BAB V

KESIMPULAN DAN SARAN

A. Kesimpulan

Berdasarkan dari hasil penelitian dan pembahasan dapat ditarik kesimpulan sebagai berikut,

1. Kombinasi zat pengatur tumbuh 2,4-D dan Kinetin dalam media New Phalaenopsis (NP) mampu menginduksi kalus daun Stevia
2. Konsentrasi penambahan zat pengatur tumbuh Kinetin 1 ppm mampu mempengaruhi pembentukan kalus daun Stevia lebih banyak dari konsentrasi lainnya.
3. Kadar steviosida dalam kalus daun Stevia lebih tinggi bila dibandingkan dengan kadar dalam daun Stevia hasil adaptasi di Solo.

B. Saran

Pertama, perlu diadakan penelitian lebih lanjut untuk mengetahui pengaruh zat pengatur tumbuh 2,4-D dan kinetin terhadap pertumbuhan kalus dan kadar steviosida dalam kalus daun Stevia.

Kedua, perlu dikaji secara lebih mendalam untuk mengetahui kandungan kimia dari kalus daun Stevia selain steviosida yang nantinya dapat berguna dan bermanfaat terutama bagi dunia kesehatan.

Ketiga, perlu diadakan penelitian lebih lanjut dengan membandingkan kadar kalus hasil adaptasi dengan kadar kalus asli dari Tawangmangu.

DAFTAR PUSTAKA

Depkes. 2000. *Inventaris Tanaman Obat Indonesia*. Jilid 1. Jakarta: Departemen Kesehatan dan Kesejahteraan Sosial RI. Badan Litbangkes

Bawane, Adesh A. 2005. *Standaritation Of Botanical : Stevia reboudiana Leaves And AEvaluation For Antioxidant Activity.* Departemen Of Pharmacognosy K. L. E. Society's College of Pharmacy. Karnataka. India.

Brandle, J.E. & Telmer, P.G. 2007. *Steviol Glycoside Biosynthesis*, Southern Crop Protection and Food Research Centre, London, *Phytochemistry*, 68; 1855-1863.

Gandjar, IG & Rohman, A. 2007. *Kmia Farmasi Analisis.* Yogyakarta: Pustaka Pelajar. Hlm 353-361

Gardana *et al.* (2003). *Metabolism of Stevioside and Rebaudioside A from Stevia rebaudiana Extracts by Human Microflora*. Jurnal of Agricurtural Food Chemistry. 2003, 51, 6618-6622

Geuns J.M.C. 2003. *Molecules of Interest - Stevioside*. Phytochem 64, 913-921.

Hendaryono, D.P.S. & Wijayani, A., 1994, *Teknik Kultur Jaringan Tanaman*. Yogyakarta: Kanisius Press, 115-125

Kinghorn, A.D. 2002. *Stevia, The genus Stevia*. Departemen of Medicinal Chemistry and Pharmacognosy University of Illinois at Chicago. USA

Liudianto, E.R. 2003. I*dentifikasi Senyawa Steviosida Dalam Kalus Daun Stevia rebaudiana Bertonii M. Dengan Kombinasi Hormon 2,4-D dan BAP Pada Media Murashige-Skoog* [Skripsi]. Surakarta: Fakultas Farmasi, Universitas Setia Budi.

Mahadi *et al*. (2013): *Pengaruh Pemberian Naa Dan Kinetin Terhadap Pertumbuhan Eksplan Buah Naga (Hylocereus Costaricensis) Melalui Teknik Kultur Jaringan Secara In VITRO.* Jurnal Biogenesis, Vol. 9, Nomor 2, Februari 2013

Mawarni, Lisa (2011): *Produksi Tanaman Stevia (Stevia Rebaudiana* Bertonii M.*) dengan Perlakuan Setek dan Auksin.* Departemen Budidaya Pertanian. STEVIA. Vol. 1. No 01. ISSN No. 2087-6939. Januari 2011

Nugroho A & Sugito H. 2004. *Pedoman Pelaksanaan Teknik Kultur Jaringan.* Depok: Penebar Swadaya. Hlm 1-52

Nurwardani P. 2008. Teknik *Pembibitan Tanaman Dan Produksi Benih*. Jilid 2. Jakarta : Direktorat Pembinaan Sekolah Menengah Kejuruan

Raini M & Isnawati A. 2011. *Kajian: Khasiat dan Keamanan Stevia Sebagai Pemanis Pengganti Gula.* Media Litbang Kesehatan. Volume 21. No. 4. Hlm 146-149

Ratnani RD. & Anggraeni R. 2005. *Ekstraksi Gula Stevia Dari Tanaman Stevia Rebaudiana Bertonii.* Momentum. Vol 1, No. 2. Hlm 27-32

Ritonga A.W, 2007. *Pembuatan Media Kultur Jaringan Tanaman.* Laporan Pratikum Dasar-Dasar Bioteknologi Tanaman. Bogor: Institut Pertanian

Rodiansyah A. 2007. *Induksi Mutasi Kromosom Dengan Kolkisin Pada Tanaman Stevia (Stevia Rebaudiana Bertoni) Klon Zweeteners Secara In Vitro* [Skripsi]. Bogor: Fakultas Pertanian, Institut Pertanian Bogor

Santoso, U. & Nursandi, F. 2002. *Kultur Jaringan Tanaman*. Edisi Pertama, Malang: UMM Press. 131-136

Sumaryono & Sinta M. (2011). *Peningkatan Laju Multiplikasi Tunas Dan Keragaan Planlet Stevia Rebaudiana Pada Kultur In Vitro.* Menara Perkebunan 2011 79(2), 49-56

Wahidah, Sri. (2011). *Pengaruh Hormon Kinetin Terhadap Pertumbuhan Kalus Rumput Laut Kappaphycus alvarezii Melalui Kultur In Vitro*. Jurnal Vokasi. Vol. 7. No. 2. 192-197. Juli 2011.

Wiryosoendjoyo *et al*. (2011): *Pengaruh Penambahan Zat Pengatur Tumbuh 2,4-D Dan Kinetin Pada Media New Phalaenopsis (Np) Terhadap Kandungan Steviosida Dalam Kalus Daun Stevia Rebaudiana Bertonii M.* Biomedika. Jurnal Ilmiah Biologi dan Kesehatan. Vol. 4. No. 2. ISSN no. 1979-035 X. September 2011.

Zulkarnain. 2009. *Kultur Jaringan Tanaman Solusi Perbanyakan Tanaman Budi Daya.* Jakarta: Penerbit Bumi Aksara.

Lampiran 1. Surat keterangan determinasi tanaman Stevia.

UNIVERSITAS SETIA BUDI

UPT- LABORATORIUM

No : 163/DET/UPT-LAB/09/VI/2014
Hal : Surat Keterangan Determinasi Tumbuhan

Menerangkan bahwa :
Nama : Rini Elviyah
NIM : 15113357 A
Fakultas : Farmasi Universitas Setia Budi

Telah mendeterminasikan tumbuhan : **Stevia (*Stevia rebaudiana* Bertonii M.)**

Hasil determinasi berdasarkan : **Baker: Flora of Java**

1b – 2b – 3b – 4b – 12b – 13b – 14b – 17b – 18b – 19b – 20b – 21b – 22b – 23b – 24b – 25b – 26b – 27b – 799a. Familia 166. Asteraceae. 1b – 3a – 4b – 5b – 23b – 28a – 29b. 11. *Stevia sp.*

Deskripsi *Stevia rebaudiana* Bertonii M.

Habitus : Semak, semusim, tinggi dapat mencapai 90 cm.
Batang : Bulat, hijau, beruas, berbulu.
Daun : Tunggal, berhadapan, bulat telur, berbulu, ujung tumpul, pangkal runcing, tepi bergerigi, tulang daun menyirip, tangkai pendek, hijau.
Bunga : Majemuk malai, di ujung dan di ketiak daun,
Buah : Kotak, berambut, coklat.
Biji : Bentuk jarum.
Akar : Tunggang.

Pustaka : Backer C.A. & Brink R.C.B. (1965): *Flora of Java* (Spermatophytes only). N.V.P. Noordhoff – Groningen – The Netherlands.

Surakarta, 09 Juni 2014
Tim determinasi

Dra.Kartinah Wiryosoendjojo, SU.

Jl. Let.jen Sutoyo, Mojosongo-Solo 57127 Telp.0271-852518, Fax.0271-853275
Homepage : www.setiabudi.ac.id, e-mail : info@setiabudi.ac.id

Lampiran 2. Certificate of Analysis steviosida standard.

NINGBO HAISHU J S TRADING CO.,LTD.

ADD: 529 YUANBAOSHAN ROAD, BEILUN DISTRICT, NINGBO, CHINA **E-mail:** jasonji@vip.163.com
TEL: 0086-574-87897188/27851288 **FAX:**0086-574-87897189 **URL:** http://www.jsbotanics.com

Certificate of Analysis

Product Name:	**Stevioside**	**Manufacture Date:**	**2009-05-01**
Latin Name:	***Stevia Rebaudiana Hemsl***	**Testing Date:**	**2009-05-05**
Batch Number:	**SST20090501**	**Expire Date:**	**2011-04-30**
Quantity:	**500KGS**	**Shelf Life:**	**24 MONTHS**

ITEM	SPECIFICATION	TEST RESULT
PHYSICAL TESTS:		
DESCRIPTION:		
APPEARANCE	WHITE FINE POWDER	COMPLIES
AROMA	CHARACTERISTIC	COMPLIES
TASTE	CHARACTERISTIC	COMPLIES
PARTICLE SIZE	80 MESH	COMPLIES
BULK DENSITY	0.35-0.55G/ML	0.35G/ML
CHEMICAL TESTS:		
ASSAY	≥95.00%	95.48%
SPECIFIC ROTATION	-30° ~-38°	-37°
SPECIFIC ABSOROANCE	≤0.050	0.038
LOSS ON DRYING	≤4.0%	3.5%
ASH	≤0.2%	0.11%
HEAVY METAL	≤10PPM	<10PPM
AS	≤1PPM	<1PPM
MICROBIOLOGICAL RESULTS		
TOTAL AEROBIC PLATE COUNT	≤1000CFU/G	<10 CFU/G
YEAST & MOLD	≤100 CFU/G	10 CFU/G
E.COLI	NEGATIVE	NEGATIVE
SALMONELLA	NEGATIVE	NEGATIVE
STAPHYLOCOCCUS AUREUS	NEGATIVE	NEGATIVE
STORAGE	STORE IN COOL & DRY PLACE. KEEP AWAY FROM STRONG LIGHT AND HEAT.	

QUALITY ASSURANCE OFFICER S.L.AN

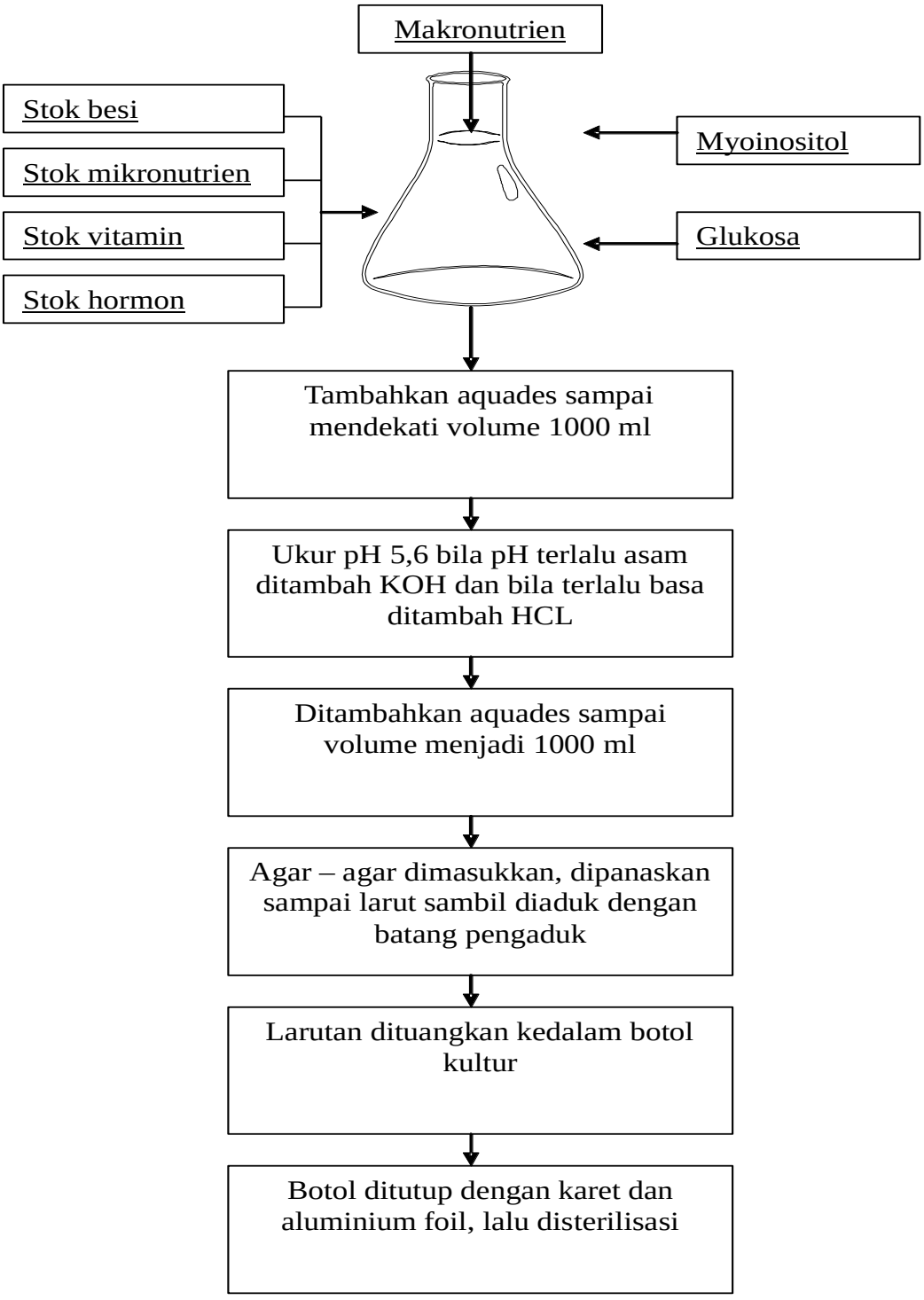
CORRECTOR LI YI

ANALYST YUJIAO WA

Lampiran 3. Komposisi media New Phalaenopsis (NP).

