respons®910

ALAT (GPT) FS* (IFCC mod.)

with/without pyridoxal-5-phosphate

Diagnostic reagent for quantitative in vitro determination of ALAT (GPT) in serum or plasma on DiaSys respons®910

Order Information

Cat. No. 1 2701 99 10 920
4 twin containers for 200 tests each

Pyridoxal-5-phosphate FS
Cat. No. 2 5010 99 10 030
6 x 3 mL

Method

Optimized UV-test according to IFCC (International Federation of Clinical Chemistry and Laboratory Medicine) [modified]

Principle

L-Alanine + 2-Oxoglutarate $\xleftarrow{ALAT}$ L-Glutamate + Pyruvate

Pyruvate + NADH + H^+ $\xleftarrow{LDH}$ D-Lactate + NAD^+

Addition of pyridoxal-5-phosphate (P-5-P) stabilizes the activity of transaminases and avoids falsely low values in samples containing insufficient endogenous P-5-P, e.g. from patients with myocardial infarction, liver disease and intensive care patients [1].

Reagents

Components and Concentrations

R1:	TRIS	pH 7.15	140 mmol/L
	L-Alanine		700 mmol/L
	LDH (lactate dehydrogenase)		≥ 2300 U/L
R2:	2-Oxoglutarate		85 mmol/L
	NADH		1 mmol/L
Pyridoxal-5-phosphate FS			
	Good's buffer	pH 9.6	100 mmol/L
	Pyridoxal-5-phosphate		13 mmol/L

Storage Instructions and Reagent Stability

The reagents are stable up to the end of the indicated month of expiry, if stored at 2 - 8 °C, protected from light and contamination is avoided. DiaSys respons containers provide protection from light. Do not freeze the reagents!

Warnings and Precautions

1. The reagents contain sodium azide (0.95 g/L) as preservative. Do not swallow! Avoid contact with skin and mucous membranes.
2. In very rare cases, samples of patients with gammopathy might give falsified results.
3. Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.

Waste Management

Please refer to local legal requirements.

Reagent Preparation

The reagents are ready to use. The bottles are placed directly into the reagent rotor. For the determination with P-5-P add 350 µL of P-5-P to reagent 1 and mix gently.

Stability after mixing: 6 days at 2 - 8 °C
24 hours at 15 - 25 °C

Specimen

Serum, heparin plasma or EDTA plasma

Stability [2]:
3 days at 20 - 25 °C
7 days at 4 - 8 °C
7 days at -20 °C

Discard contaminated specimens. Freeze only once.

Calibrators and Controls

For calibration, DiaSys TruCal U calibrator is recommended. This method has been standardized against the original IFCC formulation (molar extinction coefficient 340 nm). For internal quality control DiaSys TruLab N and P controls should be assayed. Each laboratory should establish corrective action in case of deviations in control recovery.

	Cat. No.	Kit size
TruCal U	5 9100 99 10 063	20 x 3 mL
	5 9100 99 10 064	6 x 3 mL
TruLab N	5 9000 99 10 062	20 x 5 mL
	5 9000 99 10 061	6 x 5 mL
TruLab P	5 9050 99 10 062	20 x 5 mL
	5 9050 99 10 061	6 x 5 mL

Performance Characteristics

with P-5-P activation

Measuring range up to 600 U/L ALAT (in case of higher activities re-measure samples after manual dilution or use rerun function).	
Limit of detection**	3 U/L ALAT
On-board stability	6 days
Calibration stability	6 days

Interfering substance	Interferences < 10%	ALAT [U/L]
Ascorbate	up to 30 mg/dL	121
Hemoglobin	up to 500 mg/dL	50.9
	up to 850 mg/dL	107
Bilirubin, conjugated	up to 50 mg/dL	49.8
	up to 55 mg/dL	93.8
Bilirubin, unconjugated	up to 45 mg/dL	46.1
	up to 45 mg/dL	85.7
Lipemia (triglycerides)	up to 1000 mg/dL	35.5
	up to 1000 mg/dL	114
For further information on interfering substances refer to Young DS [4].		

Precision			
Within run (n=20)	**Sample 1**	**Sample 2**	**Sample 3**
Mean [U/L]	21.2	47.4	132
Coefficient of variation [%]	2.88	1.41	0.95
Between run (n=20)	**Sample 1**	**Sample 2**	**Sample 3**
Mean [U/L]	19.5	51.2	126
Coefficient of variation [%]	4.02	2.03	1.63

Method comparison (n=107)	
Test x	DiaSys ALAT (GPT) FS (Hitachi 911)
Test y	DiaSys ALAT (GPT) FS (respons®910)
Slope	1.021
Intercept	-1.094 U/L
Coefficient of correlation	0.9996

without P-5-P activation

Measuring range up to 600 U/L ALAT (in case of higher activities re-measure samples after manual dilution or use rerun function)	
Limit of detection**	3 U/L ALAT
On-board stability	4 weeks
Calibration stability	4 weeks

Interfering substance	**Interferences < 10%**	**ALAT [U/L]**
Ascorbate	up to 30 mg/dL	81.1
Hemoglobin	up to 500 mg/dL	36.0
	up to 850 mg/dL	78.1
Bilirubin, conjugated	up to 50 mg/dL	46.7
	up to 55 mg/dL	70.3
Bilirubin, unconjugated	up to 45 mg/dL	33.5
	up to 45 mg/dL	63.5
Lipemia (triglycerides)	up to 1000 mg/dL	40.3
	up to 1000 mg/dL	131
For further information on interfering substances refer to Young DS [4].		

