

## GPT (ALT) mod. IFCC

Diagnostic reagent for quantitative in vitro determination of GPT (ALT) in human serum or plasma on photometric systems

REF	Kit Size	Configuration
D03116B	1 x 1.25 L	1 x 1 L R1 + 1 x 0.25 L R2
D94620	5 x 100 mL	4 x 100 mL R1 + 1 x 100 mL R2
D98624	5 x 50 mL	4 x 50 mL R1 + 1 x 50 mL R2
D00640	5 x 25 mL	4 x 25 mL R1 + 1 x 25 mL R2
D98625	5 x 10 mL	4 x 10 mL R1 + 1 x 10 mL R2
D73911	10 x 50 mL	10 x 40 mL R1 + 4 x 25 mL R2
D0428917	5 x 62.5 mL	4 x 62.5mL R1 + 1 x 62.5mL R2
DA0830	5 x 50 mL	5 x 40 mL R1 + 5 x 10 mL R2
DT1030	4 x 62.5 mL	4 x 50 mL R1 + 4 x 12.5mL R2
DK0729	5 x 50 mL	4 x 50 mL R1 + 1 x 50 mL R2
DE1830	4 x 62.5 mL	4 x 50 mL R1 + 4 x 12.5 mL R2
DB20323	4 x 62.5 mL	4 x 50 mL R1 + 4 x 12.5 mL R2

Additionally available:

D98485	5 x 3 mL	Calibrator	Diacal Auto
D98485SV	1 x 3 mL	Calibrator	Diacal Auto
D98481	12 x 5 mL	Control normal	Diacon N
D14481	5 x 5 mL	Control normal	Diacon N
D98481SV	1 x 5 mL	Control normal	Diacon N
D98482	12 x 5 mL	Control abnormal	Diacon P
D14482	5 x 5 mL	Control abnormal	Diacon P
D98482SV	1 x 5 mL	Control abnormal	Diacon P

For professional in vitro diagnostic use only.

### GENERAL INFORMATION

<b>Method</b>	UV, Kinetic, Decreasing Reaction, modified IFCC
<b>Shelf life</b>	15 months
<b>Storage</b>	2 – 8 °C
<b>Wavelength</b>	340 nm, Hg 334 nm, Hg 365 nm
<b>Temperature</b>	37°C
<b>Sample</b>	Serum, EDTA-plasma, heparinized plasma

### INTENDED USE

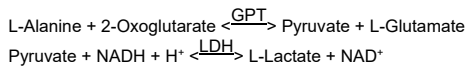
Diagnostic reagent for quantitative in vitro determination of GPT (ALT) in human serum or plasma on photometric systems.

### DIAGNOSTIC SIGNIFICANCE [1, 2]

Alanine Aminotransferase (ALAT/ALT), also called Glutamic Pyruvic Transaminase (GPT) and Aspartate Aminotransferase (ASAT/AST), formerly called Glutamic Oxalacetic Transaminase (GOT) are the most important representatives of a group of enzymes, the aminotransferases or transaminases, which catalyse the conversion of α-keto acids into amino acids by transfer of amino groups.

As liver specific enzyme GPT is only significantly elevated in hepatobiliary diseases, increased GOT levels, however, can occur in connection with damages of heart or skeletal muscle as well as of liver parenchyma. Parallel measurement of GPT and GOT is therefore applied to distinguish liver from heart or skeletal muscle damages. The GOT/GPT ratio is used for differential diagnosis in liver diseases. While ratios < 1 indicate mild liver damage, ratios > 1 are associated with severe, often chronic liver diseases.

### TEST PRINCIPLE



NADH is oxidized to NAD<sup>+</sup>, the resulting decrease in absorbance at 340 nm is directly proportional to the activity of GPT in the sample.

This is a modified formulation for the assay of GPT, as recommended by the IFCC (International Federation of Clinical Chemistry). The IFCC reference method includes pyridoxal phosphate (P-5-P). P-5-P serves as coenzyme in AA transfer and stabilizes the activity of transaminases. Therefore addition of P-5-P 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, 3].

AA	= Amino Acid
GPT	= Glutamate Pyruvate Transaminase
NAD <sup>+</sup>	= Nicotinamide Adenine Dinucleotide
NADH	= reduced NAD
LDH	= Lactate Dehydrogenase

### REAGENT COMPOSITION

COMPONENTS	CONCENTRATION
<b>Reagent 1</b>	
Tris, pH 7.5	138 mmol/L
L-Alanine	709 mmol/L
LDH	1500 U/L
<b>Reagent 2</b>	
2-Oxoglutarate	85 mmol/L
NADH	≥ 1 mmol/L

### MATERIAL REQUIRED BUT NOT PROVIDED

- NaCl solution (9 g/L).
- Clinical chemistry analyser.
- Dialab Pyridoxal-5'-Phosphate in case of determination with P-5-P.

### REAGENT PREPARATION

#### Substrate Start:

Reagents are ready to use.

#### Sample Start:

Mix 4 parts of Reagent 1 with 1 part of Reagent 2.  
(= working reagent)

### STORAGE AND STABILITY

Conditions:	Protect from direct light. Avoid contamination. Close immediately after use. Do not freeze the reagents!
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#### Substrate Start:

Storage:	at 2 – 8 °C
Stability:	up to the expiration date

#### Sample Start (Working Reagent):

Stability:	at 2 – 8 °C	4 weeks
	at 15 – 25 °C	5 days

The working reagent must be protected from light!

