

BAB V

KESIMPULAN DAN SARAN

A. Kesimpulan

Berdasarkan dari hasil penelitian yang telah dilakukan dapat diperoleh kesimpulan bahwa:

Pertama, *Nanostructured Lipid Carriers* (NLC) fisetin dapat dibuat menggunakan lipid golongan gliserida (Precirol) dengan metode emulsifikasi sonikasi.

Kedua, karakterisasi NLC Fisetin menghasilkan ukuran partikel yaitu F1 651,1 nm $\pm$ 7,11; F2 246,2 nm $\pm$ 2,45 dan F3 119,6 nm $\pm$ 5,36. Menghasilkan PDI yaitu pada F1 0,315; F2 0,266 dan F3 0,619. Menghasilkan zeta potensial yaitu pada F1 -13,13 mV; F2 -20,36 mV dan F3 -20,53 mV.

Ketiga, karakterisasi serum NLC Fisetin pada uji organoleptik menghasilkan warna kuning keruh, bau khas dan sedikit kental. Pada uji pH menghasilkan pH sebesar 6,27, pada uji homogenitas menghasilkan sediaan yang homogen dan pada uji viskositas menghasilkan viskositas sebesar 1000 cPs.

B. Saran

Penelitian ini masih banyak kekurangan, maka perlu dilakukan penelitian lebih lanjut mengenai :

Pertama, perlu dilakukan analisis *screening* lipid dengan menggunakan jenis lipid yang lebih beragam.

Kedua, perlu dilakukan uji *Transminssion Electron Microscopy* (TEM) untuk mengetahui morfologi NLC (*Nanostructured Lipid Carriers*) fisetin.

Ketiga, perlu dilakukan uji pelepasan sediaan (Difusi Franz).

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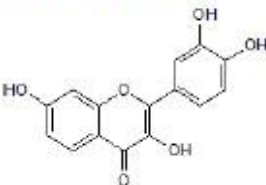
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Lampiran 1. Certificate of Analysis (CoA) Fisetin

		Product Information		Print Date: Jan 14 th 2016 www.tocris.com
Product Name:	Fisetin	Catalog No.:	5016	Batch No.: 1
CAS Number:	528-48-3			
IUPAC Name:	2-(3,4-Dihydroxyphenyl)-3,7-dihydroxy-4H-1-benzopyran-4-one			
Description: Naturally occurring flavonoid and antioxidant. Inhibits PI 3-K, Akt, mTOR and Cdk6. Displays antiproliferative activity in prostate cancer cells. Shown to activate ERK; exhibits neuroprotective activity in Huntington's disease models. Also a DNMT1 inhibitor.		Storage: Store at -20°C		
Physical and Chemical Properties: Batch Molecular Formula: C ₁₅ H ₁₀ O ₆ Batch Molecular Weight: 286.24 Physical Appearance: Yellow solid Minimum Purity: >98%		Solubility & Usage Info: DMSO to 100 mM ethanol to 10 mM		
Batch Molecular Structure: 		Stability and Solubility Advice: Some solutions can be difficult to obtain and can be encouraged by rapid stirring, sonication or gentle warming (in a 45-60°C water bath). Information concerning product stability, particularly in solution, has rarely been reported and in most cases we can only offer a general guide. Our standard recommendations are: SOLIDS: Provided storage is as stated on the product label and the vial is kept tightly sealed, the product can be stored for up to 6 months from date of receipt. SOLUTIONS: We recommend that stock solutions, once prepared, are stored aliquoted in tightly sealed vials at -20°C or below and used within 1 month. Wherever possible solutions should be made up and used on the same day.		
References: Lu et al (2005) Crystal structure of a human cyclin-dependent kinase 6 complex with a flavonol inhibitor, fisetin. <i>J.Med.Chem.</i> 48 737. PMID: 15689157. Lee et al (2005) Mechanisms for the inhibition of DNA methyltransferases by tea catechins and bioflavonoids. <i>Mol.Pharmacol.</i> 68 1018. PMID: 16037419. Haddad et al (2010) Antiproliferative mechanisms of the flavonoids 2,2'-dihydroxychalcone and fisetin in human prostate cancer cells. <i>Nutr.Cancer</i> 62 668. PMID: 20574928. Maher et al (2011) ERK activation by the polyphenols fisetin and resveratrol provides neuroprotection in multiple models of Huntington's disease. <i>Hum.Mol.Genet.</i> 20 261. PMID: 20952447. Adhami et al (2012) Dietary flavonoid fisetin: a novel dual inhibitor of PI3K/Akt and mTOR for prostate cancer management. <i>Biochem.Pharmacol.</i> 84 1277. PMID: 22842629.				
Caution - Not Fully Tested • Research Use Only • Not For Human or Veterinary Use				
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Lampiran 2. Certificate of Analysis (CoA) Precirol



GATTEFOSSÉ Certificate of Analysis

Product name : PRECIROL ATO 5

Item N° : 3092PPD

Batch N° : 169604

Manufacturing date : 05.2018

Expiry date : 05.2020

CHEMICAL DEFINITION :

Glycerol distearate (type II) EP / Glycerol distearate NF.

Characteristic	Method	Specification	Result
MANUFACTURING PLANT		Gattefossé : 01 / Innovia : 06	01/01
APPEARANCE	MA0193	Fine white powder	Conforms
ODOUR	MA0170	Faint	Conforms
COLOUR (Gardner Scale)	MA0214	< = 3.0	0.9
DROP POINT (METTLER)	MA0003	53.0 to 57.0 °C	56.3
ACID VALUE	MA0008	< = 6.00 mgKOH/g	1.76
SAPONIFICATION VALUE	MA0172	165 to 195 mgKOH/g	187
IODINE VALUE	MA0092	< = 3.0 gI2/100g	< = 3.0*
FREE GLYCEROL CONTENT	MA0006	< = 1.0 %	0.2
TOTAL MONOGLYCERIDES CONTENT	MA0006	8.0 to 22.0 %	17.4
TOTAL DIESTERS CONTENT	MA0006	40.0 to 60.0 %	50.7
TOTAL TRIESTERS CONTENT	MA0006	25.0 to 35.0 %	30.8
PALMITIC ACID (C16)	MA0002	40.0 to 60.0 %	44.5
STEARIC ACID (C18)	MA0002	40.0 to 60.0 %	53.2
SUM OF PALMITIC AND STEARIC ACIDS	MA0002	> = 90.0 %	97.7
WATER CONTENT	MA0007	< = 1.0 %	< 0.1
TOTAL ASHES CONTENT	MA0028	< = 0.1 %	< = 0.1*
NICKEL CONTENT	MA0256	< 1 ppm	Conforms
HEAVY METALS CONTENT (Pb)	MA0032	< 10 ppm	Conforms *

* Characteristic guaranteed but not tested.

This batch was released by our authorized Quality Manager and was found to meet the specifications as stated above. The above results do not discharge the customer from the obligation to carry out an inspection of goods received.

This report has been produced electronically and does not require a signature.

