636 \text{ U/l}$ $r = 0,997$

Interferences

No interference was observed by the presence of :

hemoglobin ≤ 400 mg/dl
bilirubin ≤ 25 mg/dl
lipids ≤ 500 mg/dl

WASTE DISPOSAL

Product is intended for professional laboratories. Waste products must be handled as per relevant security cards and local regulations.

PACKAGING

CODE CC00500 (200 TESTS)
Reagent A 4 x 40 ml (liquid)
Reagent B 1 x 40 ml (liquid)






REFERENCES

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HU Bergmeyer – Methods of enzymatic analysis, (1987).
CCLM 2002; 40(7):725-733, Schumann et al. – IFCC reference procedure for alanine aminotransferase.

MANUFACTURER

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SYMBOLS

- IVD** Only for IVD use
- LOT** Lot of manufacturing
- REF** Code number
-  Storage temperature interval
-  Expiration date
-  Warning, read enclosed documents
-  Read the directions
-  Biological risk

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