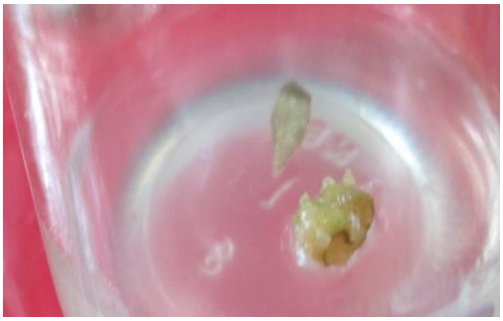
Bahan	Jumlah (mg/L)
I. Makronutrien	
NH_4NO_3	32
$(NH_4)_2SO_4$	303,9
KNO_3	424,6
$Mg(NO_3)_2.6H_2O$	256,4
$Ca(NO_3)_2.4H_2O$	637,6
KH_2PO_4	462,7
II. Besi	
Na2EDTA	37,3
FeSO4 7H2O	27,8
III. Mikronutrien	
$MnSO_4$ H_2O	11,15
$ZnSO_4$ $4H_2O$	4,3
H_3BO_3	3,1
KI	0,415
$NaMoO_4$ $2H_2O$	0,125
$CuSO_4$ $5H_2O$	0,0125
$CoCl_2$ $6H_2O$	0,0125
IV. Vitamin	
Glycine	2
Nicotinic acid	0,5
Pyridoxine-HCl	0,5
Thiamine-HCl	0,1
Myoinositol	100
Glukosa	20.000
Agar	7000
pH	5,6

Lampiran 4. Skema pembuatan media New Phalaenopsis (NP) 1 liter.



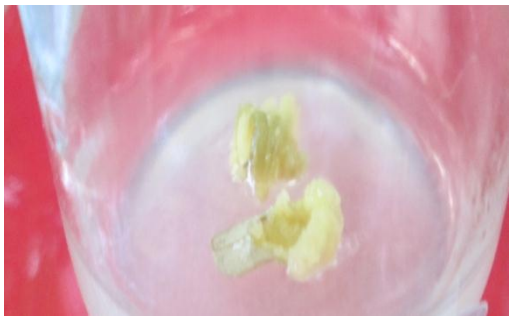
Lampiran 5. Foto kalus *Stevia rebaudiana* Bertonii M.

ZPT kinetin 1 ppm
Eksplan setelah diinkubasi selama 1 minggu



ZPT kinetin 1 ppm
Eksplan setelah diinkubasi selama 2 minggu

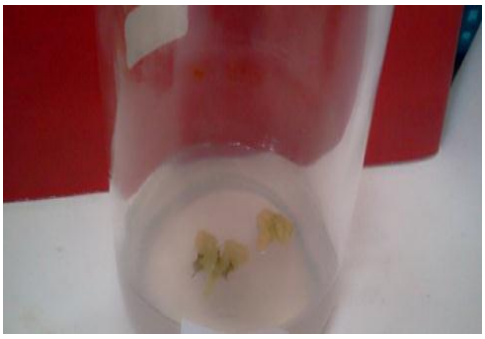
ZPT kinetin 1 ppm
Eksplan setelah diinkubasi selama 3 minggu



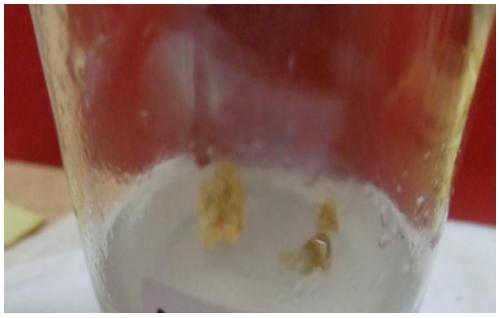
ZPT kinetin 1 ppm
Eksplan setelah diinkubasi selama 4 minggu

Subkultur kalus ZPT kinetin 1 ppm

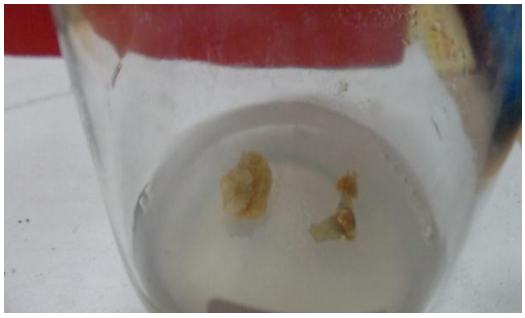
ZPT 2,4-D 0,25 ppm dan kinetin 0,75 ppm
Eksplan setelah diinkubasi selama 1 minggu



ZPT 2,4-D 0,25 ppm dan kinetin 0,25 ppm
Eksplan setelah diinkubasi selama 2 minggu

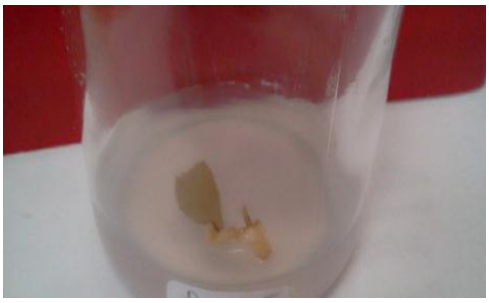


ZPT 2,4-D 0,25 ppm dan kinetin 0,75 ppm
Eksplan setelah diinkubasi selama 3 minggu

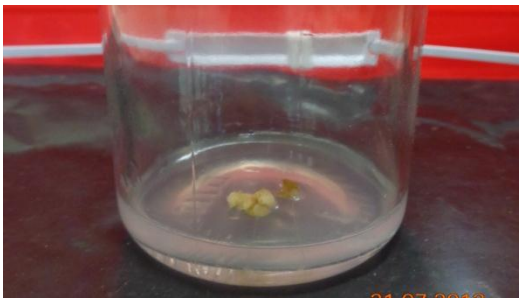


ZPT 2,4-D 0,25 ppm dan kinetin 0,75 ppm
Eksplan setelah diinkubasi selama 4 minggu

Subkultur kalus ZPT 2,4-D 0,25 ppm dan kinetin 0,75 ppm

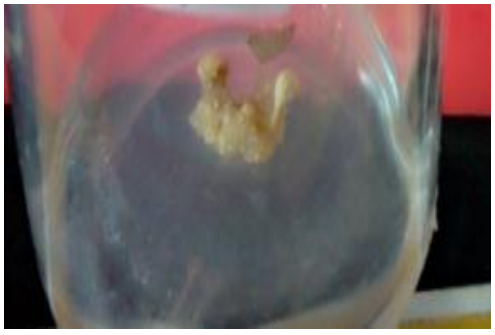


ZPT 2,4-D 0,5 ppm dan kinetin 0,5 ppm
Eksplan setelah diinkubasi selama 1 minggu



ZPT 2,4-D 0,5 ppm dan kinetin 0,5 ppm
Eksplan setelah diinkubasi selama 2 minggu

ZPT 2,4-D 0,5 ppm dan kinetin 0,5 ppm
Eksplan setelah diinkubasi selama 3 minggu



ZPT 2,4-D 0,5 ppm dan kinetin 0,5 ppm
Eksplan setelah diinkubasi selama 4 minggu

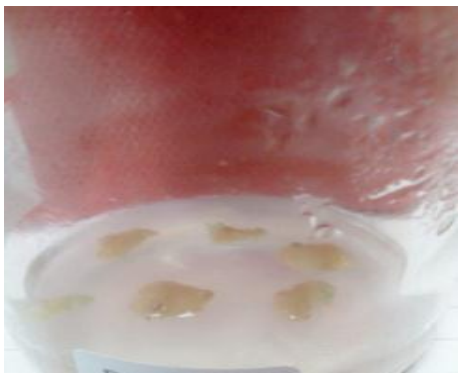
Subkultur kalus ZPT 2,4-D 0,5 ppm dan kinetin 0,5 ppm

ZPT 2,4-D 0,75 ppm dan kinetin 0,25 ppm
Eksplan setelah diinkubasi selama 1 minggu

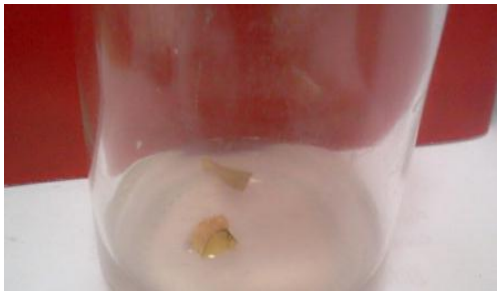
ZPT 2,4-D 0,75 ppm dan kinetin 0,25 ppm
Eksplan setelah diinkubasi selama 2 minggu

ZPT 2,4-D 0,75 ppm dan kinetin 0,25 ppm
Eksplan setelah diinkubasi selama 3 minggu

ZPT 2,4-D 0,75 ppm dan kinetin 0,25 ppm
Eksplan setelah diinkubasi selama 4 minggu

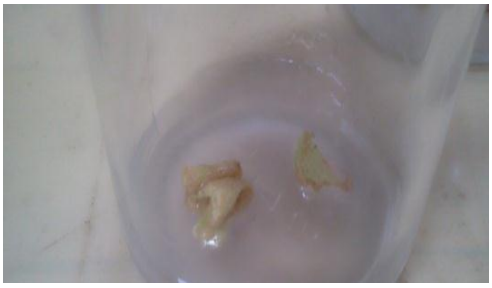


Subkultur kalus ZPT 2,4-D 0,75 ppm dan kinetin 0,25 ppm



ZPT 2,4-D 1 ppm
Eksplan setelah diinkubasi selama 1 minggu

ZPT 2,4-D 1 ppm
Eksplan setelah diinkubasi selama 2 minggu



ZPT 2,4-D 1 ppm
Eksplan setelah diinkubasi selama 3 minggu

ZPT 2,4-D 1 ppm
Eksplan setelah diinkubasi selama 4 minggu

Subkultur kalus ZPT 2,4-D 1 ppm

Lampiran 6. Foto ekstrak daun dan kalus daun Stevia

Ekstrak daun Stevia asal Solo

Lampiran 7. Sampel daun Stevia hasil adaptasi di Solo

Lampiran 8. Kromatogram steviosida standard dan daun Stevia sebanyak 2 replikasi

Replikasi 1

winCATS Planar Chromatography Manager

Detection - CAMAG TLC Scanner 3

Information

Application position	8,0 mm
Solvent front position	75,0 mm

Instrument

Executed by	CAMAG TLC Scanner 3 "Scanner3_160602" S/N 160602 (1.14.28)
Number of tracks	farmasiusd 25 Oktober 2013 14:09:04
Position of first track X	7
Distance between tracks	15,0 mm
Scan start pos. Y	10,0 mm
Scan end pos. Y	20,0 mm
Slit dimensions	154,0 mm
Optimize optical system	6,00 x 0,10 mm, Micro
Scanning speed:	Light
Data resolution:	20 mm/s
	100 µm/step

daun 10 µl

Measurement Table

Wavelength	400
Lamp	D2 & W
Measurement Type	Remission
Measurement Mode	Absorption
Optical filter	Second order
Detector mode	Automatic
PM high voltage	462 V

Detector properties

Y-position for 0 adjust	20,0 mm
Track # for 0 adjust	0
Analog Offset	10%
Sensitivity	Automatic (36)

Integration

Properties

Data filtering	Savitsky-Golay 7
Baseline correction	Lowest Slope
Peak threshold min. slope	15
Peak threshold min. height	40 AU
Peak threshold min. area	200
Peak threshold max. height	990 AU
Track start position	20,0 mm
Track end position	154,0 mm
Display scaling	Automatic

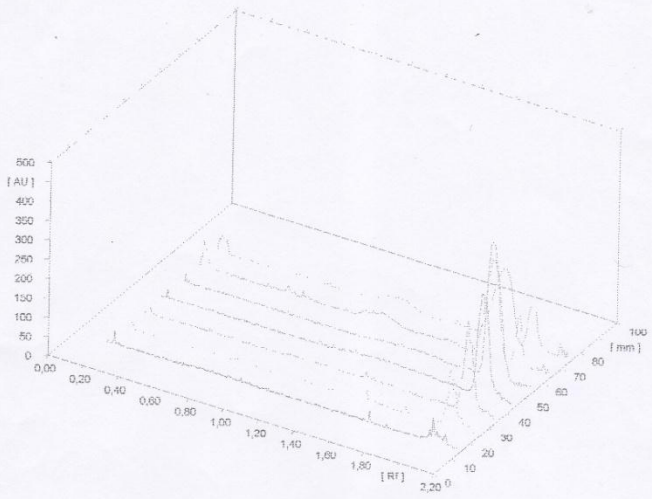
User : farmasiusd
25 Oktober 2013 14:09:19

Approved :. . . . .
Report ID : 07DD0A19060E090C

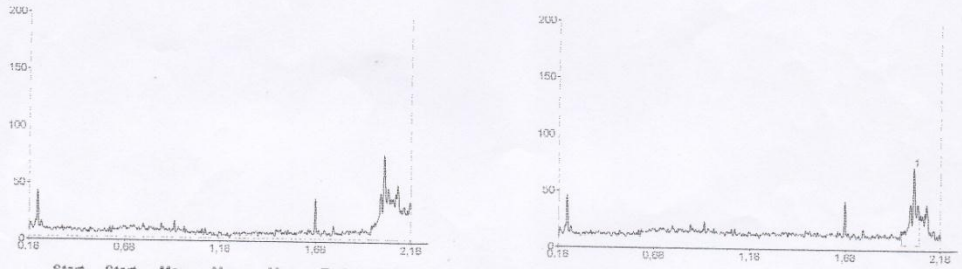
SN 1607W007, V1.4.4
Page 1 of 4

winCATS Planar Chromatography Manager

All tracks at Wavelength



Track 1, ID:



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,98	4,3	2,04	63,0	100,00	2,07	18,1	1328,2	100,00	unknown *

