BAB V

PENUTUP

5.1 Kesimpulan

Dari data pemeriksaan kadar SGOT dan SGPT pada petani yang terpapar pestisida dapat disimpulkan bahwa :

1. Dari 28 sampel, 2 sampel (7,14%) mengalami peningkatan kadar SGOT
2. Dari 28 sampel, 1 sampel (3,57%) mengalami peningkatan kadar SGPT
3. Dari 28 sampel, 5 sampel (17,86%) mengalami peningkatan kadar SGOT dan SGPT
4. Dari 28 sampel, 20 sampel (71,43%) tidak mengalami peningkatan kadar SGOT dan SGPT.

5.2 Saran

1. Bagi petani yang bekerja dianjurkan untuk menggunakan dosis pestisida sesuai dengan batas dosis yang telah ditetapkan
2. Bagi Petani yang bekerja dianjurkan untuk menggunakan APD yang lengkap untuk dapat mengurangi resiko masuknya pestisida ke dalam tubuh.

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L A M P I R A N

Lampiran 1. Hasil Penelitian Kadar SGOT dan SGPT pada Petani

NO	NAMA	UMUR	JK	SGOT	KET	SGPT	KET
1	WIR	73	L	46 U/L	> Normal	36 U/L	Normal
2	SUR	60	L	23 U/L	Normal	29 U/L	Normal
3	SUM	63	L	39 U/L	> Normal	30 U/L	Normal
4	REJ	32	L	19 U/L	Normal	15 U/L	Normal
5	SUG	51	L	47 U/L	> Normal	57 U/L	> Normal
6	PI	60	L	48 U/L	>Normal	63 U/L	> Normal
7	ISB	36	L	20 U/L	Normal	26U/L	Normal
8	PAR	60	L	29 U/L	Normal	23 U/L	Normal
9	NIQ	57	L	18 U/L	Normal	17 U/L	Normal
10	MUL	53	L	34 U/L	Normal	26 U/L	Normal
11	PON	53	L	23 U/L	Normal	17 U/L	Normal
12	FAT	47	L	19 U/L	Normal	15 U/L	Normal
13	TRI	50	L	41 U/L	> Normal	53 U/L	> Normal
14	SWN	34	P	18 U/L	Normal	11 U/L	Normal
15	NAR	60	P	22 U/L	Normal	29 U/L	Normal
16	PAN	58	P	20 U/L	Normal	13 U/L	Normal
17	KAS	56	P	32 U/L	Normal	45 U/L	> Normal
18	SUR	38	P	30 U/L	Normal	17 U/L	Normal
19	PUJ	28	P	21 U/L	Normal	16 U/L	Normal
20	WAG	45	P	16 U/L	Normal	19 U/L	Normal
21	SAD	65	P	36 U/L	> Normal	46 U/L	> Normal
22	ROH	62	P	29 U/L	Normal	28 U/L	Normal
23	KAT	50	P	20 U/L	Normal	13 U/L	Normal
24	SAR	52	P	41 U/L	> Normal	51 U/L	> Normal
25	RUM	49	P	25 U/L	Normal	20 U/L	Normal
26	RAT	37	P	22 U/L	Normal	15 U/L	Normal
27	MAR	54	P	19 U/L	Normal	18 U/L	Normal
28	MAS	57	P	20 U/L	Normal	16 U/L	Normal