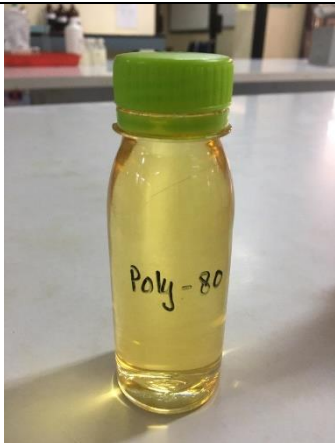
Printing Date : 09.26.2018

Quality Manager
Magali DRONNIER

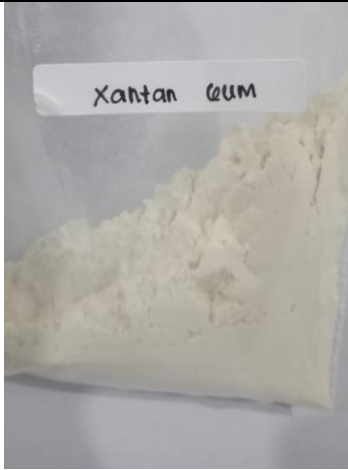
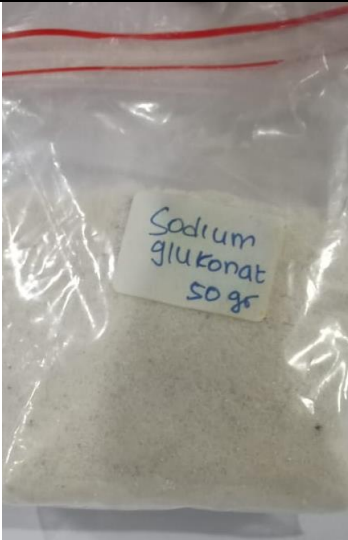
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Lampiran 3. Bahan yang digunakan untuk penelitian

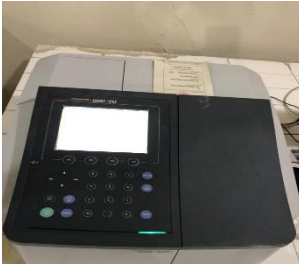
Nama Bahan	Foto Bahan	Kegunaan
Serbuk Fisetin		Zat aktif
Precirol		Sebagai Lipid padat dalam komponen NLC.
Isopropil Miristat		Sebagai lipid cair dalam komponen NLC.

Tween80		Sebagai surfaktan dalam komponen NLC.
Aquade-mineralisata		Sebagai pelarut.
Ethanol PA		Sebagai pelarut.

Hyaluronic Acid		Berfungsi sebagai humektan
Propilen Glikol		Sebagai humektan
Distilled Water		Sebagai pelarut pada sediaan serum.

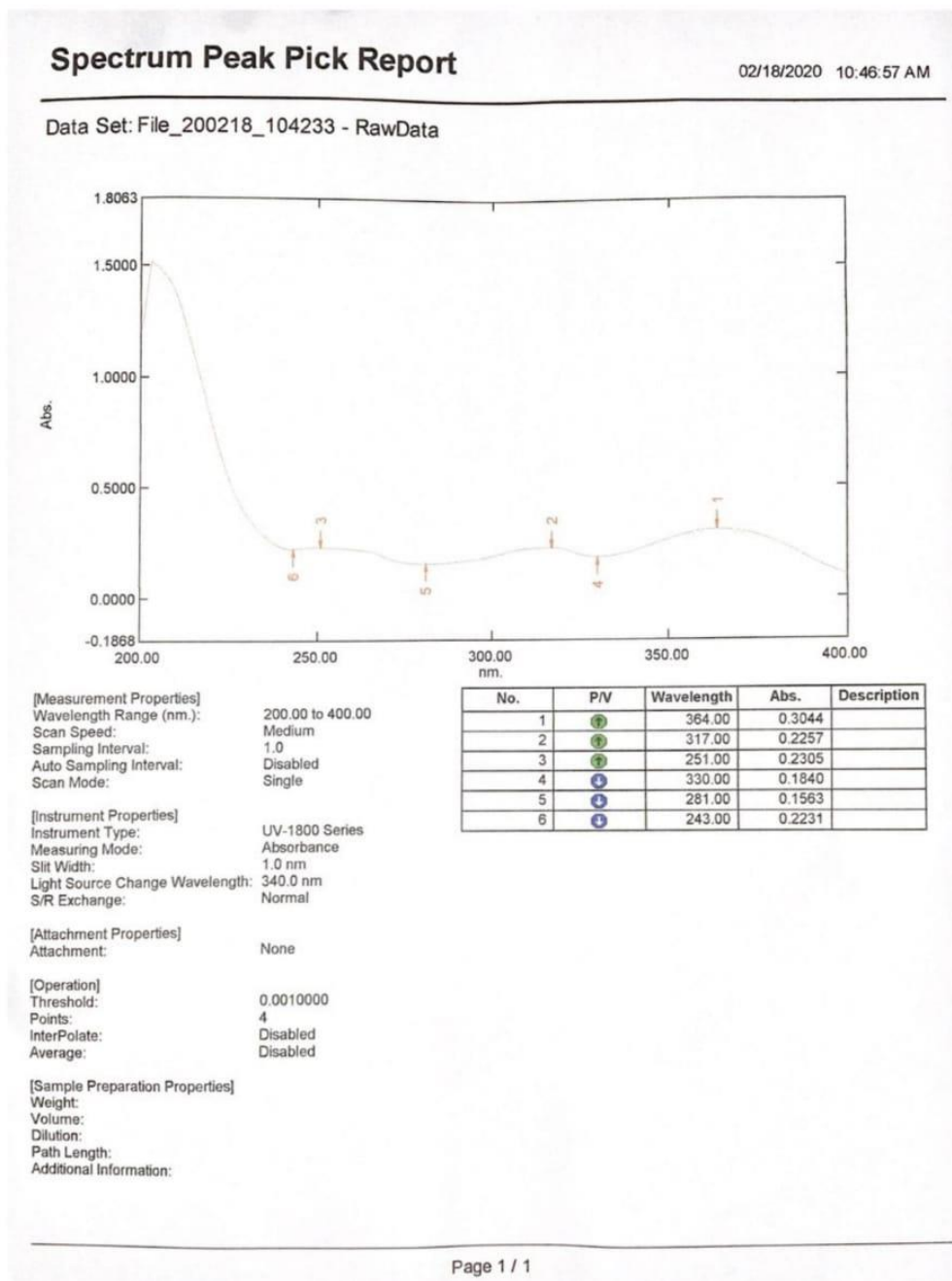
Xanthan Gum		Sebagai pelembut dalam serum.
Sodium Gluconate		Sebagai stabilizer dalam sediaan serum
Optiphen		Berfungsi sebagai pengawet

Lampiran 4. Alat yang digunakan dalam penelitian

Nama Alat	Foto Alat	Kegunaan
Spektrofotometri Uv-vis		Merupakan spektrofotometer yang digunakan untuk pengukuran serapan cahaya di daerah ultraviolet (200-350nm) dan sinar tampak (350-800nm) oleh suatu senyawa.
Particle Size Analyzer (PSA)		Untuk mengukur ukuran partikel.
Zeta nansizer		Analisa zeta potensial menggunakan hamburan cahaya elektroforesis.
Magnetic Stirrer		Berfungsi untuk menghomogenkan suatu larutan dengan pengadukan serta suhu.