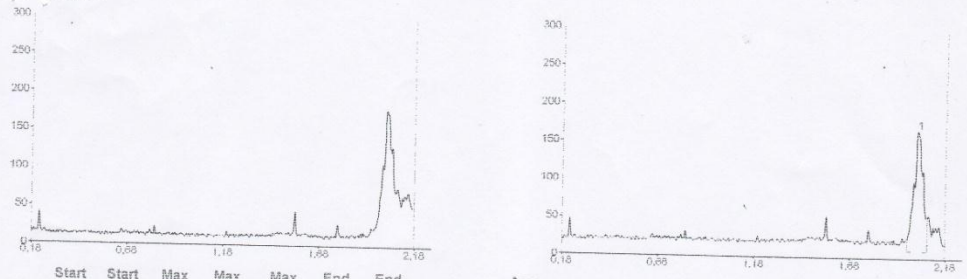
User : farmasiusd
25 Oktober 2013 14:09:19

Approved :. . . . . .
Report ID : 07DD0A19060E090C

SN 1607W007, V1.4.4
Page 2 of 4

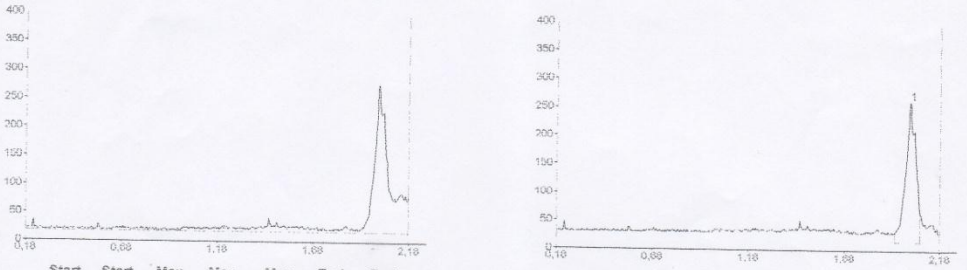
winCATS Planar Chromatography Manager

Track 2, ID:



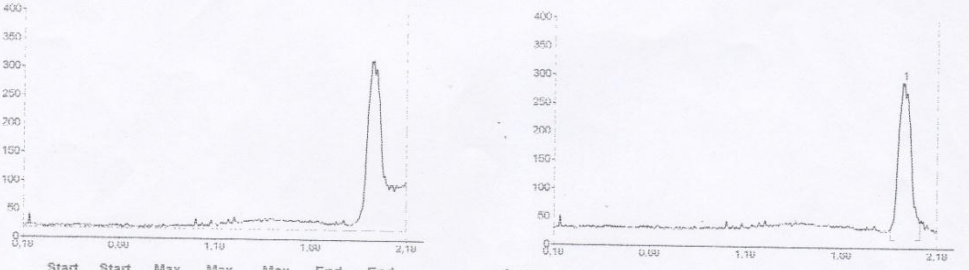
Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,98	6,2	2,03	150,7	100,00	2,09	34,0	5324,8	100,00	unknown *

Track 3, ID:



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,94	1,5	2,02	235,2	100,00	2,08	36,6	9147,5	100,00	unknown *

Track 4, ID:



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,93	2,8	2,00	263,7	100,00	2,09	14,6	12955,1	100,00	unknown *

User : farmasiusd
25 Oktober 2013 14:09:19

Approved : .....
Report ID : 07DD0A19060E090C

SN 1607W007, V1.4.4
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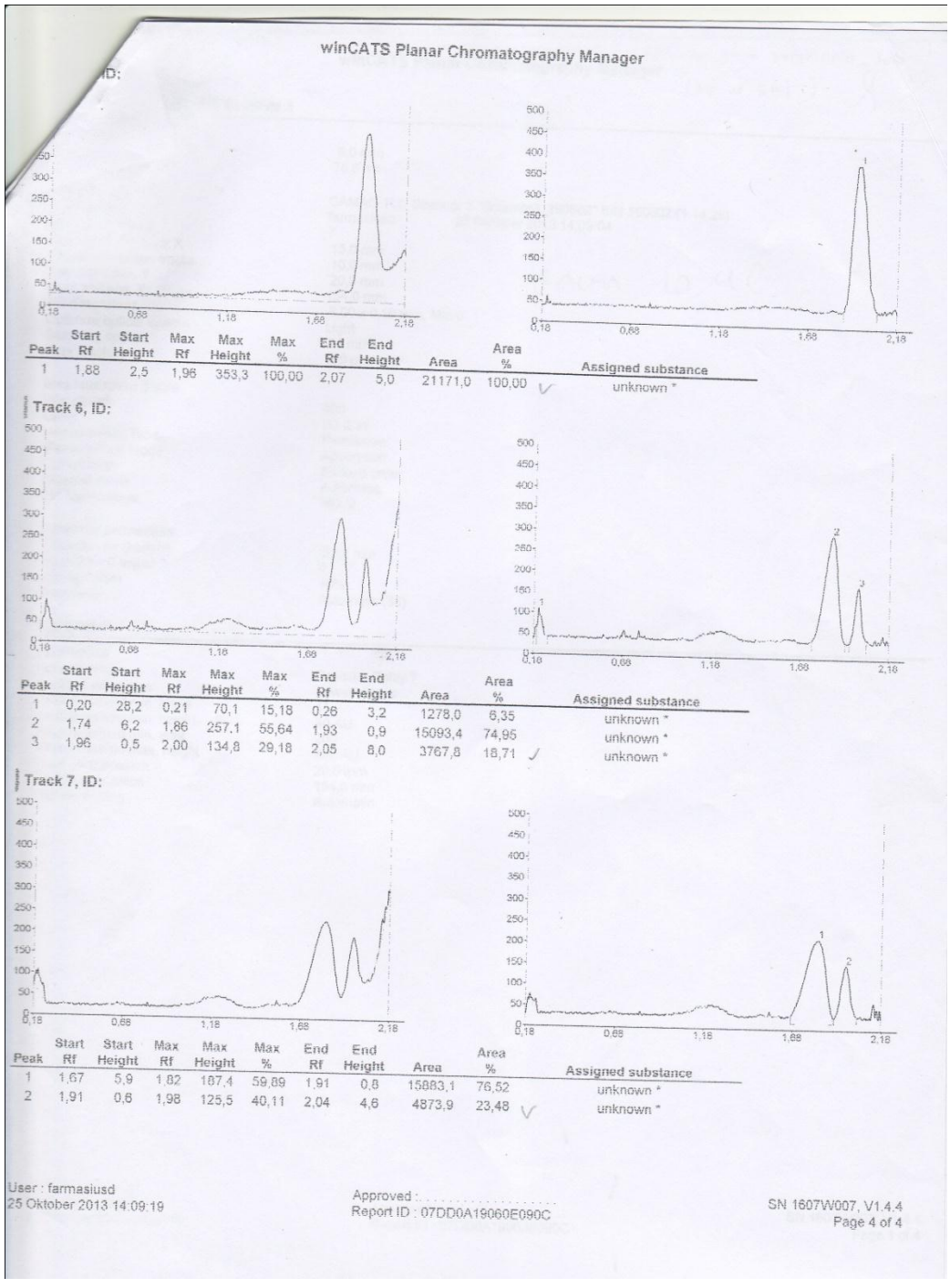

winCATS Planar Chromatography Manager

ID:

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,88	2,5	1,96	353,3	100,00	2,07	5,0	21171,0	100,00	unknown *

Track 6, ID:

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0,20	28,2	0,21	70,1	15,18	0,26	3,2	1278,0	6,35	unknown *
2	1,74	6,2	1,86	257,1	55,64	1,93	0,9	15093,4	74,95	unknown *
3	1,96	0,5	2,00	134,8	29,18	2,05	8,0	3767,8	18,71	unknown *

Track 7, ID:

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,67	5,9	1,82	187,4	59,89	1,91	0,8	15883,1	76,52	unknown *
2	1,91	0,6	1,98	125,5	40,11	2,04	4,6	4873,9	23,48	unknown *

User : farmasiusd
25 Oktober 2013 14:09:19

Approved : .....
Report ID : 07DD0A19060E090C

SN 1607W007, V1.4.4
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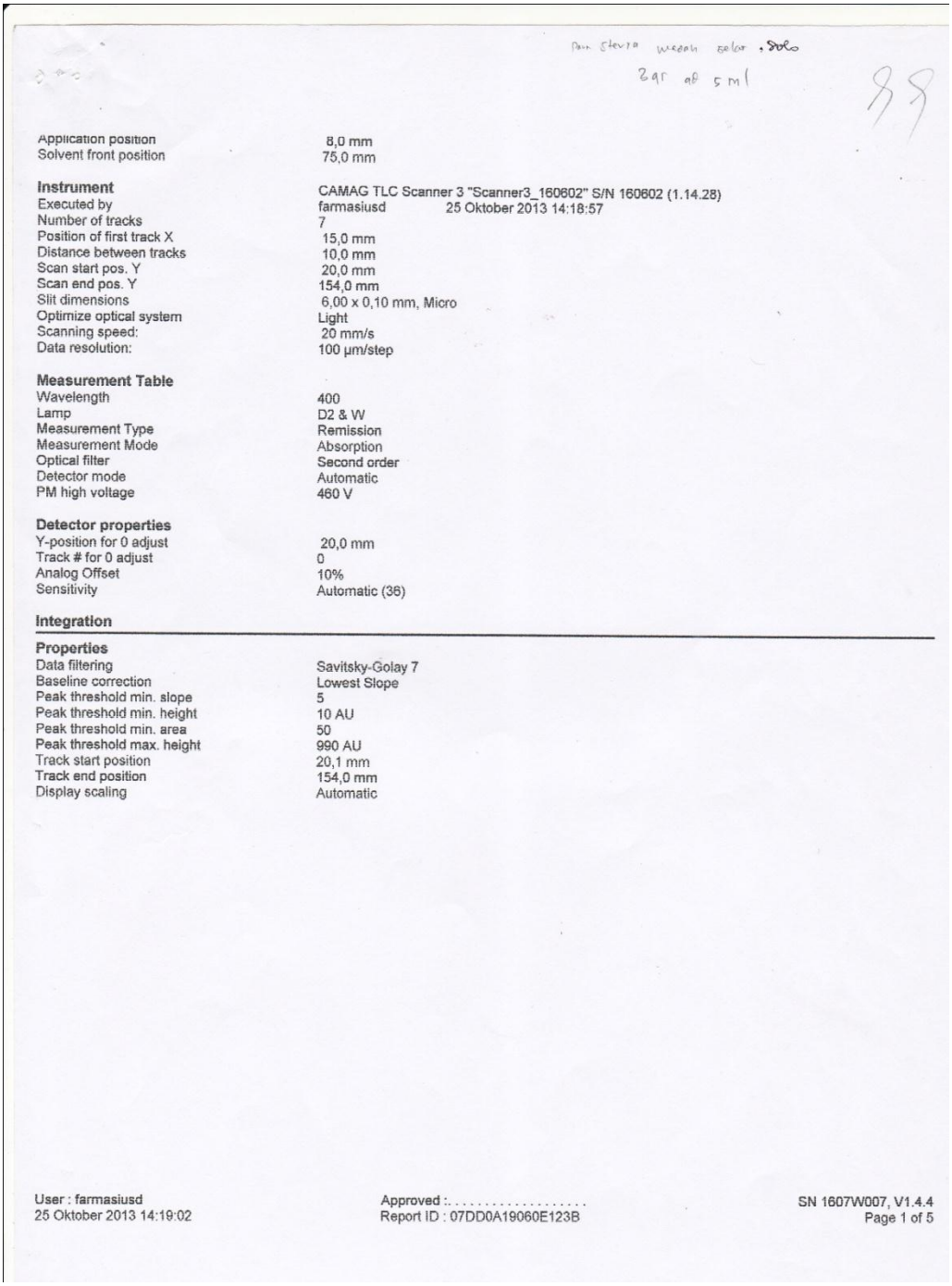
Replikasi 2



Pair Stevia wiedeh eelat .500
2gr ad 5ml

99

Application position	8,0 mm
Solvent front position	75,0 mm
Instrument	
Executed by	CAMAG TLC Scanner 3 "Scanner3_160602" S/N 160602 (1.14.28)
	farmasiusd 25 Oktober 2013 14:18:57
Number of tracks	7
Position of first track X	15,0 mm
Distance between tracks	10,0 mm
Scan start pos. Y	20,0 mm
Scan end pos. Y	154,0 mm
Slit dimensions	6,00 x 0,10 mm, Micro
Optimize optical system	Light
Scanning speed:	20 mm/s
Data resolution:	100 µm/step
Measurement Table	
Wavelength	400
Lamp	D2 & W
Measurement Type	Remission
Measurement Mode	Absorption
Optical filter	Second order
Detector mode	Automatic
PM high voltage	460 V
Detector properties	
Y-position for 0 adjust	20,0 mm
Track # for 0 adjust	0
Analog Offset	10%
Sensitivity	Automatic (36)
Integration	
Properties	
Data filtering	Savitsky-Golay 7
Baseline correction	Lowest Slope
Peak threshold min. slope	5
Peak threshold min. height	10 AU
Peak threshold min. area	50
Peak threshold max. height	990 AU
Track start position	20,1 mm
Track end position	154,0 mm
Display scaling	Automatic