Harga Normal SGOT : Wanita < 31 µ/L

Laki-laki < 35 µ/L

Harga Normal SGPT : Wanita < 31 µ/L

Laki-laki < 41 µ/L

Data Induk

NO	NAMA	UMUR	JK	TINGKAT PENDIDIKAN	SGOT (µ/L)	SGPT (µ/L)	MASA KERJA (TAHUN)	LAMA TIAP PENYEMPROTAN (JAM)	MENGKONSUMSI ALKOHOL
1	WIR	73	L	SMP	46	36	> 2	≤ 5 jam	TIDAK
2	SAD	65	P	SMA	36	46	> 2	≤ 5 jam	TIDAK
3	SUM	63	L	SD	39	30	> 2	> 5 jam	TIDAK
4	ROH	62	P	SD	29	28	> 2	≤ 5 jam	TIDAK
5	PI	60	L	SD	46	63	> 2	> 5 jam	TIDAK
6	PAR	60	L	SMP	29	23	> 2	≤ 5 jam	TIDAK
7	SUR	60	L	SD	23	29	> 2	> 5 jam	TIDAK
8	NAR	60	P	SD	22	29	> 2	> 5 jam	TIDAK
9	PAN	58	P	SMP	20	13	> 2	≤ 5 jam	TIDAK
0	NIQ	57	L	SD	18	17	> 2	> 5 jam	TIDAK
11	MAS	57	P	SMP	20	16	> 2	≤ 5 jam	TIDAK
12	KAS	56	P	SMP	32	45	> 2	≤ 5 jam	TIDAK
13	MAR	54	P	Perguruan Tinggi	19	18	> 2	≤ 5 jam	TIDAK
14	PON	53	L	SD	23	17	> 2	> 5 jam	TIDAK
15	MUL	53	L	SMP	34	26	> 2	≤ 5 jam	TIDAK
16	SAR	52	P	SMP	41	51	> 2	≤ 5 jam	TIDAK
17	SUG	51	L	SD	47	57	> 2	> 5 jam	TIDAK
18	TRI	50	L	SMP	41	53	> 2	> 5 jam	TIDAK
19	KAT	50	P	SMA	36	46	> 2	≤ 5 jam	TIDAK
20	RUM	49	P	SMA	25	20	> 2	≤ 5 jam	TIDAK

21	FAT	47	L	SMA	19	15	> 2	≤ 5 jam	TIDAK
22	WAG	45	P	SMA	16	19	≤ 2	≤ 5 jam	TIDAK
23	SUR	38	P	Perguruan Tinggi	30	16	≤ 2	≤ 5 jam	TIDAK
24	RAT	37	P	Perguruan Tinggi	22	15	≤ 2	≤ 5 jam	TIDAK
25	ISB	36	L	SMA	20	26	≤ 2	> 5 jam	TIDAK
26	SWN	34	P	Perguruan Tinggi	18	11	≤ 2	≤ 5 jam	TIDAK
27	REJ	32	L	SMP	19	15	≤ 2	> 5 jam	TIDAK
28	PUJ	28	P	Perguruan Tinggi	21	16	≤ 2	≤ 5 jam	TIDAK

Lampiran 2. Kuisioner

LEMBAR OBSERVASI PENELITIAN

Identitas Responden

Nama :

Umur :

Jenis Kelamin : Laki-laki / Perempuan

Alamat :

Tingkat Pendidikan Terakhir : Tidak sekolah/ SD/ SMP /SMA /Perguruan tinggi

Pekerjaan :

Hasil Pemeriksaan Lab : a. SGOT μ/L

b. SGPT μ/L

Lingkarilah jawaban sesuai dengan pilihan anda!

1. Berapa lamakah anda bekerja sebagai petani (.....Tahun)
   a. Kurang dari 2 tahun
   b. Lebih dari 2 tahun

2. Apakah anda menggunakan pestisida untuk tanaman ?
   a. Ya b. Tidak

3. Berapa lama anda setiap menyemprot pestisida ?
   a. ≤ 5 jam
   b. > 5 jam

4. Apakah anda menggunakan pakaian pelindung waktu anda kontak dengan pestisida ?
   a. Ya b. Tidak

5. Jika Ya, apa saja ? **(Lingkari, jawaban bisa lebih dari satu)**
   a. Sarung tangan e. Baju lengan panjang
   b. Sepatu boot f. Masker

6. Setelah menyemprot, pernahkah anda mengalami :
   (beri tanda silang (X) pada tiap jawaban)

No.	Gejala	Ya	Tidak
1	Mual dan muntah		
2	Pusing		
3	Sakit kepala		
4	Susah bernafas		
5	Dada sesak		
6	Kejang		
7	Gemetar/ tremor		
8	Kram		
9	Sakit otot		
10	nyeri perut sebelah kanan saat berjalan cepat atau lari		
11	Diare		
12	Keringat berlebihan		
13	Pandangan kabur		
14	Lemas		
15	Warna kuku berubah		

   Lainnya.....