Sonikator		Berfungsi memecah partikel dengan energi suara.
Viskometer		Untuk mengukur viskositas (kekentalan) suatu sediaan.
pH meter		Untuk mengukur pH (kadar keasaman) suatu sediaan.

Lampiran 5. Penentuan panjang gelombang dan pembuatan kurva baku



Hasil lamda max

Panjang gelombang maksimum yang diperoleh dari scanning larutan fisetin dalam etanol pro analisis sebesar 364 nm dengan serapan 0,3044 nm.

a. Penentuan *operating time*

Kinetics Data Print Report

02/18/2020 11:50:21 AM

Time (Minute)	RawData ...
0.000	0.301
1.000	0.301
2.000	0.301
3.000	0.301
4.000	0.300
5.000	0.300
6.000	0.300
7.000	0.301
8.000	0.301
9.000	0.301
10.000	0.300
11.000	0.301
12.000	0.300
13.000	0.300
14.000	0.300
15.000	0.300
16.000	0.300
17.000	0.300
18.000	0.300
19.000	0.300
20.000	0.300
21.000	0.300
22.000	0.300
23.000	0.300
24.000	0.300
25.000	0.300
26.000	0.300
27.000	0.300
28.000	0.300
29.000	0.300
30.000	0.300
31.000	
32.000	
33.000	
34.000	
35.000	
36.000	
37.000	
38.000	
39.000	
40.000	
41.000	
42.000	
43.000	
44.000	
45.000	
46.000	
47.000	
48.000	
49.000	
50.000	

b. Kurva kalibrasi (linieritas)

Penimbangan bahan :

Kertas + fisetin = 0,1053

Kertas kosong = 0,0807 _

0,0246 gr / 500 ml = 49,2 ppm

Perhitungan konsentrasi kurva kalibrasi :

- 1) $V_1 \times N_1 = V_2 \times N_2$
 $1 \text{ ml} \times 49,2 \text{ ppm} = 10 \text{ ml} \times N_2$
 $N_2 = 4,92 \text{ ppm}$
- 2) $V_1 \times N_1 = V_2 \times N_2$
 $1,2 \text{ ml} \times 49,2 \text{ ppm} = 10 \text{ ml} \times N_2$
 $N_2 = 5,904 \text{ ppm}$
- 3) $V_1 \times N_1 = V_2 \times N_2$
 $1,4 \text{ ml} \times 49,2 \text{ ppm} = 10 \text{ ml} \times N_2$
 $N_2 = 6,888 \text{ ppm}$
- 4) $V_1 \times N_1 = V_2 \times N_2$
 $1,6 \text{ ml} \times 49,2 \text{ ppm} = 10 \text{ ml} \times N_2$
 $N_2 = 7,872 \text{ ppm}$
- 5) $V_1 \times N_1 = V_2 \times N_2$
 $1,8 \text{ ml} \times 49,2 \text{ ppm} = 10 \text{ ml} \times N_2$
 $N_2 = 8,856 \text{ ppm}$
- 6) $V_1 \times N_1 = V_2 \times N_2$
 $2 \text{ ml} \times 49,2 \text{ ppm} = 10 \text{ ml} \times N_2$
 $N_2 = 9,84 \text{ ppm}$

Konsentrasi (ppm)	Absorbansi
4,920	0,315
5,904	0,427
6,888	0,527
7,872	0,646
8,856	0,741
9,840	0,860

Persamaan regresi linier antara konsentrasi (ppm) dan serapan diperoleh nilai :

$$a = -0,225285714$$

$$b = 0,109930314$$

$$r = 0,999669338$$

$$y = a + bx$$

$$y = -0,225285714 + 0,109930314x$$

keterangan :

x = konsentrasi ($\mu\text{g/ml}$)

y = serapan

Hasil linearitas diperoleh $R = 0.999669338$; sehingga dapat disimpulkan bahwa data tersebut linier.

c. Akurasi

X (ppm)	Y (abs)	Konsentrasi	Replikasi	Abs	Konsentrasi	Konsentrasi Sebenarnya	%	Rata-rata	Hasil
4,920	0,315	80%	1	0,326	5,0149	4,92	102%	99,89%	99,95%
5,904	0,427		2	0,304	4,8147	4,92	98%		
6,888	0,527		3	0,315	4,9148	4,92	100%		
7,872	0,646	100%	1	0,431	5,9700	5,904	101%	100,86%	
8,856	0,741		2	0,425	5,9154	5,904	100%		
9,840	0,860		3	0,432	5,9791	5,904	101%		
		120%	1	0,525	6,8251	6,888	99%	99,09%	
			2	0,527	6,8433	6,888	99%		
			3	0,523	6,8069	6,888	99%		

d. Presisi

Konsentrasi (ppm)	Absorbansi
6,888	0,552
6,888	0,537
6,888	0,545
6,888	0,557
6,888	0,558
6,888	0,525
6,888	0,543
6,888	0,547
6,888	0,552
6,888	0,529
Rata-rata	7,0025
SD	0,102222
CV	0,014598 %

Nilai CV dilihat dari data diatas adalah 0,014598%, hasil ini sesuai dengan persyaratan presisi yaitu $\leq 2\%$.

Lampiran 6. Foto NLC Fisetin



Lampiran 7. Foto NLC fisetin setelah disimpan selama 2 minggu



Lampiran 8. Uji ukuran partikel formula 1 (Precirol 2%)

a. Replikasi I

Size Distribution Report by Intensity v2.2



Sample Details

Sample Name: NLC 2% RIZKY 1
SOP Name: mansettings.nano
General Notes: 2 tetes ke 5 ml

File Name: Rizky_120320.dts Dispersant Name: Water
Record Number: 1 Dispersant RI: 1,330
Material RI: 1,52 Viscosity (cP): 0,8872
Material Absorbtion: 0,100 Measurement Date and Time: 12 March 2020 09:57:42

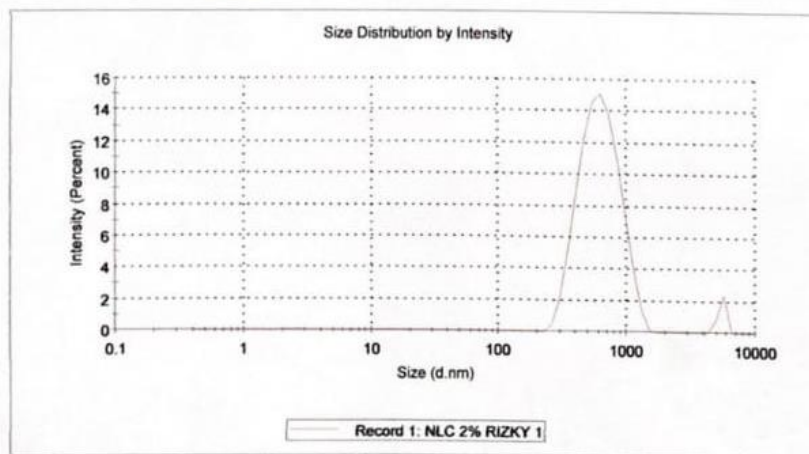
System

Temperature (°C): 25,0 Duration Used (s): 60
Count Rate (kcps): 227,4 Measurement Position (mm): 4,65
Cell Description: Disposable sizing cuvette Attenuator: 5