### 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. Reagent 1 contains animal material. Handle the product as potentially infectious according to universal precautions and good clinical laboratory practices.
3. In very rare cases, samples of patients with gammopathy might give falsified results [4].
4. Sulfasalazine and sulfapyridine medication may lead to false results in patient samples. Blood collection must be done before drug administration.
5. Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents.
6. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
7. For professional use only!

### SPECIMEN COLLECTION AND STORAGE

Stability [5]:		
In serum/plasma	at 20 – 25 °C	3 days
	at 4 – 8 °C	7 days
	at -20 °C	7 days

Discard contaminated specimens. Freeze only once!

### TEST PROCEDURE

Bring reagents and samples to room temperature.

**Note:** If pyridoxal phosphate (PP) is used, please consult instruction insert for PP before performing test (for Substrate Start only).

#### Substrate Start

Pipette into test tubes	37°C
Reagent 1	1000 µl
Sample	100 µl
Mix. Incubate for 5 min. Then add:	
Reagent 2	250 µl
Mix. Read initial absorbance against air after 1 min. and start a timer. Read absorbance again after exactly 1, 2 and 3 min.	

#### Sample Start (Do not use sample start with P-5-P)

Pipette into test tubes	37°C
Working reagent	1000 µl
Sample	100 µl
Mix. Read initial absorbance against air after 1 min. and start a timer. Read absorbance again after exactly 1, 2 and 3 min.	

### Automation

Special adaptations for automated analysers can be made on request.

### INTERPRETATION OF RESULTS

#### Calculation

**With factor** (light path 1 cm):

From absorbance readings calculate ΔA/min and multiply by the corresponding factor:

$$\text{GPT [U/L]} = \Delta A/\text{min} \times \text{factor}$$

Factors (37 °C):

<b>Substrate Start:</b>	
Factor at 340 nm	2143
Factor at 334 nm	2184
Factor at 365 nm	3971

<b>Sample Start:</b>	
Factor at 340 nm	1745
Factor at 334 nm	1780
Factor at 365 nm	3235

#### With calibrator:

$$\text{GPT [U/L]} = \frac{\Delta A/\text{min Sample}}{\Delta A/\text{min Calibrator}} \times \text{Conc. Calibrator [U/L]}$$

### Unit Conversion

$$\text{GPT [U/L]} \times 0.0167 = \text{GPT [\mu katal/L]}$$

## QUALITY CONTROL AND CALIBRATION

All control sera with GPT values determined by this method can be used. We recommend the Dialab serum controls **Diacon N** (control serum with values in the normal range) and **Diacon P** (control serum with values in the abnormal range). Each laboratory should establish corrective action in case of deviations in control recovery.

### Calibration

The use of a GPT Calibrator is optional.  
 We recommend the Dialab multi calibration serum **Diacal Auto**.

## PERFORMANCE CHARACTERISTICS

### LINEARITY, MEASURING RANGE

On automated systems the test is suitable for the determination of GPT activities up to 600 U/L.  
 In case of manual procedure, the test is suitable for GPT activities which correspond to a maximum  $\Delta A/\text{min} = 0.16$  at 340 nm and 334 nm or 0.08 at 365 nm.  
 Above this concentration the samples should be diluted 1+9 with NaCl solution (9 g/L) and the results multiplied by 10.

### SENSITIVITY/LIMIT OF DETECTION

The lower limit of detection is 4 U/L.

### PRECISION (at 37 °C)

#### Without P-5-P

Intra-assay, n = 20	Mean [U/L]	SD [U/L]	CV [%]
Sample 1	22.2	1.38	6.22
Sample 2	44.8	1.17	2.62
Sample 3	101	1.02	1.00

Inter-assay, n = 20	Mean [U/L]	SD [U/L]	CV [%]
Sample 1	22.8	0.70	3.08
Sample 2	42.6	0.68	1.60
Sample 3	99.3	0.92	0.92

#### With P-5-P

Intra-assay, n = 20	Mean [U/L]	SD [U/L]	CV [%]
Sample 1	33.8	1.25	3.71
Sample 2	72.0	2.04	2.83
Sample 3	128	2.77	2.16

Inter-assay, n = 20	Mean [U/L]	SD [U/L]	CV [%]
Sample 1	33.3	0.99	2.96
Sample 2	72.1	1.36	1.88
Sample 3	133	1.76	1.32

## SPECIFICITY/INTERFERENCES

no interference up to:

Ascorbic acid	30 mg/dL
Bilirubin	40 mg/dL
Hemoglobin	400 mg/dL
Triglycerides	2000 mg/dL

For further information on interfering substances refer to Young DS [6].

## METHOD COMPARISON

### Without P-5-P

A comparison between Dialab GPT (ALT) without P-5-P (y) and a commercially available test (x) using 105 samples gave following results:  $y = 1.024x - 1.199$  U/L;  $r = 0.999$ .

### With P-5-P

A comparison between Dialab GPT (ALT) with P-5-P (y) and a commercially available test (x) using 107 samples gave following results:  $y = 1.027x - 0.189$  U/L;  $r = 1.000$ .

## TRACEABILITY

This method has been standardized against the original IFCC formulation.

## EXPECTED VALUES\*

### With P-5-P:

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

### Without P-5-P:

Women [8, 9]	< 31 U/L	< 0.52 µkat/L
Men [8, 9]	< 41 U/L	< 0.68 µkat/L

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

## LIMITATIONS

- Eventual GPT (ALT), mod. IFCC carry-over to reagents Carbon Dioxide (PEP-C) and Protein Total in Urine/CSF (Pyrogallol red). The actual carry-over depends on the analyser.

## WASTE MANAGEMENT

Please refer to local legal requirements.

## LITERATURE

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