7. Apakah anda mengkonsumsi obat-obatan ?
   a. Ya b. Tidak

8. Apakah anda mengkonsumsi alkohol ?
   a. Ya b. Tidak

Lampiran 3 Informent Consent

SURAT PERSETUJUAN TINDAKAN

INFORMED CONSENT

Saya yang bertanda tangan dibawah ini :

Nama :

Umur :

Jenis kelamin :

Pekerjaan :

Alamat :

Telpon :

Dengan ini menyatakan SETUJU untuk dilakukan tindakan pengambilan darah vena dalam penelitian dengan judul “Pemeriksaan Kadar SGOT dan SGPT pada Petani yang Terpapar Pestisida” yang dilakukan oleh saudari Vini Meidy Syafitri Mahasiswa Fakultas Ilmu Kesehatan Universitas Setia Budi.

Dari penjelasan yang diberikan, saya telah mengerti segala resiko yang dapat timbul akibat tindakan tersebut diatas.

Klaten, Januari 2019

Peneliti

Yang membuat pernyataan

(Vini Meidy Syafitri)

( )

Lampiran 4. Surat Keterangan Ijin Penelitian

UNIVERSITAS SETIA BUDI
FAKULTAS ILMU KESEHATAN

Nomor : 441 / H6 – 04 / 11.01.2019
Lamp. : - helai
Hal : *Ijin Penelitian*

Kepada :
Yth. Kepala
UPT. Laboratorium
Universitas Setia Budi
Di Surakarta

Dengan Hormat,

Guna memenuhi persyaratan untuk keperluan penyusunan Karya Tulis Ilmiah (KTI) bagi Mahasiswa Semester Akhir Program Studi D-III Analis Kesehatan Fakultas Ilmu Kesehatan Universitas Setia Budi, terkait bidang yang ditekuni dalam melaksanakan kegiatan tersebut bersamaan dengan ini kami menyampaikan ijin bahwa :

NAMA : VINI MEIDY SYAFITRI
NIM : 34162936 J
PROGDI : D-III Analis Kesehatan
JUDUL : Pemeriksaan Kadar SGOT dan SGPT pada yang terpapar Pestisida

Untuk ijin penelitian di laboratorium Universitas Setia Budi tentang Pemeriksaan Kadar SGOT dan SGPT pada yang terpapar Pestisida di Instansi Bapak / Ibu

Demikian atas bantuan dan kerjasamanya kami ucapkan terima kasih.

Surakarta, 11 Januari 2019
Dekan,

UNIVERSITAS SETIA BUDI
FAKULTAS ILMU KESEHATAN

Prof. dr. Marsetyawan HNE Soesatyo, M.Sc., Ph.D.

Jl. Let. Jend. Sutoyo Mojosongo – Solo 57127, Telp. 0271 – 852518, Fax. 0271 – 853275
Homepage : www.setiabudi.ac.id, e-mail : usbsolo@yahoo.com

Lampiran 5. Surat keterangan ijin Pengambilan Sampel

UNIVERSITAS SETIA BUDI

FAKULTAS ILMU KESEHATAN

Nomor : 441 / H6 – 04 / 11.01.2019
Lamp. : - helai
Hal : *Ijin Penelitian*

Kepada :
Yth. Kepala
Desa Carikan Kec. Juwiring
Kabupaten Klaten
Jawa Tengah

Dengan Hormat,

Guna memenuhi persyaratan untuk keperluan penyusunan Karya Tulis Ilmiah (KTI) bagi Mahasiswa Semester Akhir Program Studi D-III Analis Kesehatan Fakultas Ilmu Kesehatan Universitas Setia Budi, terkait bidang yang ditekuni dalam melaksanakan kegiatan tersebut bersamaan dengan ini kami menyampaikan ijin bahwa :

NAMA : VINI MEIDY SYAFITRI
NIM : 34162936 J
PROGDI : D-III Analis Kesehatan
JUDUL : Pemeriksaan Kadar SGOT dan SGPT pada yang terpapar Pestisida

Untuk ijin pengambilan sampel tentang Pemeriksaan Kadar SGOT dan SGPT pada yang terpapar Pestisida di daerah Desa Carikan Kec. Juwiring Kabupaten Klaten Jawa Tengah

Demikian atas bantuan dan kerjasamanya kami ucapkan terima kasih.