Results

	Size (d.n...	% Intensity:	St Dev (d.n...
Z-Average (d.nm): 658,8	Peak 1: 634,9	96,9	219,3
Pdl: 0,315	Peak 2: 5376	3,1	324,8
Intercept: 0,949	Peak 3: 0,000	0,0	0,000

Result quality **Good**



b. Replikasi II

Size Distribution Report by Intensity

v2.2



Sample Details

Sample Name: NLC 2% RIZKY 2
 SOP Name: mansettings.nano
 General Notes: 2 tetes ke 5 ml

File Name: Rizky_120320.dts Dispersant Name: Water
 Record Number: 2 Dispersant RI: 1,330
 Material RI: 1,52 Viscosity (cP): 0,8872
 Material Absorbion: 0,100 Measurement Date and Time: 12 March 2020 09:59:55

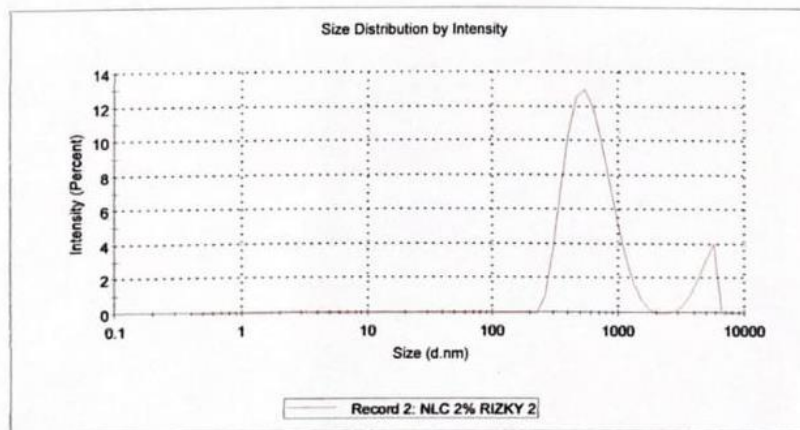
System

Temperature (°C): 25,0 Duration Used (s): 60
 Count Rate (kcps): 257,3 Measurement Position (mm): 4,65
 Cell Description: Disposable sizing cuvette Attenuator: 5

Results

	Size (d.n...	% Intensity:	St Dev (d.n...
Z-Average (d.nm): 649,6	Peak 1: 606,2	89,6	246,8
PdI: 0,350	Peak 2: 4788	10,4	755,6
Intercept: 0,942	Peak 3: 0,000	0,0	0,000

Result quality Good



c. Replikasi III

Size Distribution Report by Intensity

v2.2



Sample Details

Sample Name: NLC 2% RIZKY 3

SOP Name: mansettings.nano

General Notes: 2 tetes ke 5 ml

File Name: Rizky_120320.dts

Dispersant Name: Water

Record Number: 3

Dispersant RI: 1,330

Material RI: 1,52

Viscosity (cP): 0,8872

Material Absorbtion: 0,100

Measurement Date and Time: 12 March 2020 10:02:09

System

Temperature (°C): 25,0

Duration Used (s): 60

Count Rate (kcps): 216,1

Measurement Position (mm): 4,65

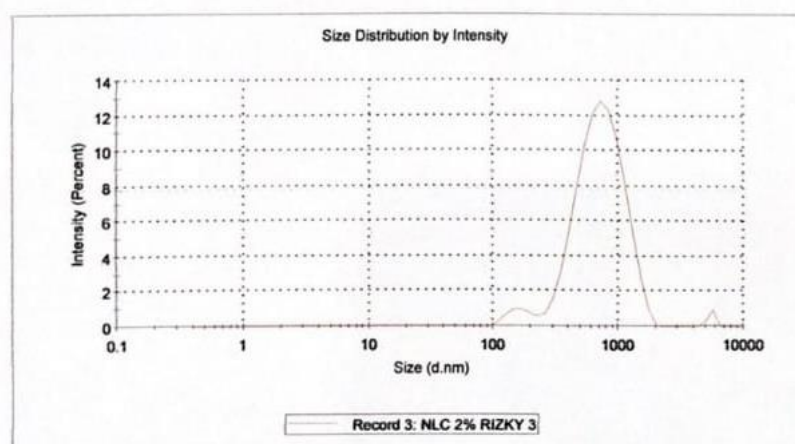
Cell Description: Disposable sizing cuvette

Attenuator: 5

Results

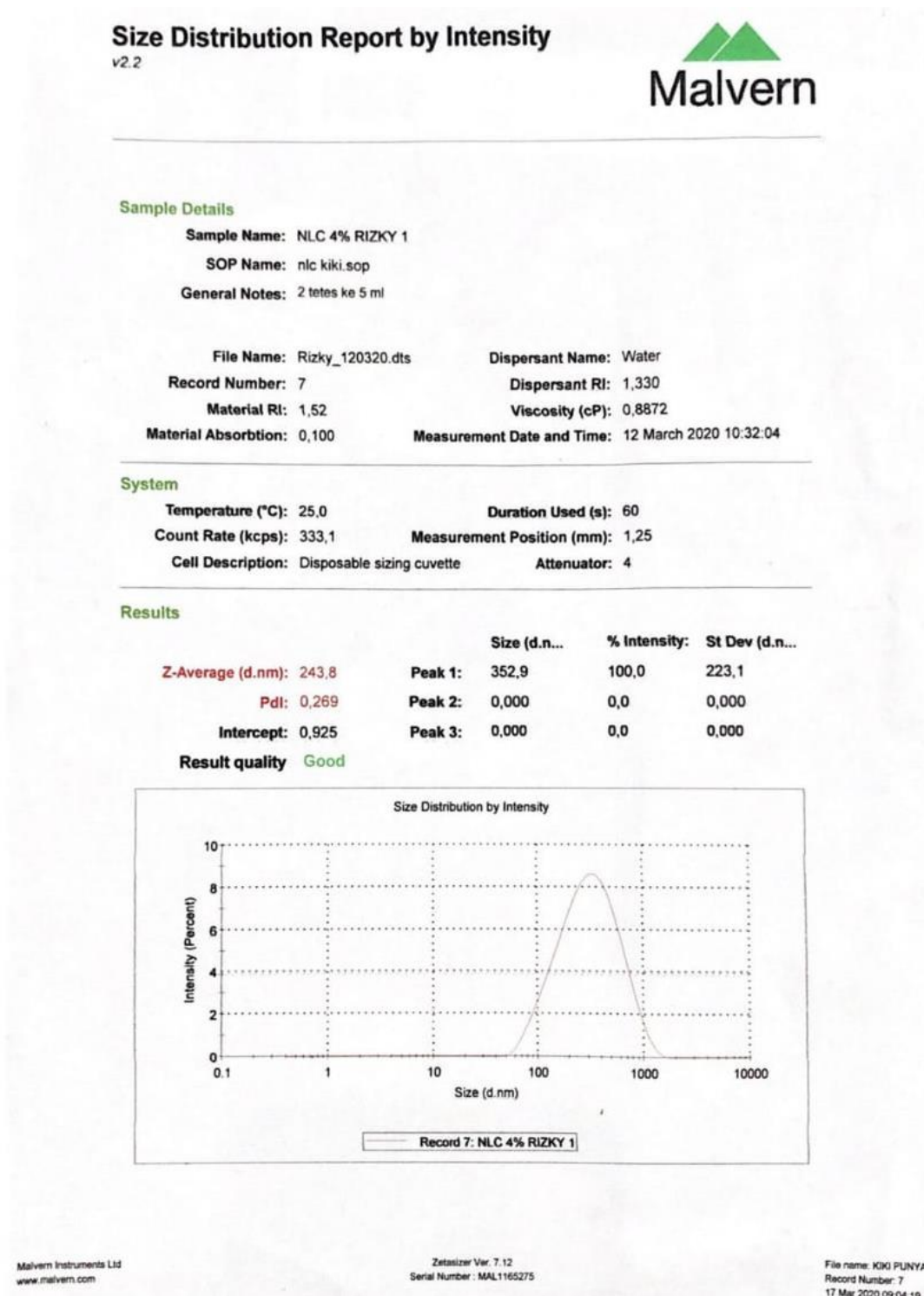
	Size (d.n...	% Intensity:	St Dev (d.n...
Z-Average (d.nm): 644,8	Peak 1: 756,1	94,5	305,6
Pdl: 0,279	Peak 2: 161,7	4,3	33,65
Intercept: 0,945	Peak 3: 5407	1,2	304,5