User : farmasiusd
25 Oktober 2013 14:19:02

Approved :. . . . . .
Report ID : 07DD0A19060E123B

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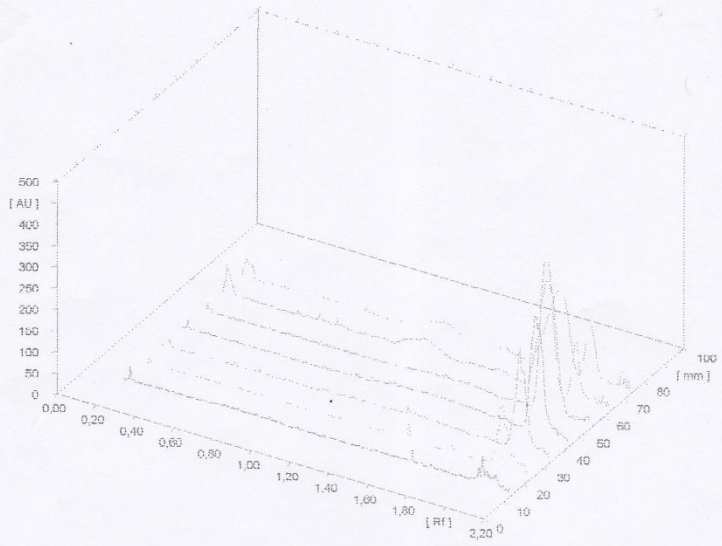
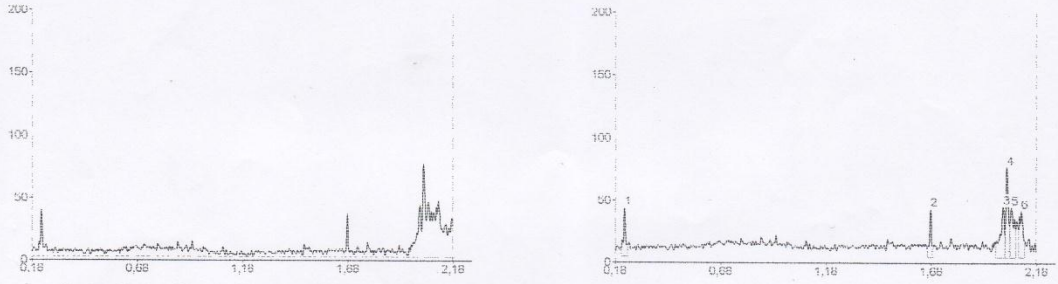
winCATS Planar Chromatography Manager

All tracks at Wavelength



Track 1, ID: baku 1



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %		Assigned substance
1	0,21	0,9	0,22	33,8	14,22	0,24	3,5	223,9	9,17		unknown *
2	1,66	2,4	1,68	33,8	14,22	1,69	4,2	192,6	7,89		unknown *
3	1,99	5,4	2,02	35,6	15,00	2,03	14,3	484,8	19,85	V	unknown *
4	2,03	18,0	2,04	67,1	28,24	2,05	21,1	599,4	24,55	V	unknown *
5	2,05	22,7	2,06	34,9	14,69	2,08	18,1	494,0	20,23	✓	unknown *
6	2,09	17,0	2,11	32,4	13,63	2,12	7,7	447,1	18,31		unknown *

1578,2

User : farmasiusd
25 Oktober 2013 14:19:02

Approved :. . . . . .
Report ID : 07DD0A19060E123B

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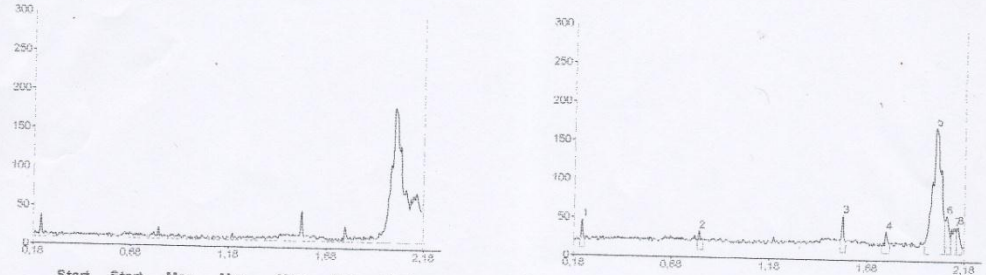
winCATS Planar Chromatography Manager

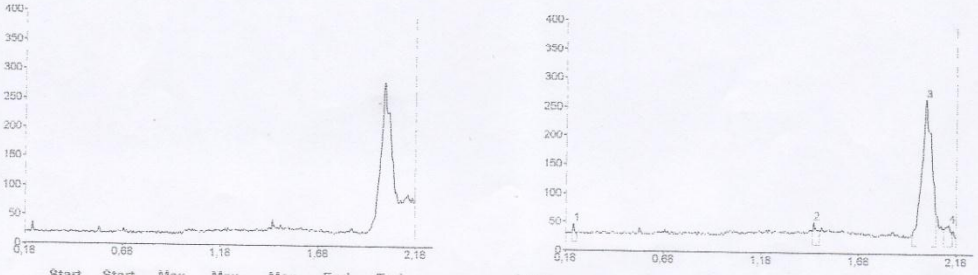
winCATS Planar Chromatography Manager

Track 2, ID: baku 2



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %		Assigned substance
1	0,21	1,9	0,22	26,6	7,70	0,23	1,2	159,4	2,13		unknown *
2	0,81	3,5	0,82	14,6	4,23	0,84	1,9	121,8	1,63		unknown *
3	1,54	6,9	1,56	38,6	11,15	1,57	4,9	325,3	4,35		unknown *
4	1,76	1,2	1,78	18,8	5,43	1,80	2,4	174,9	2,34		unknown *
5	1,97	5,0	2,03	155,0	44,83	2,08	35,7	5400,8	72,18	✓	unknown *
6	2,08	35,8	2,09	40,3	11,66	2,11	12,1	620,8	8,30		unknown *
7	2,11	14,4	2,13	25,1	7,27	2,14	19,3	400,4	5,35		unknown *
8	2,14	20,9	2,14	26,6	7,74	2,17	0,4	279,4	3,73		unknown *

Track 3, ID: baku 4



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %		Assigned substance
1	0,21	1,0	0,22	16,6	5,47	0,23	2,3	93,4	0,92		unknown *
2	1,44	8,7	1,45	24,0	7,93	1,48	9,9	336,5	3,32		unknown *
3	1,95	3,2	2,02	240,4	79,32	2,08	39,2	9267,7	91,43	✓	unknown *
4	2,12	13,8	2,14	22,1	7,28	2,16	5,1	438,9	4,33		unknown *

User : farmasiusd
25 Oktober 2013 14:19:02

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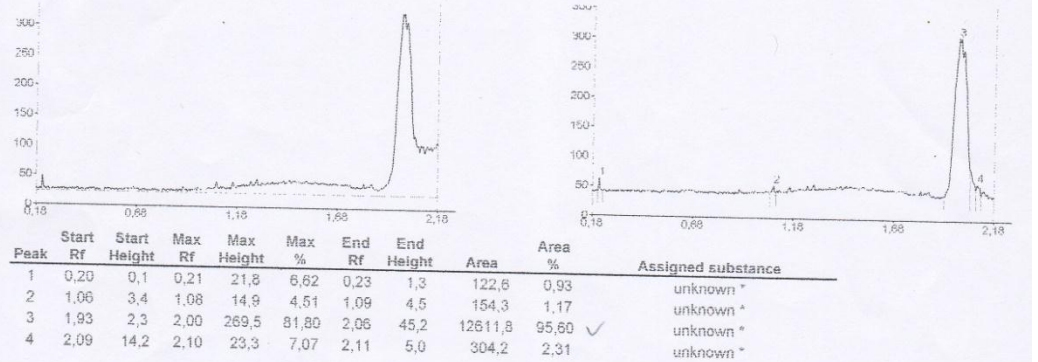
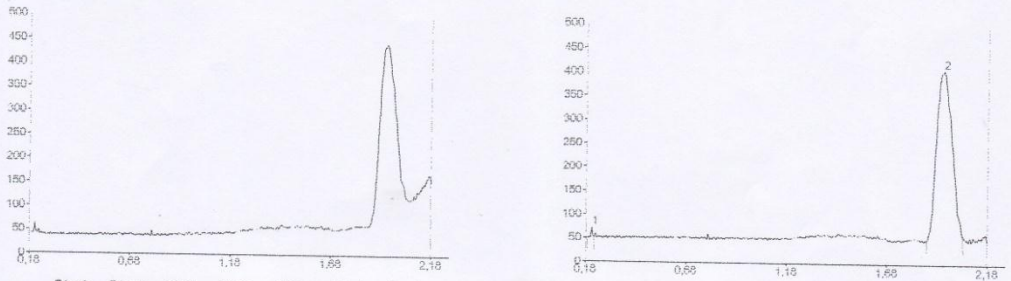


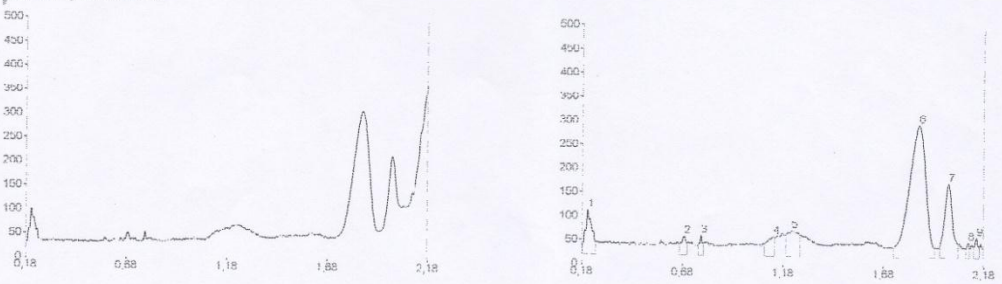
Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0,20	0,1	0,21	21,8	6,62	0,23	1,3	122,6	0,93	unknown *
2	1,06	3,4	1,08	14,9	4,51	1,09	4,5	154,3	1,17	unknown *
3	1,93	2,3	2,00	269,5	81,80	2,06	45,2	12611,8	95,60	unknown *
4	2,09	14,2	2,10	23,3	7,07	2,11	5,0	304,2	2,31	unknown *

Track 5, ID: baku 16



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0,19	0,1	0,21	21,8	5,67	0,22	2,4	124,5	0,58	unknown *
2	1,88	2,2	1,96	359,5	94,33	2,06	12,2	21404,4	99,42	unknown *

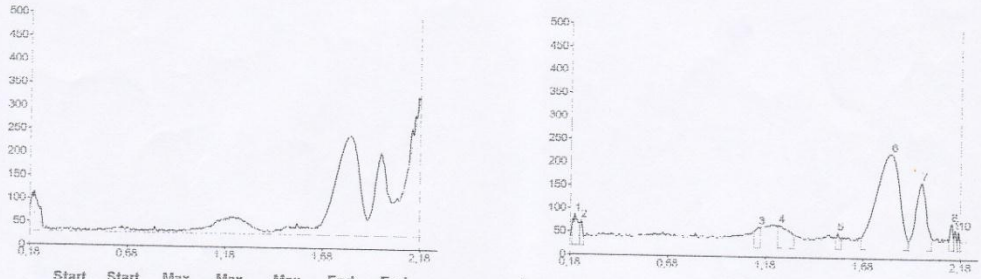
Track 6, ID: daun 1



winCATS Planar Chromatography Manager

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0,18	0,6	0,21	71,5	12,12	0,25	5,5	1451,9	6,38	unknown *
2	0,66	1,5	0,69	19,6	3,32	0,70	3,9	262,3	1,15	unknown *
3	0,76	0,6	0,77	21,4	3,63	0,79	5,0	142,6	0,63	unknown *
4	1,09	6,1	1,13	20,6	3,49	1,14	18,0	485,6	2,13	unknown *
5	1,20	23,5	1,22	33,3	5,64	1,27	24,2	1453,3	6,38	unknown *
6	1,73	2,3	1,86	256,5	43,48	1,93	0,4	14999,6	65,88	unknown *
7	1,96	5,1	2,00	134,6	22,81	2,05	8,6	3717,5	16,33 ✓	unknown *
8	2,09	0,1	2,10	11,1	1,88	2,11	0,3	72,8	0,32	unknown *
9	2,13	1,5	2,14	21,3	3,62	2,16	0,0	184,3	0,81	unknown *

Track 7, ID: daun 2



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0,19	22,7	0,20	47,2	8,56	0,23	28,3	906,8	3,71	unknown *
2	0,23	27,8	0,24	34,0	6,18	0,25	0,9	316,3	1,29	unknown *
3	1,12	14,3	1,15	25,0	4,54	1,15	22,6	464,4	1,90	unknown *
4	1,24	28,7	1,25	30,5	5,53	1,33	8,4	1211,3	4,96	unknown *
5	1,54	5,5	1,55	15,4	2,79	1,57	3,2	152,3	0,62	unknown *
6	1,67	5,6	1,82	187,2	33,97	1,91	0,1	15961,9	65,34	unknown *
7	1,91	0,0	1,98	126,7	22,99	2,03	7,1	4760,1	19,49 ✓	unknown *
8	2,12	0,5	2,13	39,4	7,15	2,14	7,0	405,3	1,66	unknown *
9	2,15	9,1	2,15	24,1	4,37	2,16	0,3	156,3	0,64	unknown *
10	2,16	1,3	2,17	21,5	3,91	2,18	0,7	93,3	0,38	unknown *

User : farmasiusd
25 Oktober 2013 14:19:02

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Report ID : 07DD0A19060E123B