Surakarta, 11 Januari 2019
Dekan

UNIVERSITAS SETIA BUDI FAKULTAS ILMU KESEHATAN

Prof. dr. Marsetyawan HNE Soesatyo, M.Sc., Ph.D.

Jl. Let. Jend. Sutoyo Mojosongo – Solo 57127, Telp. 0271 – 852518, Fax. 0271 – 853275
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Lampiran 6. Leaflet Reagen

ASAT (GOT) FS* (IFCC mod.)

with/without pyridoxal-5-phosphate

Diagnostic reagent for quantitative in vitro determination of ASAT(GOT) in serum or plasma on photometric systems

Order Information

Cat. No.	Kit size						
1 2601 99 10 021	R1	5 x	20 mL	+	R2	1 x	25 mL
1 2601 99 10 026	R1	5 x	80 mL	+	R2	1 x	100 mL
1 2601 99 10 023	R1	1 x	800 mL	+	R2	1 x	200 mL
1 2601 99 10 704	R1	8 x	50 mL	+	R2	8 x	12.5 mL
1 2601 99 10 917	R1	8 x	60 mL	+	R2	8 x	15 mL
1 2601 99 90 314	R1	10 x	20 mL	+	R2	2 x	30 mL

For determination with pyridoxal-5-phosphate activation additionally required:

2 5010 99 10 030	6 x	3 mL

Summary [1,2]

Alanine Aminotransferase (ALAT/ALT), formerly 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 catalyze the conversion of α-keto acids into amino acids by transfer of amino groups.
As a liver specific enzyme ALAT is only significantly elevated in hepatobiliary diseases. Increased ASAT levels, however, can occur in connection with damages of heart or skeletal muscle as well as of liver parenchyma. Parallel measurement of ALAT and ASAT is, therefore, applied to distinguish liver from heart or skeletal muscle damages. The ASAT/ALAT 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.

Method

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

Principle

L-Aspartate + 2-Oxoglutarate $\xleftarrow{ASAT}$ L-Glutamate + Oxalacetate

Oxalacetate + NADH + H^+ $\xleftarrow{MDH}$ L-Malate + 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 7.65	110 mmol/L
	L-Aspartate		320 mmol/L
	MDH (malate dehydrogenase)		≥ 800 U/L
	LDH (lactate dehydrogenase)		≥ 1200 U/L
R2:	2-Oxoglutarate		65 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. 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. Reagent 1 contains biological 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 [6].
4. 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.
5. For professional use only!

Waste Management

Please refer to local legal requirements.

Reagent Preparation

Substrate Start

The reagents are ready to use.
For the determination with pyridoxal-5-phosphate mix 1 part of P-5-P with 100 parts of reagent 1,
e.g. 100 µL P-5-P + 10 mL R1

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

Sample Start

without pyridoxal-5-phosphate
Mix 4 parts of R1 + 1 part of R2
(e.g. 20 mL R1 + 5 mL R2) = mono reagent

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

The mono reagent must be protected from light!

Materials required but not provided

DiaSys Pyridoxal-5-Phosphate FS in case of determination with P-5-P activation (Cat.-No. 2 5010 99 10 030)
NaCl solution 9 g/L
General laboratory equipment

Specimen

Serum, heparin plasma or EDTA plasma

Stability [3]:

4 days	at	20 – 25°C
7 days	at	4 – 8°C
3 months	at	–20°C

Discard contaminated specimens. Only freeze once!

Assay Procedure

Application sheets for automated systems are available on request.

Wavelength	340 nm, Hg 365 nm, Hg 334 nm
Optical path	1 cm
Temperature	37°C
Measurement	Against air

Substrate Start

Sample/Calibrator	100 µL
Reagent 1	1000 µL
Mix, incubate for 5 min., then add:	
Reagent 2	250 µL
Mix, read absorbance after 1 min. and start stopwatch. Read absorbance again 1, 2 and 3 min thereafter.	

Sample Start

Don't use sample start with pyridoxal-5-phosphate !