Result quality **Good**

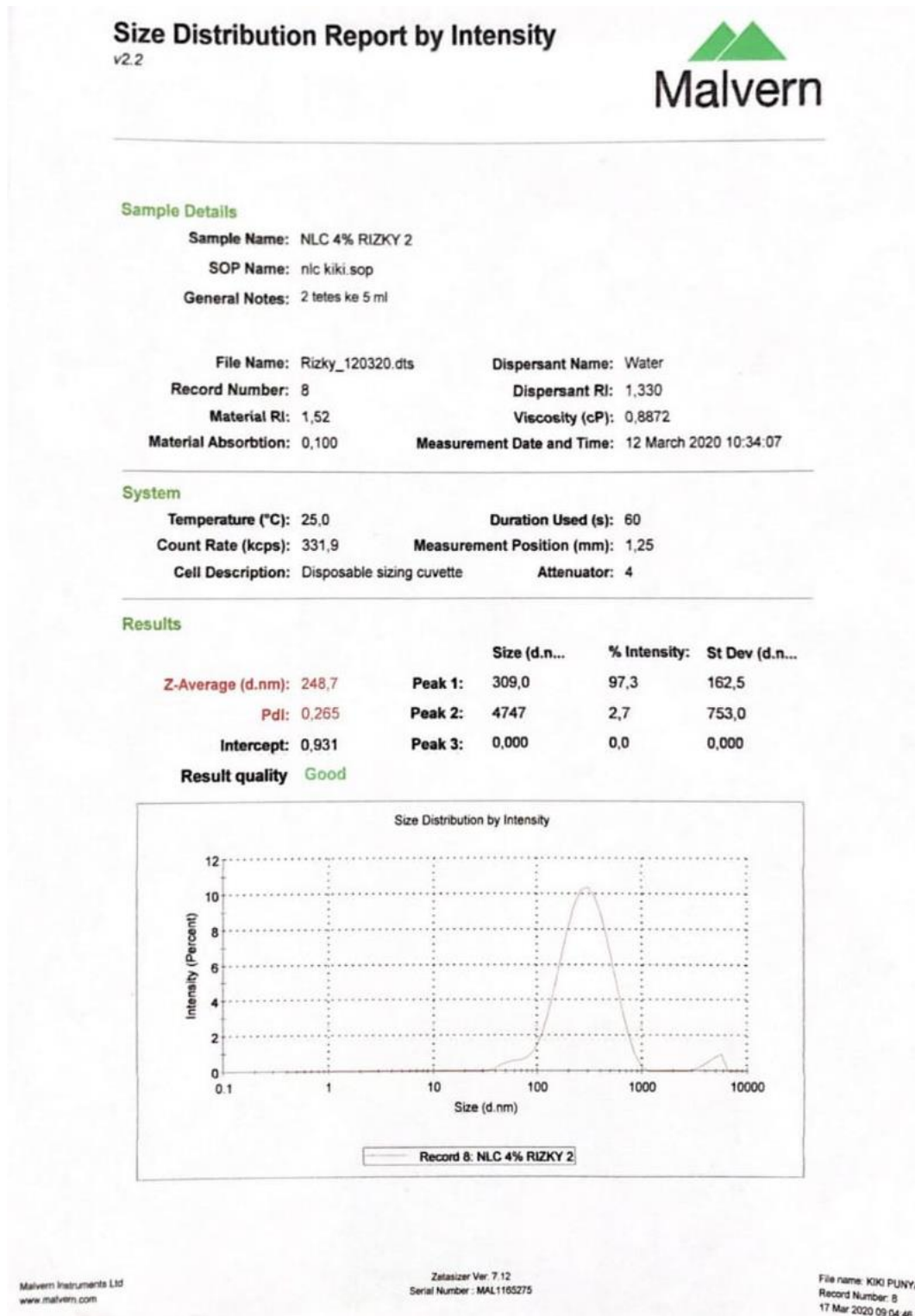


Lampiran 9. Uji ukuran partikel formula 2 (Precirol 4%)