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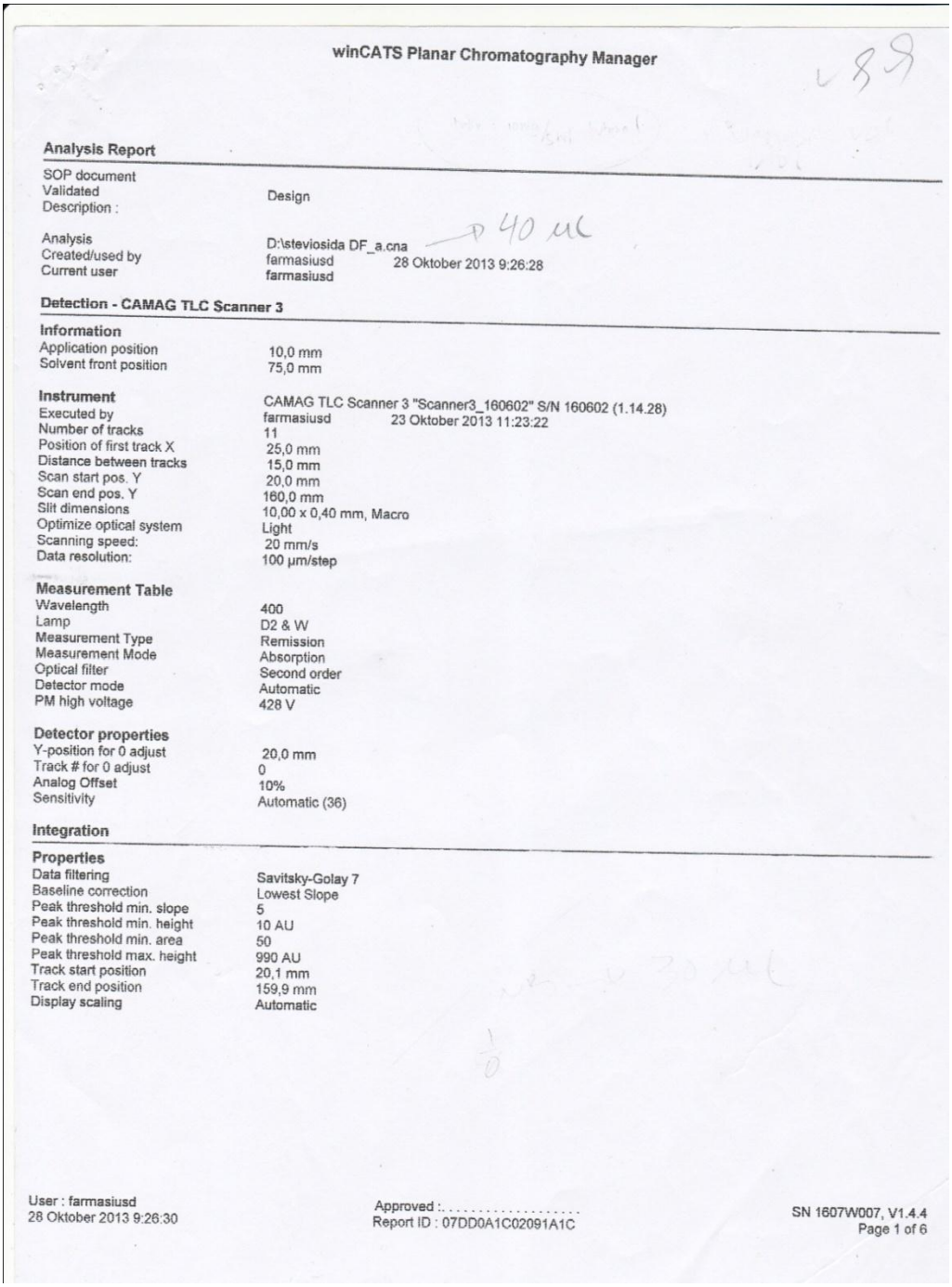
winCATS Planar Chromatography Manager

Lampiran 9. Kromatogramstandar dan kalus daun Stevia senyak 2 replikasi

Replikasi 1



winCATS Planar Chromatography Manager

Analysis Report

SOP document	
Validated	
Description :	Design
Analysis	D:\steviosida DF_a.cna
Created/used by	farmasiusd 28 Oktober 2013 9:26:28
Current user	farmasiusd

Detection - CAMAG TLC Scanner 3

Information

Application position	10,0 mm
Solvent front position	75,0 mm

Instrument

Executed by	CAMAG TLC Scanner 3 "Scanner3_160602" S/N 160602 (1.14.28) farmasiusd 23 Oktober 2013 11:23:22
Number of tracks	11
Position of first track X	25,0 mm
Distance between tracks	15,0 mm
Scan start pos. Y	20,0 mm
Scan end pos. Y	160,0 mm
Slit dimensions	10,00 x 0,40 mm, Macro
Optimize optical system	Light
Scanning speed:	20 mm/s
Data resolution:	100 µm/step

Measurement Table

Wavelength	400
Lamp	D2 & W
Measurement Type	Remission
Measurement Mode	Absorption
Optical filter	Second order
Detector mode	Automatic
PM high voltage	428 V

Detector properties

Y-position for 0 adjust	20,0 mm
Track # for 0 adjust	0
Analog Offset	10%
Sensitivity	Automatic (36)

Integration

Properties

Data filtering	Savitsky-Golay 7
Baseline correction	Lowest Slope
Peak threshold min. slope	5
Peak threshold min. height	10 AU
Peak threshold min. area	50
Peak threshold max. height	990 AU
Track start position	20,1 mm
Track end position	159,9 mm
Display scaling	Automatic

User : farmasiusd
28 Oktober 2013 9:26:30

Approved :. . . . . .
Report ID : 07DD0A1C02091A1C

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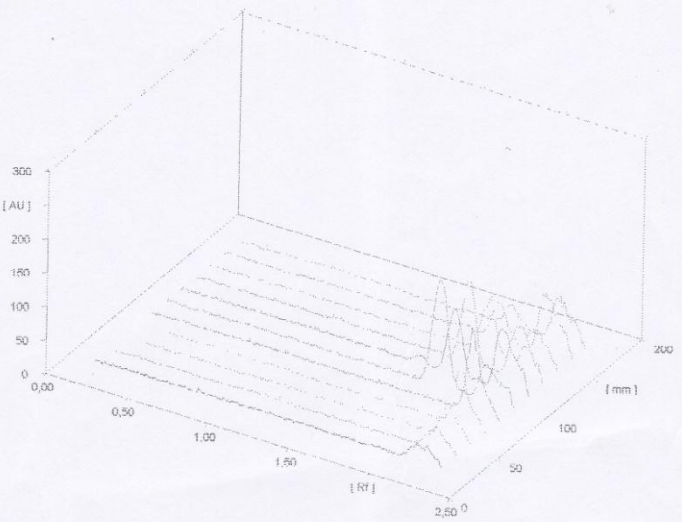
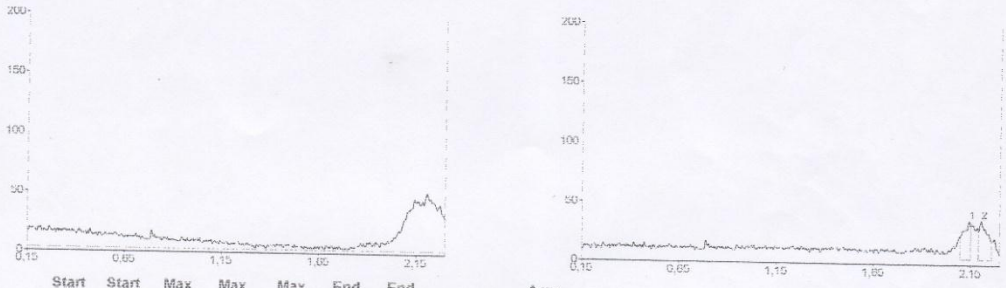
winCATS Planar Chromatography Manager

All tracks at Wavelength



Track 1, ID: baku 1



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	2,10	13,3	2,15	27,4	49,81	2,16	23,6	763,2	46,50	unknown *
2	2,20	18,8	2,21	27,6	50,19	2,27	9,8	878,1	53,50	unknown *

1641,3

User : farmasiusd
28 Oktober 2013 9:26:30

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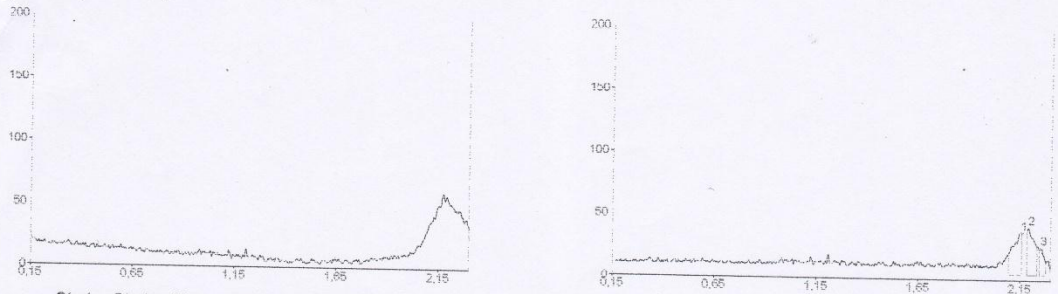
winCATS Planar Chromatography Manager

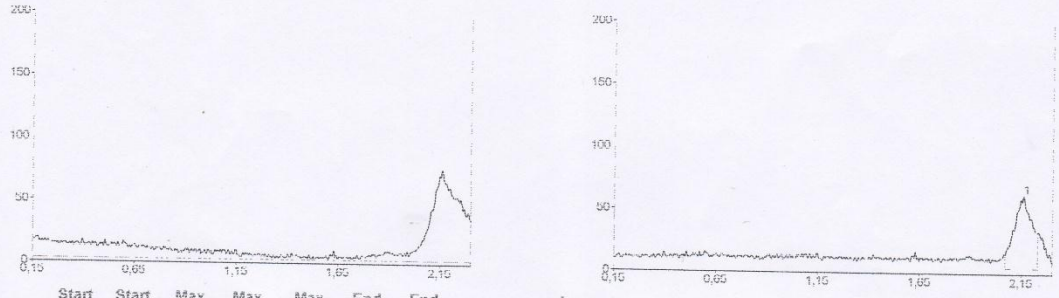
Track 2, ID: baku 2



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	2,10	11,5	2,16	28,8	36,36	2,16	28,3	915,0	44,29	unknown *
2	2,19	30,3	2,20	33,3	42,11	2,24	18,0	873,5	42,29	unknown *
3	2,25	15,6	2,26	17,0	21,54	2,28	5,4	277,2	13,42	unknown *

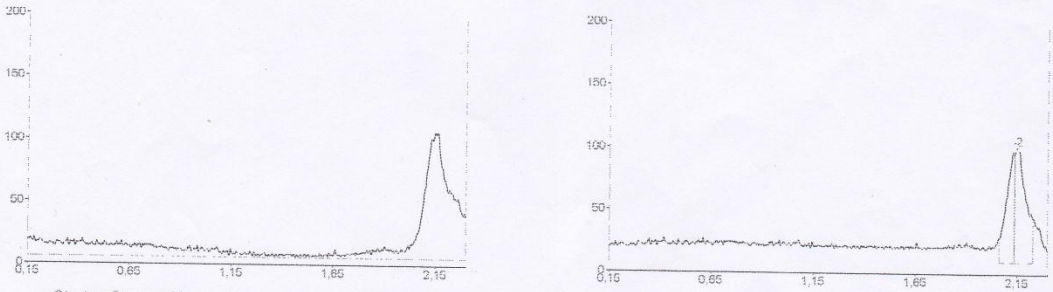
1788,5

Track 3, ID: baku 4



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	2,08	6,0	2,17	53,3	100,00	2,24	23,7	3339,1	100,00	unknown *

Track 4, ID: baku 8



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	2,07	9,5	2,14	79,2	49,08	2,14	76,8	2210,9	41,13	unknown *
2	2,15	77,3	2,15	82,2	50,92	2,24	25,5	3164,8	58,87	unknown *

5375,7

User : farmasiusd
28 Oktober 2013 9:26:30

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Report ID : 07DD0A1C02091A1C

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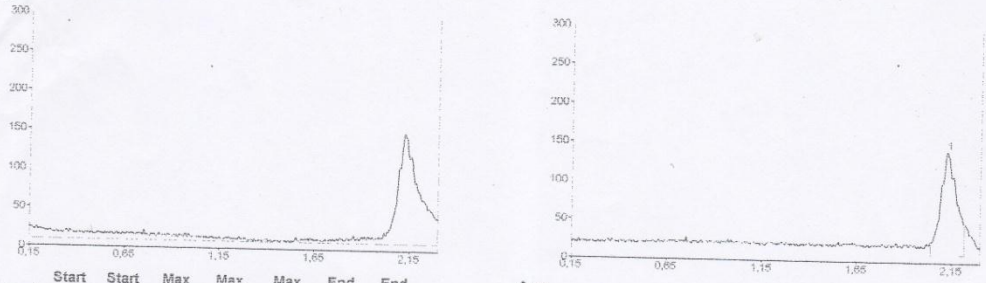
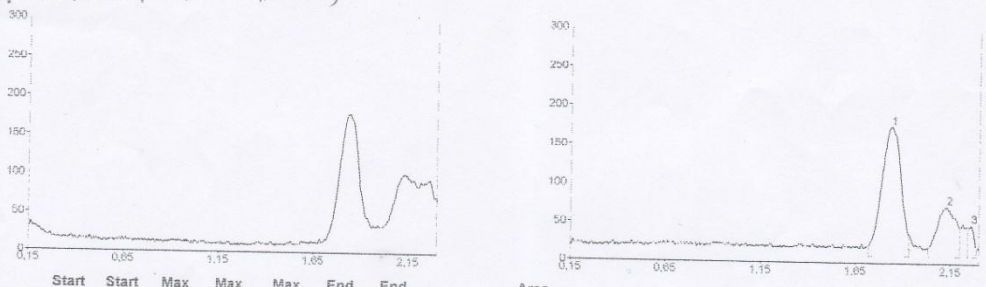
winCATS Planar Chromatography Manager

Track 5, ID: baku 16



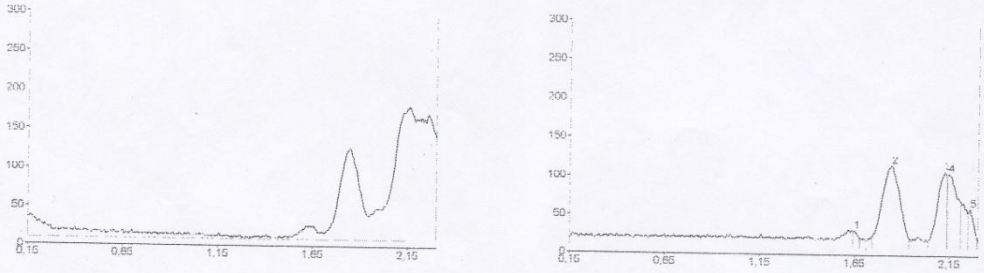
Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	2,04	6,3	2,13	124,4	100,00	2,23	30,7	7507,8	100,00	unknown *

Track 6, ID: sampel 0/1 (0,37gr ad 5ml)



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,72	3,0	1,84	157,5	64,98	1,94	17,5	11370,4	71,52	unknown *
2	2,04	1,6	2,14	54,1	22,33	2,21	28,1	3875,1	24,38	unknown *
3	2,25	27,2	2,27	30,8	12,69	2,29	2,8	651,7	4,10	unknown *

Track 7, ID: sampel 1/0 (1,75gr ad 5ml)



