



AST (GOT)

Aspartate aminotransferase

E.C.2.6.1.1.

UV Method (According to IFCC)

Cat.No. 101-0325

Size 20 x 2.5 ml

Cat.No. 101-0256

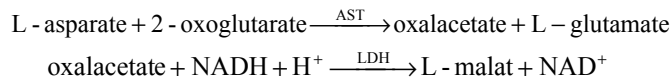
Size 10 x 10 ml

Cat.No. 101-0227

Size 8 x 100 ml

PRINCIPLE:

AST catalyzes the transfer of amino group from L-aspartate to 2-oxoglutarate resulting in the formation of oxalacetate and L-glutamate. The oxalacetate thus formed undergoes reduction with simultaneous oxidation of NADH to NAD in the malate dehydrogenase catalyzed indicator reaction. Oxidation of NADH causes a decrease in absorbance at 340 nm (334 nm or 365 nm) and the rate of absorbance change is directly proportional to AST activity. LDH is included in the reagent to prevent interference from endogenous pyruvate which is normally present in serum samples at low concentrations.



SAMPLE:

Serum, heparinized or EDTA plasma.

Stability 7 days at +2 °C to +8 °C.

REAGENTS:

1. Reagent 1 (Buffer/Substrate)	
Tris buffer, pH 7.8	80 mmol/L
L-aspartate	200 mmol/L
2. Reagent 2 (Enzyme/Coenzyme/ α -oxoglutarate)	
α -oxoglutarate	12 mmol/L
MDH	600 U/L
LDH	800 U/L
NADH	0.18 mmol/L

Store at +2 °C to +8 °C.

REAGENT PREPARATION :

Dissolve the contents of Reagent 2 with the corresponding volume of Reagent 1 (buffer/substrate).

Working reagent is stable for 30 days at +2 °C to +8 °C or 4 days at +15 °C to +25 °C.

PROCEDURE:

Method:	Kinetic (decreasing)
Wavelength:	340, 334, 365 nm
Cuvette:	1 cm light path
Temperature:	25 °C, 30 °C, 37 °C
Preincubation:	1 min
Zero:	air or H ₂ O
Read:	every 60 sec. during 3 min.

Pipette into cuvette:	
Sample	0.1 ml
Working reagent	1.0 ml
Mix well, read initial absorbance after 1 min.	
Read again after every 60 sec during 3 min.	
Calculate $\Delta A/\text{min}$.	

CALCULATION:

$$\frac{\Delta A/\text{min} \times 10^6 \times TV}{6.3 \times 10^3 \times 1 \times V} = \Delta A/\text{min} \times F = \text{U/L}$$

Where is :

ΔA	=	change in absorbance
min	=	minute
6.3×10^3	=	molar absorptivity of NADH at 340 nm
10^6	=	conversion of mol to μmol
l	=	light path in cm
TV	=	total reaction volume in ml
V	=	sample volume in ml

$\Delta A_{340 \text{ nm}}/\text{min} \times 1746 = \text{U/L AST}$

$\Delta A_{334 \text{ nm}}/\text{min} \times 1780 = \text{U/L AST}$

$\Delta A_{365 \text{ nm}}/\text{min} \times 3235 = \text{U/L AST}$

U/L x 16.67 = nkat/L

EXPECTED VALUES:

	25 °C	30 °C	37 °C	UNIT
Men	up to 19 up to 317	up to 26 up to 433	up to 38 up to 623	U/L nkat/L
Women	up to 16 up to 267	up to 22 up to 367	up to 31 up to 517	U/L nkat/L

LINEARITY:

up to 270 U/L (4500 nkat/L)

NOTE:

- If the absorbance change per minute ≥ 0.150 at 340 nm and 334 nm or ≥ 0.08 at 365 nm dilute the sample 1:10 with physiological solution and reassay (result x 10).
- Avoid hemolysis as it interferes with assay.
- Solution 1 contains sodium azide. Avoid ingestion or contact with skin or mucous membranes.

QUALITY CONTROL:

CONTRO-N	20 x 5 ml	Cat. No. 101-0083
CONTRO-P	20 x 5 ml	Cat. No. 101-0084

REFERENCES:

- Bergmeyer, H.U., Bowers, G.N., et al. Clin.Chim. Acta 70, 19-42 (1976) F and 21-22 (1977) F.
- Bergmeyer, H.U. and Wahlefeld, A. Clin.Chem. 24, 58-73 (1978).