Sample/Calibrator	100 µL
Mono reagent	1000 µL
Mix, read absorbance after 1 min. and start stopwatch. Read absorbance again 1, 2 and 3 min thereafter.	

Calculation

With factor

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

ΔA/min x factor = ASAT activity [U/L]

Substrate Start	
340 nm	2143
334 nm	2184
365 nm	3971
Sample Start	
340 nm	1745
334 nm	1780
365 nm	3235

With calibrator

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

* fluid stable

Conversion factor

ASAT [U/L] x 0.0167 = ASAT [µkat/L]

Calibrators and Controls

For the calibration of automated photometric systems, DiaSys TruCal U calibrator is recommended. This method has been standardized against the original IFCC formulation. 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

Measuring range

On automated systems the test is suitable for the determination of ASAT activities up to 700 U/L.
In case of a manual procedure, the test is suitable for ASAT activities which correspond to a maximum of ΔA/min of 0.16 at 340 and 334 nm or 0.08 at 365 nm.
If such values are exceeded the samples should be diluted 1 + 9 with NaCl solution (9 g/L) and results multiplied by 10.

Specificity/Interferences

No interference was observed by ascorbic acid up to 30 mg/dL, bilirubin up to 40 mg/dL and lipemia up to 2000 mg/dL triglycerides. The presence of hemoglobin in serum indicates destruction of erythrocytes with release of ASAT, thus producing high interference. For further information on interfering substances refer to Young DS [5].

Sensitivity/Limit of Detection

The lower limit of detection is 2 U/L.

Precision

Without P-5-P

Intra-assay precision n = 20	Mean [U/L]	SD [U/L]	CV [%]
Sample 1	25.1	0.82	3.25
Sample 2	51.3	1.57	3.06
Sample 3	116	0.90	0.77

Inter-assay precision n = 20	Mean [U/L]	SD [U/L]	CV [%]
Sample 1	25.7	1.13	4.40
Sample 2	48.6	0.67	1.38
Sample 3	115	0.80	0.69

With P-5-P

Intra-assay precision n = 20	Mean [U/L]	SD [U/L]	CV [%]
Sample 1	43.6	1.10	2.51
Sample 2	74.5	1.79	2.41
Sample 3	174	3.18	1.83

Inter-assay precision n = 20	Mean [U/L]	SD [U/L]	CV [%]
Sample 1	44.0	1.59	3.61
Sample 2	77.0	3.05	3.97
Sample 3	187	3.37	1.80

Method Comparison

With P-5-P

A comparison of DiaSys ASAT (GOT) FS with P-5-P (y) with the IFCC reference reagent (x) using 51 samples gave following results:
y = 1.000 x – 0.800 U/L; r = 0.999

A comparison of DiaSys ASAT (GOT) FS (y) with P-5-P and a commercially available test (x) using 51 samples gave following results:
y = 0.970 x + 0.350 U/L; r = 0.999

Without P-5-P

A comparison of DiaSys ASAT (GOT) FS without P-5-P (y) and a commercially available test (x) using 51 samples gave following results:
y = 0.997 x + 0.621 U/L; r = 1.000

Reference Range

With pyridoxal-5-phosphate activation

Women [4]		< 31 U/L	< 0.52 µkat/L
Men [4]		< 35 U/L	< 0.58 µkat/L
Children [1]	1 – 3 Years	< 50 U/L	< 0.83 µkat/L
	4 – 6 Years	< 45 U/L	< 0.75 µkat/L
	7 – 9 Years	< 40 U/L	< 0.67 µkat/L
	10 – 12 Years	< 40 U/L	< 0.67 µkat/L
	13 – 15 Years	< 35 U/L	< 0.58 µkat/L
	16 – 18 Years	< 35 U/L	< 0.58 µkat/L

Without pyridoxal-5-phosphate activation

Women	< 31 U/L	< 0.52 µkat/L
Men	< 35 U/L	< 0.58 µkat/L

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

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DiaSys Diagnostic Systems GmbH
Alte Strasse 9 65558 Holzheim Germany