User : farmasiusd
28 Oktober 2013 9:26:30

Approved : .....
Report ID : 07DD0A1C02091A1C

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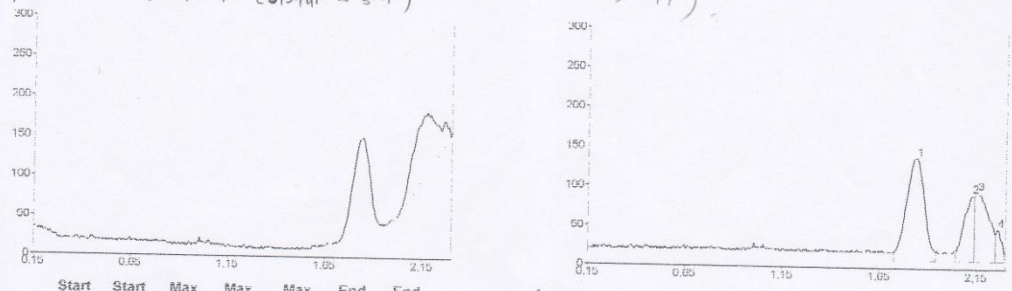
winCATS Planar Chromatography Manager

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,65	10,7	1,65	12,6	3,83	1,68	2,1	215,5	1,55	unknown *
2	1,75	5,1	1,85	97,0	29,57	1,94	2,6	6506,5	46,73	unknown *
3	2,04	0,8	2,13	89,7	27,35	2,14	86,0	3072,7	22,07	unknown *
4	2,14	85,3	2,15	87,0	26,53	2,21	52,3	3275,2	23,52	unknown *
5	2,25	35,7	2,26	41,7	12,70	2,30	7,6	854,2	6,13	unknown *

6347,9

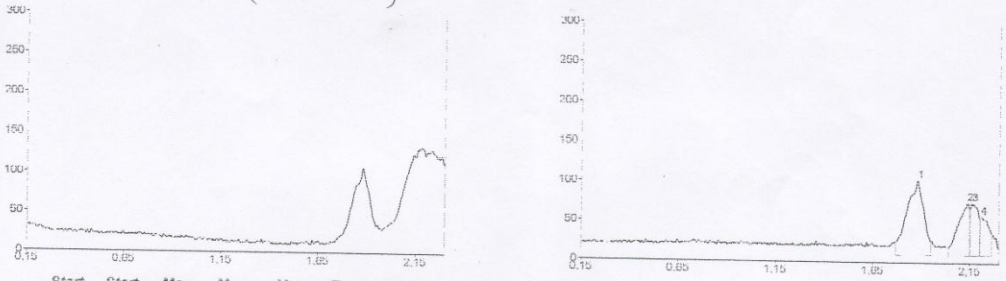
Track 8, ID: sampel 0,25/0,75 (3,34gr ad 5ml)



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,73	1,2	1,85	121,1	39,57	1,95	2,2	7880,4	51,20	unknown *
2	2,05	4,0	2,14	74,4	24,31	2,15	73,0	2768,3	17,98	unknown *
3	2,15	73,2	2,16	78,4	25,62	2,25	25,5	4089,9	26,57	unknown *
4	2,26	25,7	2,27	32,2	10,51	2,30	0,1	653,6	4,25	unknown *

6850,2

Track 9, ID: sampel 0,75/0,25 (3,32gr ad 5ml)



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,78	7,0	1,89	84,7	35,65	1,96	7,2	5079,3	49,50	unknown *
2	2,05	0,7	2,15	56,8	23,91	2,16	50,2	2225,4	21,69	unknown *
3	2,16	50,7	2,17	56,3	23,70	2,21	38,1	1677,8	16,35	unknown *
4	2,21	38,4	2,22	39,8	16,74	2,27	18,1	1278,7	12,46	unknown *

3903,2

User : farmasiusd
28 Oktober 2013 9:26:30

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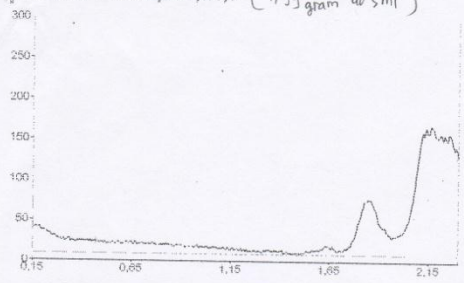
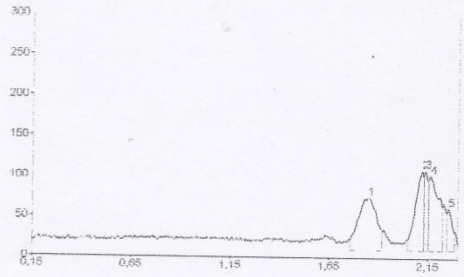
winCATS Planar Chromatography Manager

Track 10, ID: sampel 0,5/0,5 (1,99 gram ad 5ml)



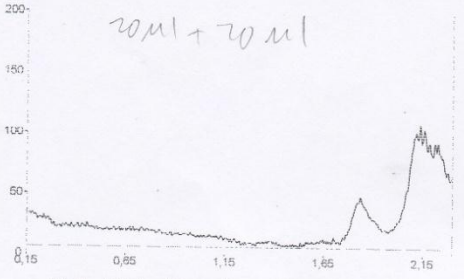


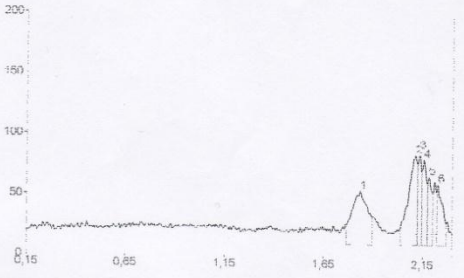
Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,76	3,7	1,85	55,9	15,35	1,92	16,6	3635,6	31,75	unknown *
2	2,05	4,7	2,13	90,4	24,83	2,14	84,3	2664,2	23,26	unknown *
3	2,14	85,4	2,14	90,8	24,94	2,16	78,5	1272,6	11,11	unknown *
4	2,16	78,8	2,17	84,1	23,09	2,23	45,7	3084,5	26,93	unknown *
5	2,25	37,9	2,26	42,9	11,79	2,29	13,4	795,3	6,94	unknown *

7021,3

Track 11, ID: kalus + baku

20 µl + 20 µl





Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,77	6,8	1,84	34,5	11,00	1,90	13,2	1868,3	25,93	unknown *
2	2,04	4,2	2,12	64,1	20,41	2,13	57,0	2063,7	28,64	unknown *
3	2,13	57,4	2,14	67,9	21,63	2,15	51,2	837,9	11,63	unknown *
4	2,16	51,8	2,16	60,8	19,37	2,18	40,9	941,3	13,06	unknown *
5	2,18	41,4	2,19	46,3	14,77	2,21	32,9	715,9	9,93	unknown *
6	2,23	38,6	2,23	40,2	12,82	2,28	7,8	778,9	10,81	unknown *

4558,8

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28 Oktober 2013 9:26:30

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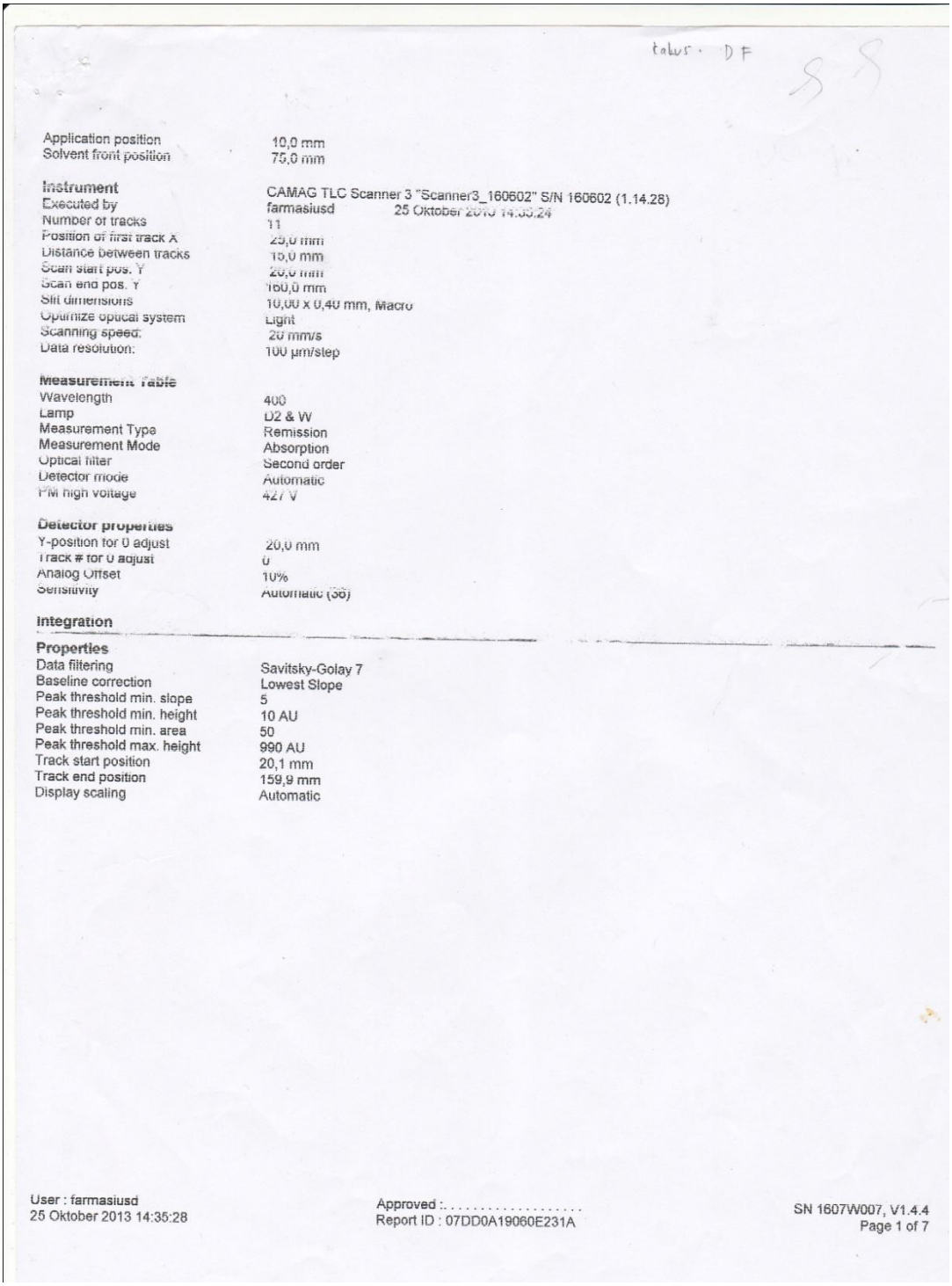
Replikasi 2



Application position	10,0 mm
Solvent front position	75,0 mm
Instrument	
Executed by	CAMAG TLC Scanner 3 "Scanner3_160602" S/N 160602 (1.14.28)
	farmasiusd 25 Oktober 2013 14:35:24
Number of tracks	11
Position of first track X	25,0 mm
Distance between tracks	15,0 mm
Scan start pos. Y	20,0 mm
Scan end pos. Y	160,0 mm
Slit dimensions	10,00 x 0,40 mm, Macro
Optimize optical system	Light
Scanning speed:	20 mm/s
Data resolution:	100 µm/step
Measurement Table	
Wavelength	400
Lamp	D2 & W
Measurement Type	Remission
Measurement Mode	Absorption
Optical filter	Second order
Detector mode	Automatic
PM high voltage	427 V
Detector properties	
Y-position for 0 adjust	20,0 mm
Track # for 0 adjust	0
Analog Offset	10%
Sensitivity	Automatic (56)
Integration	
Properties	
Data filtering	Savitsky-Golay 7
Baseline correction	Lowest Slope
Peak threshold min. slope	5
Peak threshold min. height	10 AU
Peak threshold min. area	50
Peak threshold max. height	990 AU
Track start position	20,1 mm
Track end position	159,9 mm
Display scaling	Automatic

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25 Oktober 2013 14:35:28

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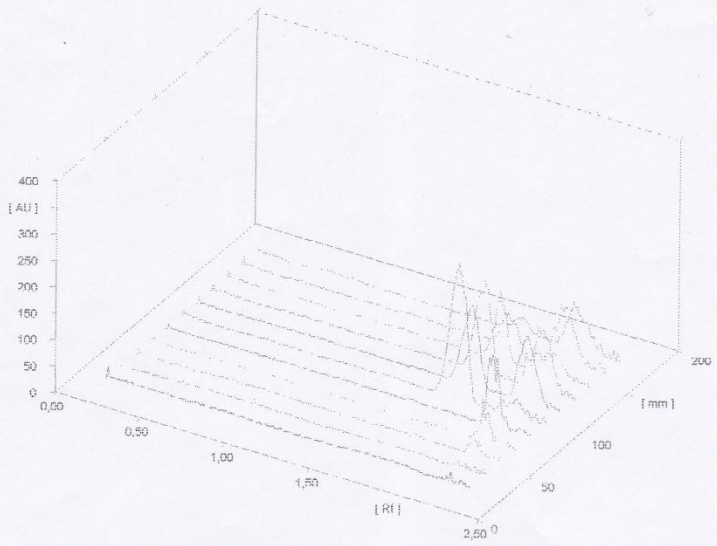
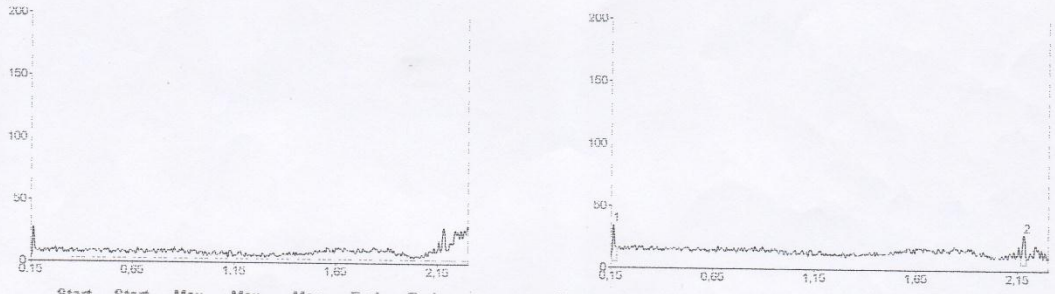
winCATS Planar Chromatography Manager