Precision			
Within run (n=20)	**Sample 1**	**Sample 2**	**Sample 3**
Mean [U/L]	20.8	36.4	125
Coefficient of variation [%]	2.12	2.04	1.02
Between run (n=20)	**Sample 1**	**Sample 2**	**Sample 3**
Mean [U/L]	20.3	40.0	122
Coefficient of variation [%]	4.24	2.28	1.67

Method comparison (n=90)	
Test x	DiaSys ALAT (GPT) FS (Hitachi 911)
Test y	DiaSys ALAT (GPT) FS (respons®910)
Slope	1.004
Intercept	-0.161 U/L
Coefficient of correlation	0.9998

** according to NCCLS document EP17-A, vol. 24, no. 34

Conversion factor

ALAT [U/L] x 0.0167 = ALAT [µkat/L]

Reference Range

With pyridoxal-5-phosphate activation

Women [3]		< 34 U/L	< 0.57 µkat/L
Men [3]		< 45 U/L	< 0.75 µkat/L
Children [1]	1 – 30 day(s)	< 25 U/L	< 0.42 µkat/L
	2 – 12 months	< 35 U/L	< 0.58 µkat/L
	1 – 3 year(s)	< 30 U/L	< 0.50 µkat/L
	4 – 6 years	< 25 U/L	< 0.42 µkat/L
	7 – 9 years	< 25 U/L	< 0.42 µkat/L
	10 – 18 years	< 30 U/L	< 0.50 µkat/L

Without pyridoxal-5-phosphate activation

Women	< 31 U/L	< 0.52 µkat/L
Men	< 41 U/L	< 0.68 µkat/L

Each laboratory should check if reference ranges are transferable to its own patient population and determine own reference ranges if necessary.

Literature

1. 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.
2. Guder WG, Zawta B et al. The Quality of Diagnostic Samples. 1st ed. Darmstadt: GIT Verlag; 2001; p. 14-5.
3. 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 alanine aminotransferase. Clin Chem Lab Med 2002; 40: 718-24.
4. Young DS. Effects of Drugs on Clinical Laboratory Tests. 5th. ed. Volume 1 and 2. Washington, DC: The American Association for Clinical Chemistry Press, 2000.
5. 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.

Manufacturer

IVD CE

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ALAT (GPT) FS (IFCC mod.)

Application for serum and plasma samples

This application was set up and evaluated by DiaSys. It is based on the standard equipment at that time and does not apply to any equipment modifications undertaken by unqualified personnel

Identification	
This method is usable for analysis:	Yes
Name:	ALT
Shortcut:	
Reagent barcode reference:	010
Host reference:	

Technic	
Type:	Linear Kinetic
First reagent:[µL]	160
Blanc correction	Yes
Second reagent:[µL]	40
Blanc correction	Yes
Main wavelength:[nm]	340
Secondary wavelength:[nm]	405
Polychromatic factor:	1.000
1 st reading time [min:sec]	6:24
Last reading time [min:sec]	10:00
Reaction way:	Decreasing
Linear Kinetics Substrate deplation: absorbance limit	0.3
Linearity: Maximum deviation [%]	100
Fixed Time Kinetics Substrate deplation: absorbance limit	
Endpoint Stability: largest remaining slope	
Prozone Limit [%]	

Sample	
Diluent	NaCl
Concentration technical limits-Lower	3
Concentration technical limits-Upper	600 (600)[1]
SERUM	
Normal volume [µL]	12
Normal dilution (factor)	1
Below normal volume [µL]	20
Below normal dilution (factor)	1
Above normal volume [µL]	2
Above normal dilution (factor)	1
URIN	
Normal volume [µL]	12
Normal dilution (factor)	1
Below normal volume [µL]	20
Below normal dilution (factor)	1
Above normal volume [µL]	2
Above normal dilution (factor)	1
PLASMA	
Normal volume [µL]	12
Normal dilution (factor)	1
Below normal volume [µL]	20
Below normal dilution (factor)	1
Above normal volume [µL]	2
Above normal dilution (factor)	1
CSF	
Normal volume [µL]	12
Normal dilution (factor)	1
Below normal volume[µL]	20
Below normal dilution (factor)	1
Above normal volume [µL]	2
Above normal dilution (factor)	1

[1]() With p5p

Results	
Decimals	1
Units	U/L
Correlation factor-Offset	0.000
Correlation factor-Slope	1.000

Range	
Genre	Male
Age	
SERUM	>= <=45.0
URINE	
PLASMA	>= <=45.0
CSF	
Genre	Female
Age	
SERUM	>= <=35.0
URINE	
PLASMA	>= <=35.0
CSF	

Contaminants	
Contaminant 1	
Wash with	
Cycle	
Volume [µL]	
Contaminant 2	
Wash with	
Cycle	
Volume [µL]	

Calibrators details	
Calibrator list	**Concentration**
Cal. 1	0
Cal. 2	*
Cal. 3	*
Cal. 4	*
Cal. 5	*
Cal. 6	*

	Max delta abs.
Cal. 1	0.015
Cal. 2	0.005
Cal. 3	
Cal. 4	
Cal. 5	
Cal. 6	
Drift limit [%]	0.8

Calculations	
Model	X degree
Degree	1

* Enter calibrator value