BIOLABO
www.biolabo.fr
MANUFACTURER:
BIOLABO SAS,
Les Hautes Rives
02160, Maizy, France

AST GOT (IFCC) Single vial

Reagent for quantitative determination of Aspartate amino transferase activity [EC 2.6.1.1] in human serum and plasma

REF 80025 **R1** 20 X 10 mL	REF 80125 **R1** 8 x 30 mL	REF 80225 **R1** 10 x 125 mL

TECHNICAL SUPPORT AND ORDERS
Tel: (33) 03 23 25 15 50
support@biolabo.fr
Latest revision : www.biolabo.fr

CE

IVD

Made in France

I: corresponds to significant modifications

INTENDED USE

This reagent is designated for professional use in laboratory (manual or automated method).
I It allows the quantitative determination of aspartate amino transferase (AST) [EC 2.6.1.1] to screen its level in human serum and plasma.

GENERALITIES (1) (2)

AST is distributed in all body tissues, but greatest activity occurs in liver, heart, skeletal muscle and in erythrocytes. Minimal activity occurs in skin, kidney and pancreas. Although serum levels of both AST and ALT become elevated whenever diseases processes affecting liver cells integrity (viral hepatitis, liver necrosis and cirrhosis), an increased AST activity in serum or plasma appears in more than 97% of cases of myocardial infarction. AST levels (and occasionally ALT) are also elevated in progressive muscular dystrophy, pulmonary emboli, acute pancreatitis…

PRINCIPLE (4) (5)

Method developed by Karmen and Al, and optimised by Henry and al. (according to modified IFCC recommendations):

L-Aspartate + 2-Oxoglutarate $\xrightleftharpoons{AST}$ Oxaloacetate + L-Glutamate

Oxaloacetate + NADH + H^+ $\xrightleftharpoons{MDH}$ L-Malate + NAD^+

The decrease in absorbance proportional to AST activity in the specimen, is measured at 340 nm.

REAGENTS

R1	**AST (GOT) IFCC**	Reagent 1
EDTA		5 mmol/L
2-Oxoglutarate		12 mmol/L
L-Aspartate		200 mmol/L
MDH		495 UI/L
LDH		820 UI/L
NADH		≤ 0.18 mmol/L
Tris Buffer		80 mmol/L
pH at 30°C		7.80 ± 0.1
Preservative		

Before reconstitution:
Danger. Acute Tox. 2: H300 - Fatal if swallowed,
Aquatic Chronic 3: H412 - Harmful to aquatic life with long lasting effects

P264: Wash hands thoroughly after handling, P270: Do not eat, drink or smoke when using this product, P301+310: IF SWALLOWED: Immediately call a POISON CENTER or doctor/physician, P330: Rinse mouth, P501: Dispose of contents/container in accordance with dangerous waste disposal regulations. Classification due to Sodium Azide < 1 %. For more details, refer to Safety Data Sheet (SDS)

Once reconstituted, working reagent is not classified as dangerous

SAFETY CAUTIONS

- Refer to current Material Safety Data Sheet available on request or on www.biolabo.fr
- Verify the integrity of the contents before use.
- Waste disposal: Respect legislation in force in the country.
- All specimens or reagents of biological origin should be handled as potentially infectious. Respect legislation in force in the country.

Any serious incident that has occurred in connection with the device is notified to the manufacturer and the competent authority of the Member State in which the user and/or patient is based.

REAGENTS PREPARATION

- REF 80025 Use a non-sharp instrument to remove aluminium cap.
- Once opened, add promptly to the contents the amount of demineralised water indicated on the label.
- Mix gently until complete dissolution.

STABILITY AND STORAGE

Stored away from light, well cap in the original vial at 2-8°C, reagent is stable when stored and used as described in the insert:

Unopened:

- Until expiry date stated on the label.

Once reconstituted:

- Working reagent is stable for 60 days when free from contamination.
- Discard any reagent if cloudy or if absorbance at 340 nm is < 1.000.
- Don't use working reagent after expiry date.

SPECIMEN COLLECTION AND HANDLING (2)

Unhemolysed serum. Do not use heparinised plasma

AST is stable in serum or plasma for:

- 24 hours at room temperature
- 28 days at 2-8°C
- at least for 1 year at –20°C.

Adding pyridoxal phosphate (0.1 mM) improves stability at room temperature to 7 days.

LIMITS (3) (6)

LDH contained in reagent allows, during pre-incubation step, reduction of endogenous pyruvate which would positively interfere.

Likewise oxaloacetate, product of the reaction, is carboxilated into pyruvate. This one will also be consumed by LDH contained in reagent and will not interfere with AST determination.

For a more comprehensive review of factors affecting this assay refer to the publication of Young D.S.