All tracks at Wavelength



Track 1, ID: baku 1



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0,16	1,5	0,17	24,8	56,40	0,18	5,4	191,6	52,62	unknown *
2	2,17	1,0	2,19	19,2	43,60	2,20	1,1	172,5	47,38	unknown *

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winCATS Planar Chromatography Manager

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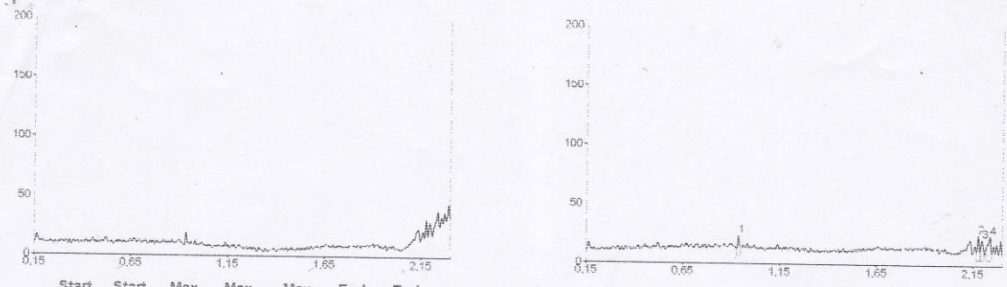
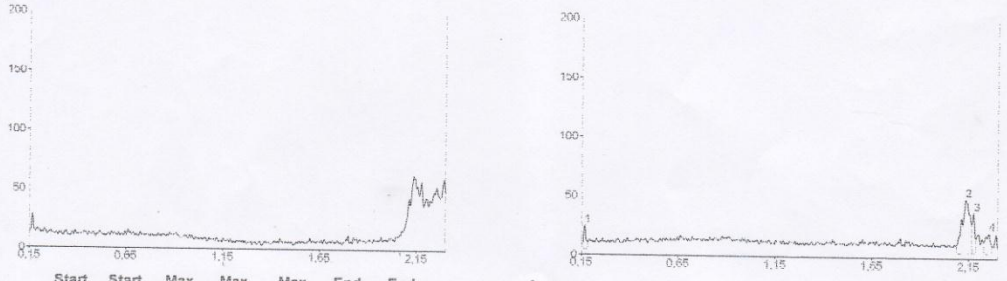
Track 2, ID: baku 2



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0,93	3,1	0,94	13,8	24,04	0,95	4,4	97,1	21,13	unknown *
2	2,17	1,6	2,18	16,0	27,80	2,19	2,0	98,5	21,43	unknown *
3	2,19	3,2	2,20	12,2	21,20	2,21	2,8	101,1	22,00	unknown *
4	2,23	6,4	2,24	15,5	26,96	2,25	1,0	162,9	35,44	unknown *

199,6

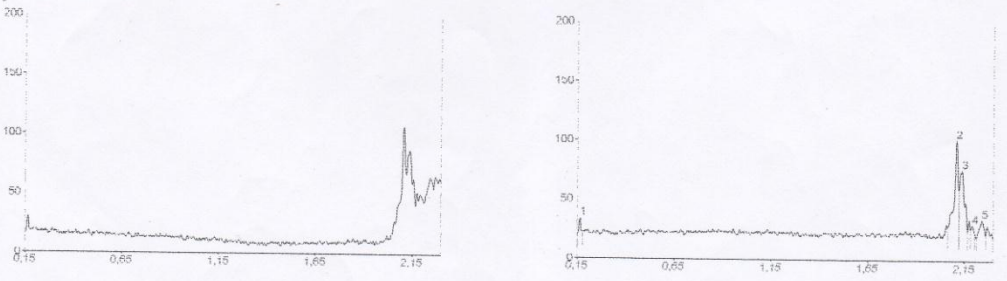
Track 3, ID: baku 4



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0,16	0,1	0,17	14,7	15,28	0,19	0,8	100,5	5,57	unknown *
2	2,09	2,2	2,14	40,3	42,02	2,17	19,4	1213,3	67,29	unknown *
3	2,17	19,9	2,18	28,8	29,98	2,19	6,8	278,7	15,46	unknown *
4	2,24	4,2	2,26	12,2	12,72	2,28	2,3	210,7	11,69	unknown *

1492

Track 4, ID: baku 8

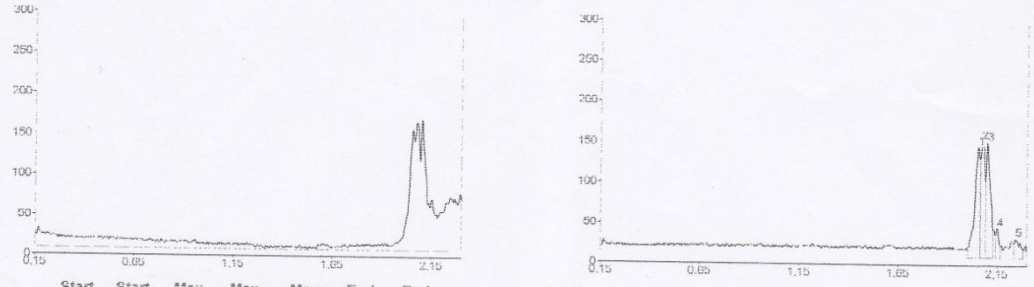


winCATS Planar Chromatography Manager

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0,16	1,1	0,17	13,6	7,66	0,18	1,9	97,8	3,30	unknown *
2	2,07	8,3	2,11	82,0	46,31	2,13	40,0	1320,3	44,49	unknown *
3	2,13	40,2	2,14	56,5	31,89	2,17	5,0	1163,6	39,20	unknown *
4	2,19	6,6	2,20	10,4	5,85	2,21	4,1	124,0	4,18	unknown *
5	2,22	0,0	2,25	14,7	8,28	2,27	2,0	262,2	8,83	unknown *

2607,9

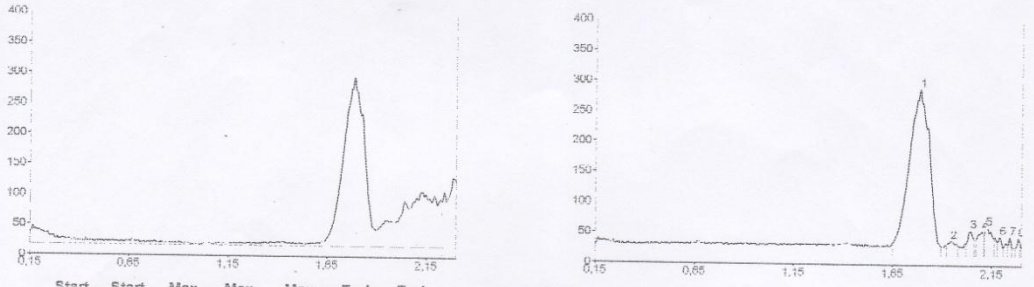
Track 5, ID: baku 16



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	2,01	3,2	2,06	128,1	29,23	2,07	113,2	2444,8	33,74	unknown *
2	2,07	114,1	2,08	135,2	30,84	2,10	84,9	2119,8	29,26	unknown *
3	2,10	90,7	2,11	132,9	30,32	2,14	24,4	2088,5	28,83	unknown *
4	2,15	19,5	2,16	27,5	6,26	2,18	6,2	326,7	4,51	unknown *
5	2,24	9,1	2,25	14,7	3,35	2,29	0,1	265,3	3,66	unknown *

6979,8

Track 6, ID: sampel 0/1



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	1,66	6,5	1,79	259,9	63,80	1,90	2,4	19108,2	87,42	unknown *
2	1,93	3,8	1,95	10,5	2,58	1,99	2,5	273,0	1,25	unknown *
3	2,03	7,0	2,05	27,9	6,86	2,07	13,8	537,4	2,46	unknown *
4	2,08	13,6	2,11	26,7	6,55	2,12	22,9	585,9	2,68	unknown *
5	2,12	23,2	2,13	32,6	8,01	2,17	15,3	869,5	3,98	unknown *
6	2,19	9,3	2,20	17,3	4,24	2,21	0,3	209,7	0,96	unknown *
7	2,24	3,5	2,25	17,1	4,20	2,26	0,2	136,4	0,62	unknown *
8	2,27	0,9	2,29	15,3	3,76	2,30	7,8	138,3	0,63	unknown *

2202,5

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25 Oktober 2013 14:35:28

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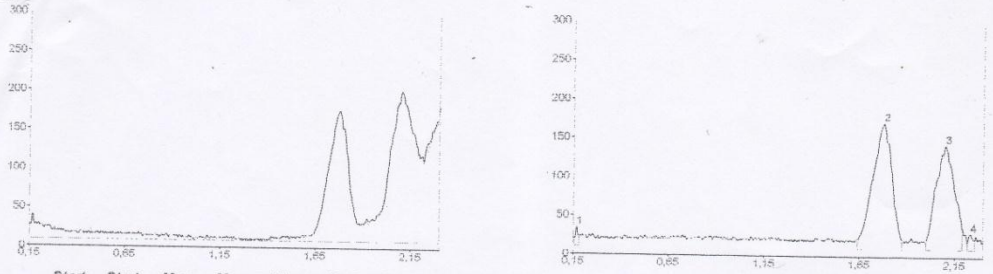
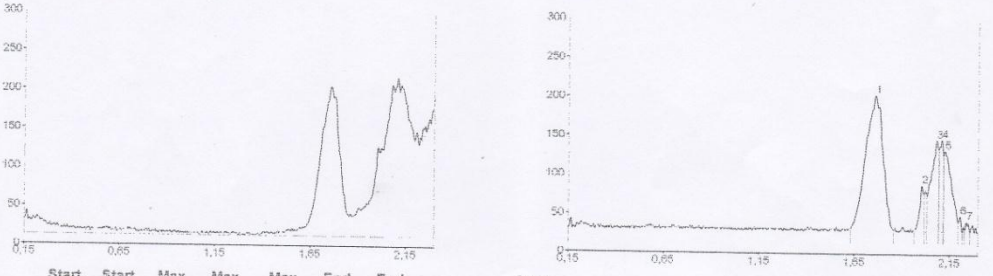
Track 7, ID: sampel 1/0



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %		Assigned substance
1	0,16	0,0	0,17	13,8	4,55	0,18	2,3	97,3	0,48		unknown *
2	1,65	3,0	1,78	153,4	50,54	1,88	2,2	11158,1	55,09		unknown *
3	2,00	2,6	2,11	125,3	41,29	2,20	17,8	8824,7	43,57	✓	unknown *
4	2,23	0,0	2,24	11,0	3,62	2,26	3,8	174,5	0,86		unknown *

Track 8, ID: sampel 0,25/0,75



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %		Assigned substance
1	1,64	3,2	1,77	174,8	28,83	1,86	1,7	12030,1	55,10		unknown *
2	1,97	2,9	2,01	59,0	9,73	2,02	49,0	974,6	4,46	✓	unknown *
3	2,04	47,2	2,09	117,3	19,34	2,10	106,6	3292,7	15,08	✓	unknown *
4	2,10	107,4	2,11	118,2	19,50	2,12	98,5	1869,8	8,56	✓	unknown *
5	2,12	99,2	2,13	103,9	17,13	2,20	12,4	3319,9	15,21	✓	unknown *
6	2,20	12,5	2,21	19,9	3,28	2,22	0,4	189,4	0,87	✓	unknown *
7	2,24	2,7	2,25	13,3	2,19	2,26	0,8	156,5	0,72		unknown *

9646,4

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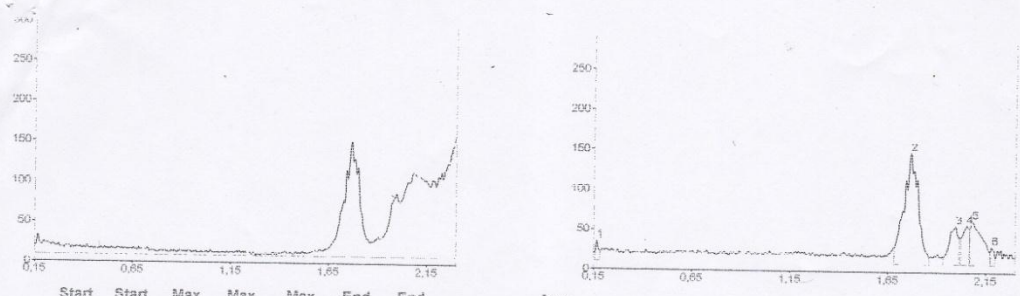
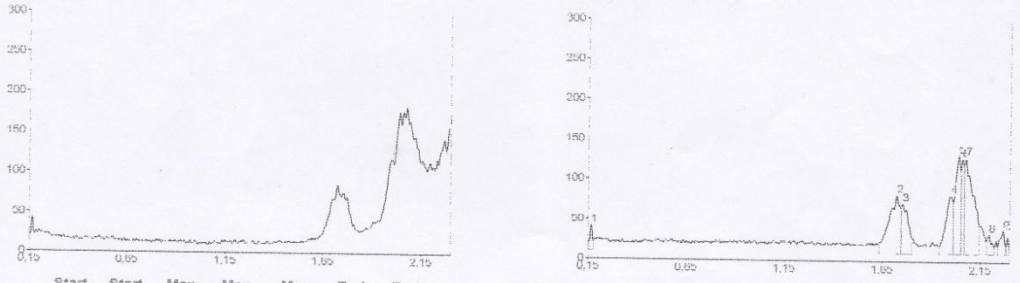
Track 9, ID : sampel 0,75 / 0,25 .