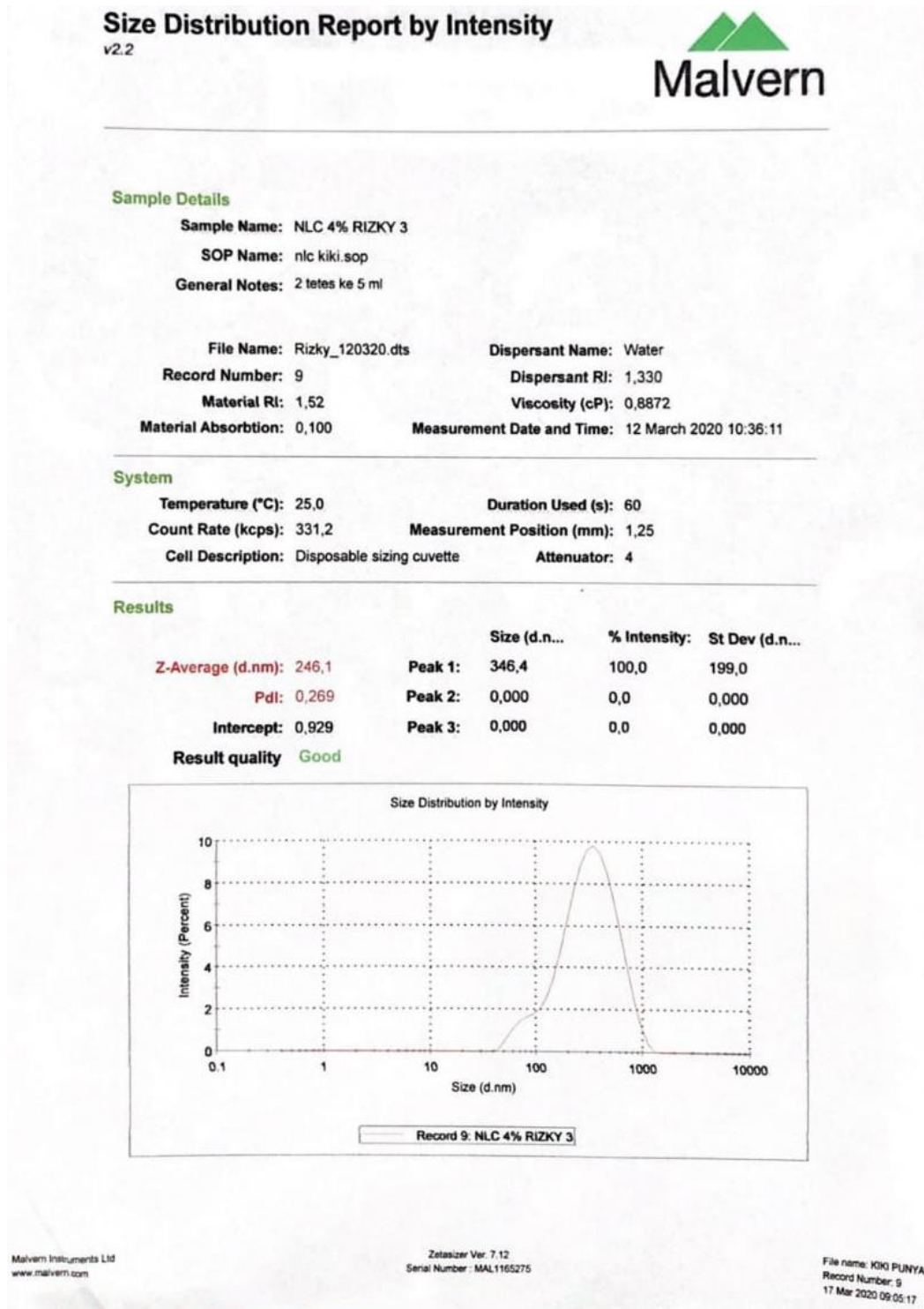
a. Replikasi I



b. Replikasi II



c. Replikasi III



Lampiran 10. Uji ukuran partikel formula 3 (Precirol 6%)

a. Replikasi I

Size Distribution Report by Intensity v2.2



Sample Details

Sample Name: NLC 6% RIZKY 1

SOP Name: nlc kiki.sop

General Notes: 2 tetes ke 5 ml

File Name: Rizky_120320.dts

Dispersant Name: Water

Record Number: 13

Dispersant RI: 1,330

Material RI: 1,52

Viscosity (cP): 0,8872

Material Absorbion: 0,100

Measurement Date and Time: 12 March 2020 11:00:24

System

Temperature (°C): 25,0

Duration Used (s): 60

Count Rate (kcps): 266,1

Measurement Position (mm): 4,65

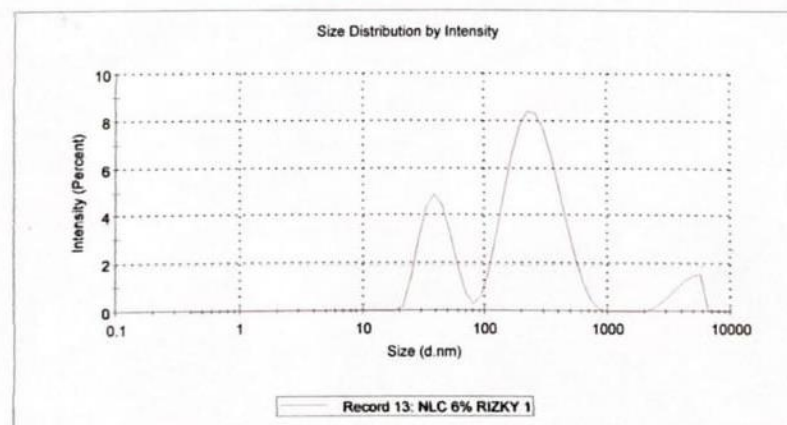
Cell Description: Disposable sizing cuvette

Attenuator: 5

Results

	Size (d.n...	% Intensity:	St Dev (d.n...
Z-Average (d.nm): 121,5	Peak 1: 267,6	69,1	124,1
Pdl: 0,575	Peak 2: 40,66	24,4	11,50
Intercept: 0,948	Peak 3: 4309	6,5	965,2

Result quality Refer to quality report



b. Replikasi II

Size Distribution Report by Intensity
v2.2

Sample Details

Sample Name: NLC 6% RIZKY 2

SOP Name: nlc kiki.sop

General Notes: 2 tetes ke 5 ml

File Name: Rizky_120320.dts

Dispersant Name: Water

Record Number: 14

Dispersant RI: 1,330

Material RI: 1,52

Viscosity (cP): 0,8872

Material Absorbion: 0,100

Measurement Date and Time: 12 March 2020 11:02:37

System

Temperature (°C): 25,0

Duration Used (s): 60

Count Rate (kcps): 263,4

Measurement Position (mm): 4,65

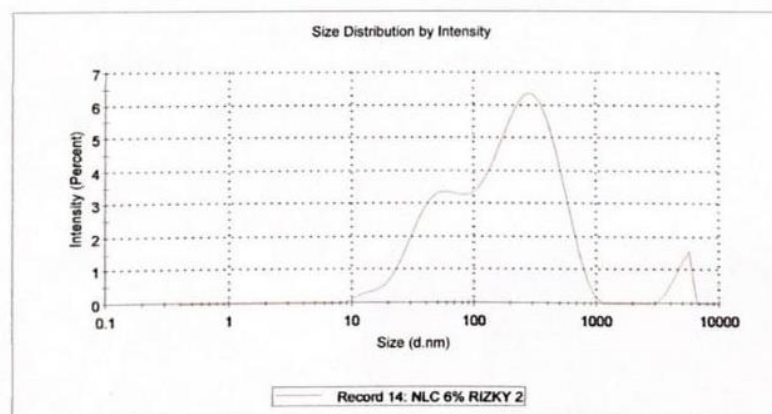
Cell Description: Disposable sizing cuvette

Attenuator: 5

Results

	Size (d.n...	% Intensity:	St Dev (d.n...
Z-Average (d.nm): 123,7	Peak 1: 282,8	69,7	167,1
Pdl: 0,515	Peak 2: 47,52	26,5	18,87
Intercept: 0,947	Peak 3: 4853	3,8	700,8