MATERIAL REQUIRED BUT NOT PROVIDED

1. Medical analysis laboratory equipment.
2. Spectrophotometer or Biochemistry Clinical Analyzer
3. Demineralised water for reagent preparation

Manufacturer	Expiry date	In vitro diagnostic	Storage temperature	Dematerialized water	Biological risk
Product Reference	See Insert	Batch number	Store away from light	Sufficient for	Dilute with

QUALITY CONTROL

- REF 95010 EXATROL-N Level I
- REF 95011 EXATROL-P Level II
- External quality control program

It is recommended to control in the following cases:

- At least once a run
- At least once within 24 hours
- When changing vial of reagent
- After maintenance operations on the instrument

If control is out of range, apply following actions:

1. Prepare a fresh control serum and repeat the test
2. If control is still out of range, use a new vial of fresh calibrator
3. If control is still out of range, use a new vial of reagent and reassay

If control is still out of range, please contact BIOLABO technical support or your local Agent.

REFERENCE INTERVALS (1) (2)

	(IU/L) 37°C
Newborn	39-117
Infant	23-94
Adult	13-31

Each laboratory should establish its own normal ranges for the population it serves.

PERFORMANCES

On Kenza 240TX, 37°C, 340 nm.

Linearity Range: between 5 and 310 IU/L

Detection limit: approx. 1.3 IU/L

Precision:

Within-run N = 20	*Low level*	*Normal level*	*High level*
Mean (IU/L)	21.8	44.2	171.9
S.D. (IU/L)	0.6	0.7	2.7
C.V. %	2.5	1.6	1.6

Between run N = 20	*Low level*	*Normal level*	*High level*
Mean (IU/L)	22.5	45.3	176.9
S.D. IU/L	0.7	1.1	4.0
C.V. %	3.1	2.5	2.3

Comparison studies with commercially available reagent:
Realised on serum specimens (n=100) between 9 and 313 IU/L

y = 1.0265 x +0.9906 r = 0.9982

Analytical Sensitivity: approx. 0.0063 abs/min for 10 IU/L

Interferences:

Turbidity	No interference up to 0.133 abs
Total bilirubin	Negative interference from 323 µmol/L
Direct bilirubin	No interference up to 328 µmol/L
Ascorbic acid	No interference up to 2500 mg/dL
Glucose	No interference up to 1176 mg/dL
Haemoglobin	Positive interference from 114 µmol/L

Other substances may interfere (see § Limits)

On the board stability: 1 month

Calibration Stability: 8 days

Make a new calibration when changing reagent batch, if quality control results are found out of the established range and after maintenance operations

CALIBRATION

- REF 95015 Multicalibrator traceable to *ERM-AD457/IFCC*

The calibration frequency depends on proper instrument functions and on the preservation of reagent.

PROCEDURE

Manual method

Let stand reagents and specimens at room temperature

Pipette in 1cm pathlength thermostated cuvette	
Reagent 1	1000 µL
Bring at 37°C, then add:	
Calibrator, Control or Specimen	100 µL
Mix. Start a timer. Record initial absorbance after 60 sec at 340 nm. Record the absorbance again every minutes during 180 sec. Measure absorbance change per minute (△Abs/min).	

1- Performances with manual procedure should be validated by user.
2- Kenza applications and other applications proposal are available on request.

CALCULATION

With Seric Muticalibrator:

$$\text{AST Activity} = \frac{(\Delta\text{Abs/min) Specimen}}{(\Delta\text{Abs/min) Calibrator}} \times \text{Calibrator Activity}$$

With Theoretical Factor:

Activity (U/L) = ΔAbs/min x Factor

$$\text{Factor} = \frac{\text{VR} \times 1000}{6.3 \times \text{VE} \times \text{P}}$$

With:
VR = Total reactional volume (mL)
VE = Specimen volume (mL)
6.3 = Molar extinction coefficient for NADH at 340nm
P = Path length (cm).

Example, with manual Procedure,
(Path length 1 cm, 37°C, 340 nm):

IU/L = (ΔAbs/min) x 1746

$$\text{µKat/L} = \frac{\text{IU/L}}{60}$$

REFERENCES

(1) *TIETZ N.W. Text book of clinical chemistry, 3rd Ed. C.A. Burtis, E.R. Ashwood, W.B. Saunders (1999) p. 652-657*
(2) *Clinical Guide to Laboratory Test, 4th Ed., N.W. TIETZ (2006) p. 154-159*
(3) *YOUNG D.S., Effect of Drugs on Clinical laboratory Tests, 4th Ed. (1995) p. 3-68 to 3-79*
(4) *HENRY R. J. and al., Am J clin Path (1960), 34, 381-398*
(5) *IFCC Method for L-Aspartate aminotransferase. J Clin. Chem. Clin. Biochem.(1986), 24, p.497-510.*
(6) *M. MATHIEU and col. SFBC. Comité de Standardisation. Recommandations pour la mesure de l'activité catalytique de l'Aspartate aminotransférase dans le sérum à 30°C. Ann. Biol. Clin. 1976. 34. 291-297*