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0,16	2,5	0,17	14,6	5,19	0,18	3,1	113,5	1,02	unknown *
2	1,69	11,6	1,77	130,3	46,32	1,87	0,2	6643,7	59,89	unknown *
3	1,94	0,6	2,00	38,8	13,81	2,02	24,3	1205,6	10,87	unknown *
4	2,03	24,6	2,06	39,8	14,13	2,07	36,5	1010,3	9,11	unknown *
5	2,07	36,6	2,09	45,1	16,03	2,18	6,8	1969,8	17,76	unknown *
6	2,18	7,7	2,19	12,7	4,52	2,20	0,4	150,2	1,35	unknown *

4335,9

Track 10, ID: sampel 0,5/0,5



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0,16	0,6	0,17	21,9	3,77	0,18	2,1	145,1	1,10	unknown *
2	1,64	3,5	1,73	62,8	10,81	1,75	48,3	2481,7	18,83	unknown *
3	1,75	48,9	1,76	52,6	9,05	1,81	9,6	1322,1	10,03	unknown *
4	1,95	0,3	2,00	63,7	10,96	2,02	54,9	1620,1	12,30	unknown *
5	2,02	55,5	2,05	113,4	19,51	2,06	95,3	2254,9	17,11	unknown *
6	2,06	96,7	2,06	108,7	18,71	2,07	100,9	1142,4	8,67	unknown *
7	2,07	101,1	2,08	110,7	19,06	2,15	26,8	3658,9	27,77	unknown *
8	2,19	8,1	2,20	15,2	2,61	2,23	0,0	201,6	1,53	unknown *
9	2,24	0,5	2,27	20,2	3,47	2,28	1,1	291,0	2,21	unknown *
10	2,29	0,7	2,30	12,0	2,07	2,30	1,6	58,4	0,44	unknown *

8877,9

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25 Oktober 2013 14:35:28

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Lampiran 10. Perhitungan kadar steviosida dalam daun dan ekstrak kalus daun Stevia

- Perhitungan kadar steviosida standard

  Kadar steviosida dalam pelarut n-butanol = 500 mg/ml

  Diambil 2 ml diencerkan sampai 10 ml, sehingga diperoleh kadar 100 mg/ml kemudian diambil 0,5 ml dan dijadikan 25 ml

  Kadar steviosida standard = 0,5 x 100 / 25 = 2 mg/ml = 2 µg/µl

- Pembuatan kurva baku steviosida standard dengan lima macam konsentrasi

  Kadar = jumlah totolan x kadar steviosida standard

  a. Jumlah totolan = 1 µl

     Kadar 1 = 1 x 2 µg/µl = 2 µg/µl

  b. Jumlah totolan = 2 µl

     Kadar 2 = 2 x 2 µg/µl = 4 µg/µl

  c. Jumlah totolan = 4 µl

     Kadar 3 = 4 x 2 µg/µl = 8 µg/µl

  d. Jumlah totolan = 8 µl

     Kadar 4 = 8 x 2 µg/µl = 16 µg/µl

  e. Jumlah totolan = 16 µl

     Kadar 5 = 16 x 2 µg/µl = 32 µg/µl

- Penetapan kadar steviosida daun Stevia (AUC steviosida pada Rf 0,79 – 0,81)

Jml totolan (µl)	Kadar (µg/µl)	AUC (luas bercak) Replikasi 1	AUC (luas bercak) Replikasi 2
1	2	1328,2	1578,2
2	4	5324,8	5400,8
4	8	9147,5	9267,7
8	16	12955,1	12611,8
16	32	21171	21404,4

Persamaan regresi : Y = A + BX

Replikasi 1 Y = 2427 + 609,54 X (perhitungan kalkulator)

r = 0,9778

Replikasi 2 Y = 2503,1875 + 608,82 X (perhitungan kalkulator)

r = 0,9805

Bobot sampel = 3 g

Volume penotolan = 10 µl

Volume pengenceran = 5 ml

Replikasi 1 (AUC = 4873,9)

Y = 2427 + 609,54 X

X = 4873,9 – 2427 / 609,54 = 4,0143 µg

Kadar dalam 5 ml = 5000 / 10 µl x 4,0143 µg / 1000 = 2,01 mg

Kadar dalam tiap sampel = 2,01 mg / 3 g = 0,67 mg/g → 0,067 %

Replikasi 2 (AUC = 4760,1)

Y = 2503,1875 + 608,82X

X = 4760,1+ 2503,1875 / 608,82 = 3,707 µg

Kadar dalam 5 ml = 5000 / 10 µl x 3,707 µg / 1000 = 1,85 mg

Kadar dalam tiap sampel = 1,85 mg / 3 g = 0,62 mg/g → 0,062 %

Rata – rata kadar = replikasi 1 + replikasi 2 / 2

= 0,67 mg/g + 0,62mg/g / 2 = 0,64 mg/g → 0,064 %

SD = 3,5 x 10^{-3} (perhitungan kalkulator)

- Penetapan kadar steviosida dalam kalus (AUC steviosida pada Rf 0,78 - 0,86)

Jml totolan (µl)	Kadar (µg/ µl)	AUC (luas bercak) Replikasi 1	AUC (luas bercak) Replikasi 2
1	2	1641,3	172,5
2	4	1788,5	199,6
4	8	3339,1	1492
8	16	5375,7	2607,9
16	32	7507,8	6979,8

Persamaan regresi : Y = A + BX

Replikasi 1 Y = 1436,304167 + 201,14 X (Perhitungan kalkulator)

r = 0,9802

Replikasi 2 Y = -546,2458 + 228,7585 X (Perhitungan kalkulator)

r = 0,9930

Bobot sampel DF 0/1 = 0,37 g

DF 0,25/0,75 = 3,34 g

DF 0,5/0,5 = 1,99 g

DF 0,75/0,25 = 3,32 g

DF 1/0 = 1,75 g

Volume penotolan = 40 µl

Volume pengenceran = 5 ml

DF 0/1

Replikasi 1 (AUC = 3875,1)

Y = 1436,304167 + 201,14 X

X = 201,14 -1436,304167 / 201,14 = 12,125 µg

Kadar dalam 5 ml = 5000 / 40 µl x 12,125 µg/ 1000 = 1,52 mg

Kadar dalam tiap sampel = 1,52 mg / 0,37 g = 4,10 mg/g → 0,410 %

Replikasi 2 (AUC = 2202,5)

Y = -546,2458 + 228,7585 X

X = 2202,5 + 546,2458 / 228,7585 = 12,016 µg

Kadar dalam 5 ml = 5000 / 40 µl x 12,016 µg / 1000 = 1,502 mg

Kadar dalam tiap sampel = 1,502 mg / 0,37 g = 4,06 mg/g → 0,406 %

Rata – rata kadar = replikasi 1 + replikasi 2 / 2

= 4,10 mg/g + 4,06 mg/g / 2 = 4,08 mg/g → 0,408 %

SD = $2,82 \times 10^{-3}$ (perhitungan kalkulator)

DF 0,25/0,75

Replikasi 1 (AUC = 6858,2)

Y = 1436,304167 + 201,14 X

X = 6858,2-1436,304167 / 201,14 = 26,955 µg

Kadar dalam 5 ml = 5000 / 40 µl x 26,955 µg / 1000 = 3,37 mg

Kadar dalam tiap sampel = 3,37 / 3,34 g = 1,01 mg/g → 0,101 %

Replikasi 2 (AUC = 9646,4)

Y = -546,2458 + 228,7585 X

X = 9646,4 + 546,2458 / 228,7585 = 44,556 µg

Kadar dalam 5 ml = 5000 / 40 µl x 44,556 µg / 1000 = 5,569 mg

Kadar dalam tiap sampel = 5,569 mg / 3,34 g = 1,66 mg/g → 0,166 %

Rata – rata kadar = replikasi 1 + replikasi 2 / 2

=1,01 + 1,66 / 2 = 1,33 mg/g → 0,133 %

SD = 0,046 (perhitungan kalkulator)

DF 0,5/0,5

Replikasi 1 (AUC = 7021,3)

Y = 1436,304167 + 201,14 X

X = 7021,3 -1436,304167 / 201,14 = 27,766 µg

Kadar dalam 5 ml = 5000 / 40 µl x 27,766 µg / 1000 = 3,47 mg

Kadar dalam tiap sampel = 3,47mg / 1,99 g = 1,74 mg/g → 0,174 %

Replikasi 2 (AUC = 8877,9)

Y = -546,2458 + 228,7585 X

X = 8877,9 + 546,2458 / 228,7585 = 41,197 µg

Kadar dalam 5 ml = 5000 / 40 µl x 41,197 µg / 1000 = 5,15 mg

Kadar dalam tiap sampel = 5,15 mg / 1,99 g = 2,58 mg/g → 0,258 %

Rata – rata kadar = replikasi 1 + replikasi 2 / 2

= 1,74 mg/g + 2,58 mg/g / 2 = 2,16 mg/g → 0,216 %

SD = 0,059 (perhitungan kalkulator)

DF 0,75/0,25

Replikasi 1 (AUC = 3903,2)

Y = 1436,304167 + 201,14 X

X = 3903,2-1436,304167 / 201,14 = 12,264 µg

Kadar dalam 5 ml = 5000 / 40 µl x 12,264 µg / 1000 = 1,53 mg

Kadar dalam tiap sampel = 1,53 mg/g / 3,32 g = 0,46 mg/g → 0,046 %

Replikasi 2 (AUC = 4335,9)

Y = -546,2458 + 228,7585 X

X = 4335,9 + 546,2458 / 228,7585 = 21,342 µg

Kadar dalam 5 ml = 5000 / 40 x 21,342 / 1000 = 2,67 mg

Kadar dalam tiap sampel = 2,67 mg / 3,32 g = 0,803 mg/g → 0,803 %

Rata – rata kadar = replikasi 1 + replikasi 2 / 2

= 0,46 mg/g + 0,803 mg/g / 2 = 0,63 mg/g → 0,063 %

SD = 0,0242 (perhitungan kalkulator)

DF 1/0

Replikasi 1 (AUC = 6347,9)

Y = 1436,304167 + 201,14 X

X = 6347,9-1436,304167 / 201,14 = 24,418 µg

Kadar dalam 5 ml = 5000 / 40 µl x 24,418µg / 1000 = 3,05 mg

Kadar dalam tiap sampel = 3,05 mg / 1,75 g = 1,74 mg/g → 0,174 %

Replikasi 2 (AUC = 8824,7)

Y = -546,2458 + 228,7585 X

X = 8824,7 + 546,2458 / 228,7585 = 40,964 µg

Kadar dalam 5 ml = 5000 / 40 µl x 40,964 µg / 1000 = 5,120 mg

Kadar dalam tiap sampel = 5,120 mg / 1,75 g = 2,93 mg/g → 0,0293 %

Rata – rata kadar = replikasi 1 + replikasi 2 / 2

=1,74 mg/g + 2,93 mg/g / 2 = 2,33 mg/g → 0,233 %

SD = $8,4 \times 10^{-3}$ (perhitungan kalkulator)

Lampiran 11. Perhitungan harga Retardian factor (hRf) daun dan ekstrak kalus dauin Stevia dengan kromatografi lapis tipis

$$hRf = \frac{\text{jarak titik pusat bercak dari titik awal}}{\text{jarak yang ditempuh pengembang}} \times 100$$

Sampel (kalus)	Jarak titik pusat bercak dari titik awal (cm)		Perhitungan hRf	
	Replikasi 1	Replikasi 2	Replikasi 1	Replikasi 2
A	13	12,4	$\frac{13}{15} \times 100 = 86,6$	$\frac{12,4}{15} \times 100 = 82,6$
B	13	12,4	$\frac{13}{15} \times 100 = 86,6$	$\frac{12,4}{15} \times 100 = 82,6$
C	13	12,4	$\frac{13}{15} \times 100 = 86,6$	$\frac{12,4}{15} \times 100 = 82,6$
D	13	12	$\frac{13}{15} \times 100 = 86,6$	$\frac{12}{15} \times 100 = 80$
E	12,8	11,7	$\frac{12,8}{15} \times 10 = 85,3$	$\frac{11,7}{15} \times 100 = 78$
1	12,9	12	$\frac{12,9}{15} \times 100 = 86$	$\frac{12}{15} \times 100 = 80$
2	12,8	11,7	$\frac{12,8}{15} \times 100 = 85,3$	$\frac{11,7}{15} \times 100 = 78$
3	12,8	11,8	$\frac{12,8}{15} \times 10 = 85,3$	$\frac{11,8}{15} \times 100 = 79$
4	12,8	12	$\frac{12,8}{15} \times 10 = 85,3$	$\frac{12}{15} \times 100 = 80$
5	12,8	11,7	$\frac{12,8}{15} \times 10 = 85,3$	$\frac{11,7}{15} \times 100 = 78$

Keterangan :

A : standar stevosida 1 µl
B : standar stevosida 2 µl
C : standar stevosida 4 µl
D : standar stevosida 8 µl
E : standar stevosida 16 µl

1 : kinetin 1 ppm
2 : 2,4-D 1 ppm
3 : 2,4-D 0,25 ppm, kinetin 0,75 ppm
4 : 2,4-D 0,75 ppm, kinetin 0,25 ppm
5 : 2,4-D 0,5 ppm, kinetin 0,5 ppm

Lampiran 12. Perhitungan harga Retardian factor (hRf) ekstrak daun Stevia dengan kromatografi lapis tipis

$$hRf = \frac{\text{jarak titik pusat bercak dari titik awal}}{\text{jarak yang ditempuh pengembang}} \times 100$$

Sampel (daun)	Jarak titik pusat bercak dari titik awal (cm)		Perhitungan hRf	
	Replikasi 1	Replikasi 2	Replikasi 1	Replikasi 2
A	12,2	12,1	$\frac{12,2}{15} \times 100 = 81,3$	$\frac{12,1}{15} \times 100 = 80,6$
B	12	12	$\frac{12}{15} \times 100 = 80$	$\frac{12}{15} \times 100 = 80$
C	12	11,9	$\frac{12}{15} \times 100 = 80$	$\frac{11,9}{15} \times 100 = 79,3$
D	11,9	11,9	$\frac{11,9}{15} \times 100 = 79,3$	$\frac{11,9}{15} \times 100 = 79,3$
E	11,9	11,9	$\frac{11,9}{15} \times 100 = 79,3$	$\frac{11,9}{15} \times 100 = 79,3$
a	11,7	11,6	$\frac{11,7}{15} \times 100 = 78$	$\frac{11,6}{15} \times 100 = 77,3$
b	12,2	12	$\frac{12,2}{15} \times 100 = 81,3$	$\frac{12}{15} \times 100 = 80$

Keterangan :

A : standar stevosida 1 µl

B : standar stevosida 2 µl

C : standar stevosida 4 µl

D : standar stevosida 8 µl

E : standar stevosida 16 µl

a : sampel daun Stevia 5 µl

b : sampel daun Stevia10 µl