Result quality Refer to quality report



c. Replikasi III

Size Distribution Report by Intensity

v2.2



Sample Details

Sample Name: NLC 6% RIZKY 3

SOP Name: nlc kiki.sop

General Notes: 2 tetes ke 5 ml

File Name: Rizky_120320.dts

Dispersant Name: Water

Record Number: 15

Dispersant RI: 1,330

Material RI: 1,52

Viscosity (cP): 0,8872

Material Absorbion: 0,100

Measurement Date and Time: 12 March 2020 11:04:51

System

Temperature (°C): 25,0

Duration Used (s): 60

Count Rate (kcps): 261,9

Measurement Position (mm): 4,65

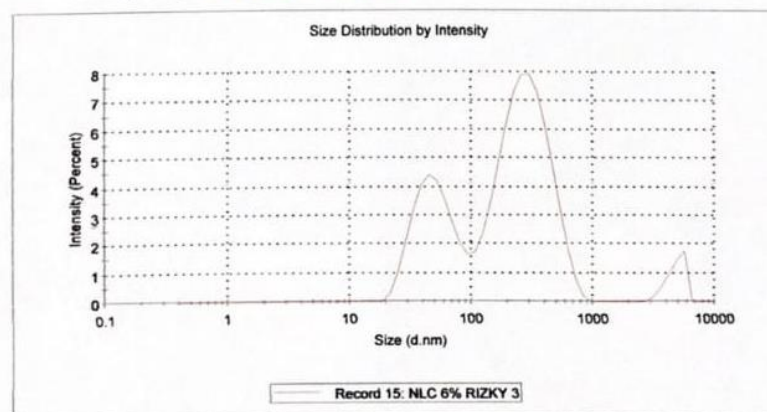
Cell Description: Disposable sizing cuvette

Attenuator: 5

Results

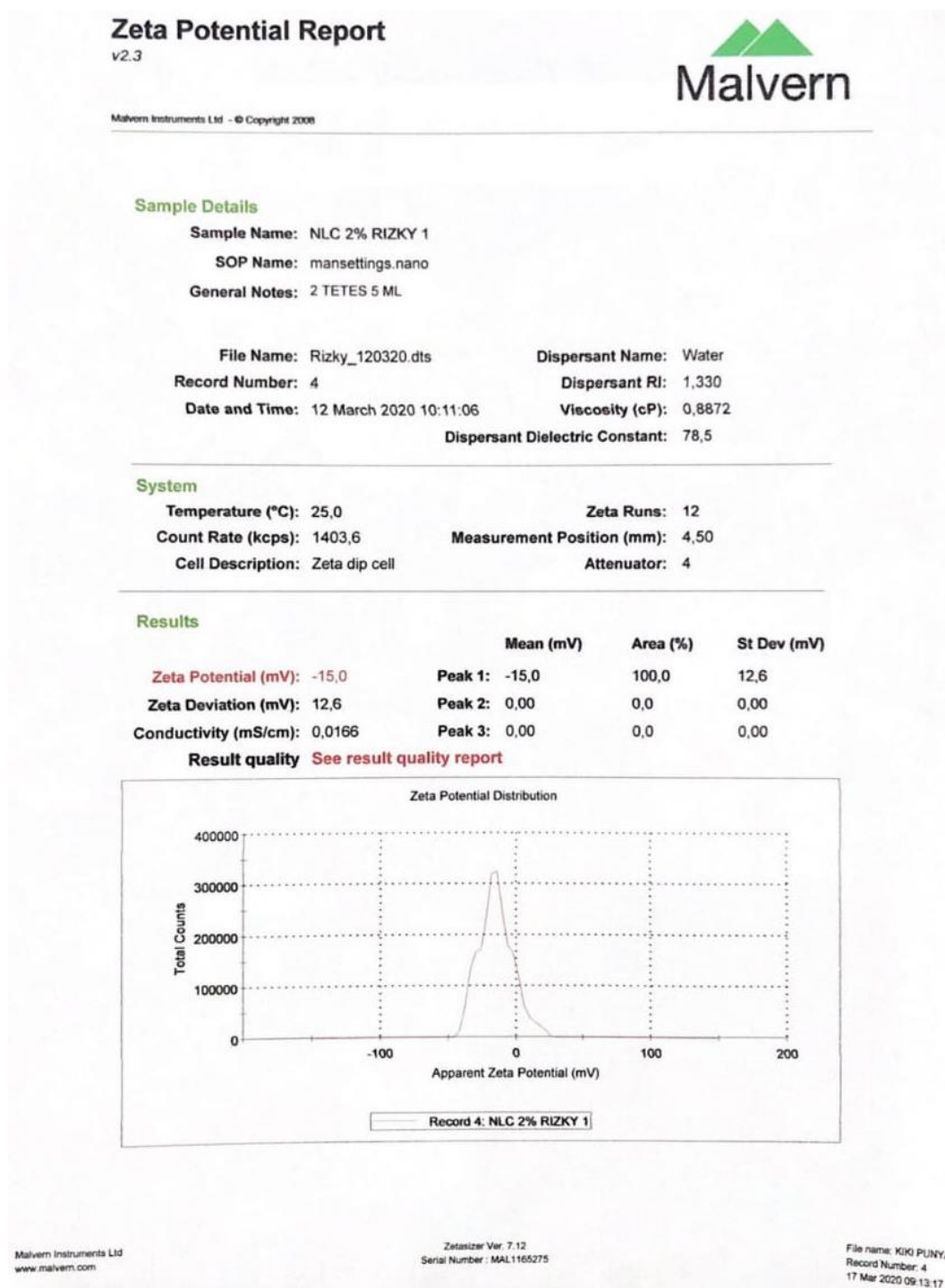
	Size (d.n...	% Intensity:	St Dev (d.n...
Z-Average (d.nm): 113,5	Peak 1: 291,6	66,4	135,8
PdI: 0,768	Peak 2: 50,06	28,8	17,79
Intercept: 0,948	Peak 3: 4749	4,8	753,0