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 photometric systems

Order Information

Cat. No.	Kit size					
1 2701 99 10 021	R1	5 x	20 mL	+ R2	1 x	25 mL
1 2701 99 10 026	R1	5 x	80 mL	+ R2	1 x	100 mL
1 2701 99 10 023	R1	1 x	800 mL	+ R2	1 x	200 mL
1 2701 99 10 704	R1	8 x	50 mL	+ R2	8 x	12.5 mL
1 2701 99 10 917	R1	8 x	60 mL	+ R2	8 x	15 mL
1 2701 99 90 314	R1	10 x	20 mL	+ R2	2 x	30 mL

For determination with pyridoxal-5-phosphate activation additionally required:

2 5010 99 10 030	6 x	3 mL

Summary [1,2]

Alanine Aminotransferase (ALAT/ALT), formerly 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 catalyze the conversion of α-keto acids into amino acids by transfer of amino groups.

As a liver specific enzyme, ALAT is only significantly elevated in hepatobiliary diseases. Increased ASAT levels, however, can occur in connection with damages of heart or skeletal muscle as well as of liver parenchyma. Parallel measurement of ALAT and ASAT is, therefore, applied to distinguish liver from heart or skeletal muscle damages. The ASAT/ALAT 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.

Method

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

Principle

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

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

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. 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

Substrate Start

The reagents are ready to use.

For the determination with pyridoxal-5-phosphate (P-5-P) mix 1 part of P-5-P with 100 parts of reagent 1,
e.g. 100 µL P-5-P + 10 mL R1

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

Sample Start

without pyridoxal-5-phosphate

Mix 4 parts of R1 + 1 part of R2
(e.g. 20 mL R1 + 5 mL R2) = mono-reagent

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

The mono-reagent must be protected from light!

Materials required but not provided

DiaSys Pyridoxal-5-Phosphate FS in case of determination with P-5-P activation (Cat. No. 2 5010 99 10 030)
NaCl solution 9 g/L; General laboratory equipment

Specimen

Serum, heparin plasma or EDTA plasma
Stability [4]:

3 days at 20 - 25 °C
7 days at 4 - 8 °C
7 days at -20 °C

Only freeze once! Discard contaminated specimens!

Assay Procedure

Application sheets for automated systems are available on request.

Wavelength	340 nm, Hg 365 nm, Hg 334 nm
Optical path	1 cm
Temperature	37 °C
Measurement	Against air

Substrate Start

Sample or calibrator	100 µL
Reagent 1	1000 µL
Mix, incubate for 5 min., then add:	
Reagent 2	250 µL
Mix, read absorbance after 1 min. and start stopwatch.	
Read absorbance again 1, 2 and 3 min thereafter.	

Sample Start

Do not use sample start with pyridoxal-5-phosphate!

Sample or calibrator	100 µL
Mono-reagent	1000 µL
Mix, read absorbance after 1 min. and start stopwatch.	
Read absorbance again 1, 2 and 3 min thereafter.	

Calculation

With factor

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

ΔA/min x factor = ALAT activity [U/L]

	Substrate Start	Sample Start
340 nm	2143	1745
334 nm	2184	1780
365 nm	3971	3235

With calibrator

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

* fluid stable

Conversion factor

ALAT [U/L] x 0.0167 = ALAT [µkat/L]

Calibrators and Controls

For the calibration of automated photometric systems the 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

Measuring range

On automated systems the test is suitable for the determination of ALAT activities up to 600 U/L.
In case of a manual procedure, the test is suitable for ALAT activities which correspond to a maximum of ΔA/min of 0.16 at 340 and 334 nm or 0.08 at 365 nm. If such values are exceeded the samples should be diluted 1 + 9 with NaCl solution (9 g/L) and results multiplied by 10.

Specificity/Interferences

No interference was observed by ascorbic acid up to 30 mg/dL, bilirubin up to 40 mg/dL, hemoglobin up to 400 mg/dL and lipemia up to 2,000 mg/dL triglycerides. For further information on interfering substances refer to Young DS [5].

Sensitivity/Limit of Detection

The lower limit of detection is 4 U/L.

Precision

Without P-5-P

Intra-assay precision 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 precision 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 precision 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 precision 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

Method Comparison

With P-5-P

A comparison of DiaSys ALAT (GPT) FS with P-5-P (y) and the IFCC reference reagent (x) using 51 samples gave following results:
y = 1.000 x – 0.200 U/L; r = 0.999.

A comparison of DiaSys ALAT (GPT) FS with P-5-P (y) and a commercially available test (x) using 51 samples gave following results:
y = 0,970 x + 0.531 U/L; r = 1.000.

Without P-5-P

A comparison of DiaSys ALAT (GPT) FS without P-5-P (y) with a commercially available test (x) using 51 samples gave following results:
y =0.971 x + 0.047 U/L; r =1.000.

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 the 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. 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.
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. Guder WG, Zawta B et al. The Quality of Diagnostic Samples. 1st ed. Darmstadt: GIT Verlag; 2001; 14-5.
5. 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.

Manufacturer

IVD CE DiaSys Diagnostic Systems GmbH
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Lampiran 7. Quality Control Alat

PELAKSANAAN *QUALITY CONTROL RAYTO*

Pelaksanaan dilakukan pada :

Hari/ tanggal : Sabtu, 29 Juni 2019

Tempat : Laboratorium 02 Universitas Setia Budi Surakarta

Hasil :

PERCOBAAN	HASIL
1	22
2	19
3	18
4	18
5	19
6	19
7	20
8	20
9	19
10	20

Diketahui :

Mean : 19,4

Standard Deviasi : 1,173788

+ 1 SD : 20,57379

+ 2 SD : 21,74758

+ 3 SD : 22,92136

- 1 SD : 18,22621

- 2 SD : 17,05242

- 3 SD : 15,87864

KOEFISIEN VARIASI/ KV

KV = (SD / Mean) x 100 %

= (1,173788 / 19,4) x 100 %

= 6,1 %

KESIMPULAN

Dari sepuluh kali percobaan yang diuji ketelitiannya, hasil analisis terhadap sampel yang sama dengan menggunakan alat *Rayto* diperoleh hasil koefisien variasi (KV) sebesar 6,1% dengan standard deviasi 1,173788

Surakarta, Juni 2019

Menyetujui,

Pendamping	Peneliti
Jatmiko. Amd	Vini Meidy Syafitri

Lampiran 8. Alat dan Bahan Penelitian

Spuit 3ml

Tourniquet

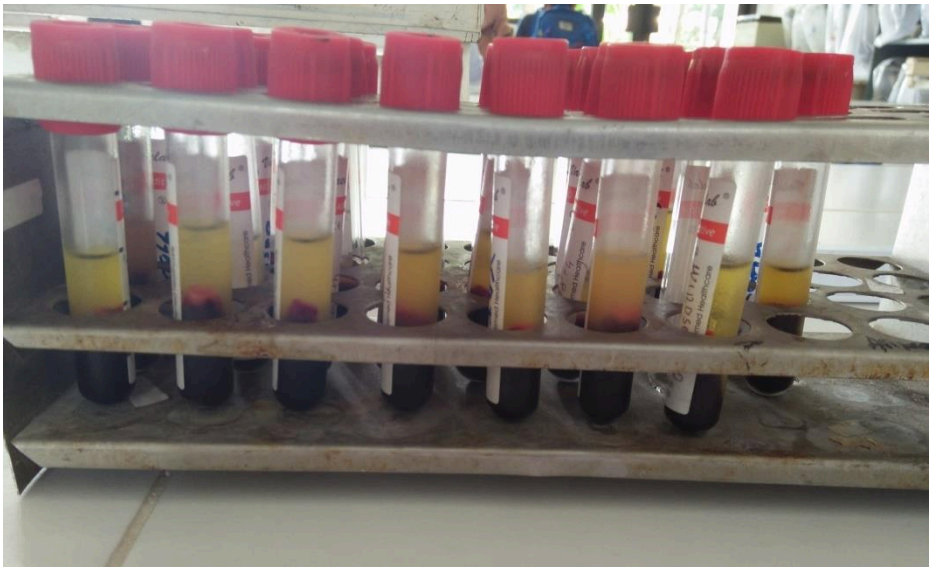
Vacutainer Tube

Alkohol Swab

Kapas Kering

Clinipet 1000 µl dan 100 µl

Yellow tip dan Blue Tip



Sampel Serum

Fotometer Star Dust FC

Centrifuge