Result quality Refer to quality report

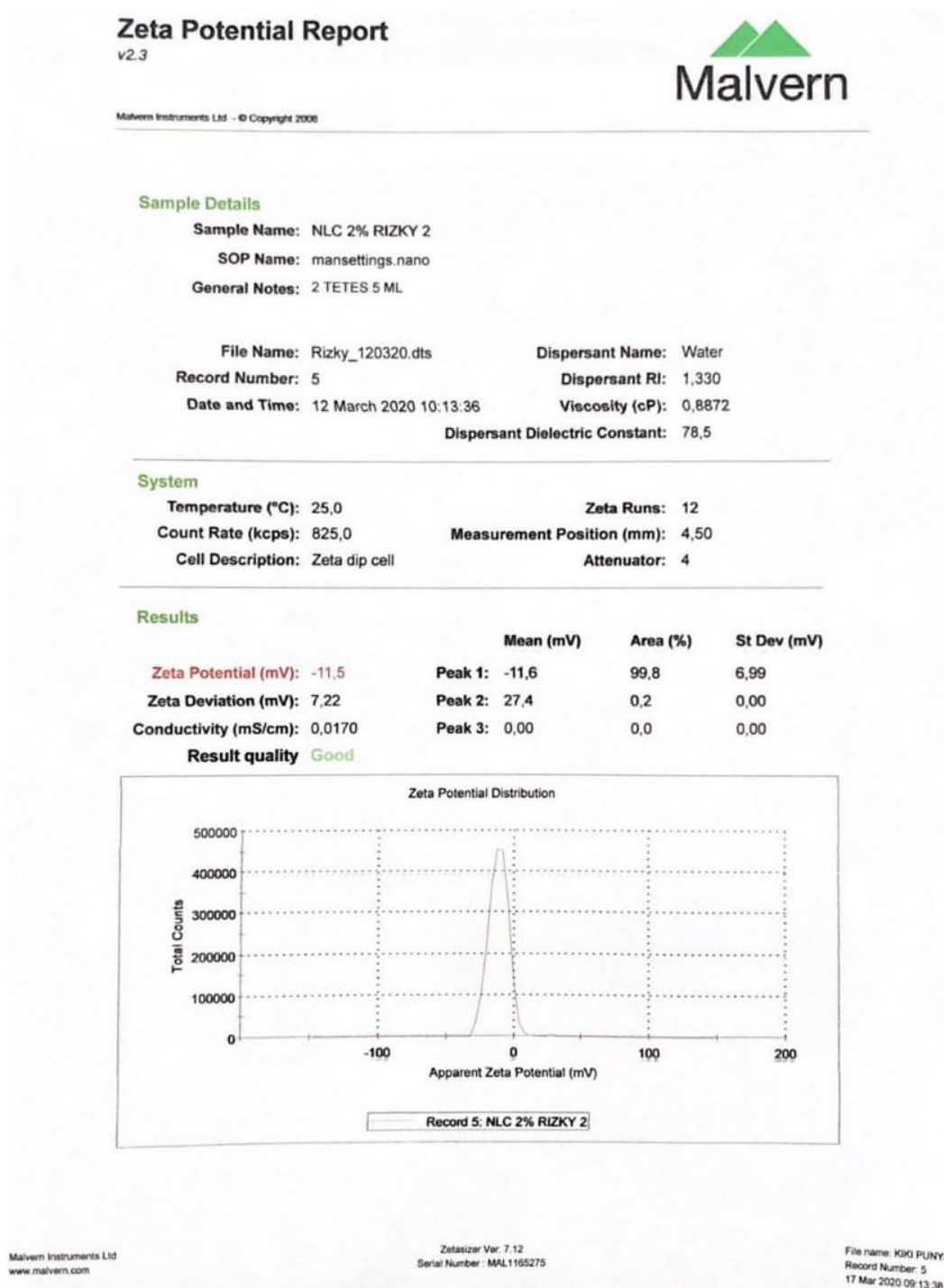


Lampiran 11. Uji zeta potensial formula I (Precirol 2%)

a. Replikasi I



b. Replikasi II



c. Replikasi III

Zeta Potential Report

v2.3



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Sample Details

Sample Name: NLC 2% RIZKY 3

SOP Name: mansettings.nano

General Notes: 2 TETES 5 ML

File Name: Rizky_120320.dts

Dispersant Name: Water

Record Number: 6

Dispersant RI: 1,330

Date and Time: 12 March 2020 10:14:16

Viscosity (cP): 0,8872

Dispersant Dielectric Constant: 78,5

System

Temperature (°C): 25,0

Zeta Runs: 12

Count Rate (kcps): 794,9

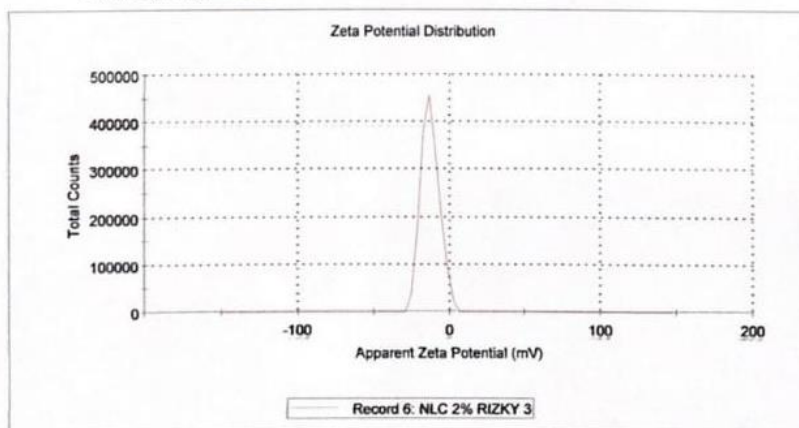
Measurement Position (mm): 4,50

Cell Description: Zeta dip cell

Attenuator: 4

Results

	Mean (mV)	Area (%)	St Dev (mV)
Zeta Potential (mV): -12,9	Peak 1: -12,9	100,0	5,90
Zeta Deviation (mV): 5,90	Peak 2: 0,00	0,0	0,00
Conductivity (mS/cm): 0,0172	Peak 3: 0,00	0,0	0,00
Result quality Good			



Lampiran 12. Uji zeta potensial formula 2 (Precirol 4%)

a. Replikasi I



b. Replikasi II

Zeta Potential Report

v2.3



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Sample Details

Sample Name: NLC 4% RIZKY 2

SOP Name: ZETA KIKI.sop

General Notes: 2 TETES 5 ML

File Name: Rizky_120320.dts

Dispersant Name: Water

Record Number: 11

Dispersant RI: 1,330

Date and Time: 12 March 2020 10:47:09

Viscosity (cP): 0,8872

Dispersant Dielectric Constant: 78,5

System

Temperature (°C): 25,0

Zeta Runs: 12

Count Rate (kcps): 534,5

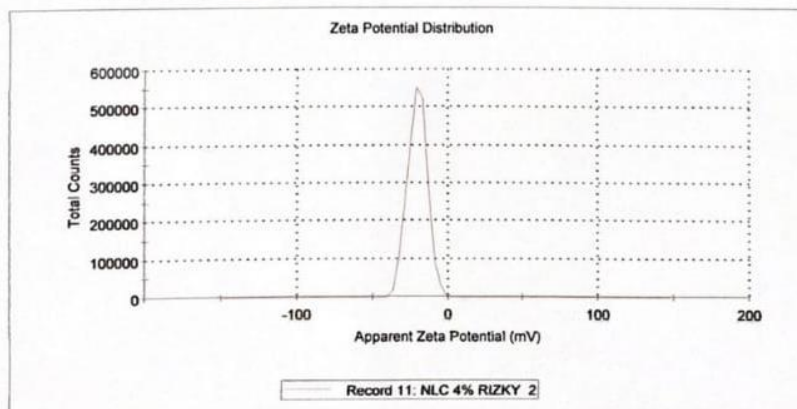
Measurement Position (mm): 4,50

Cell Description: Zeta dip cell

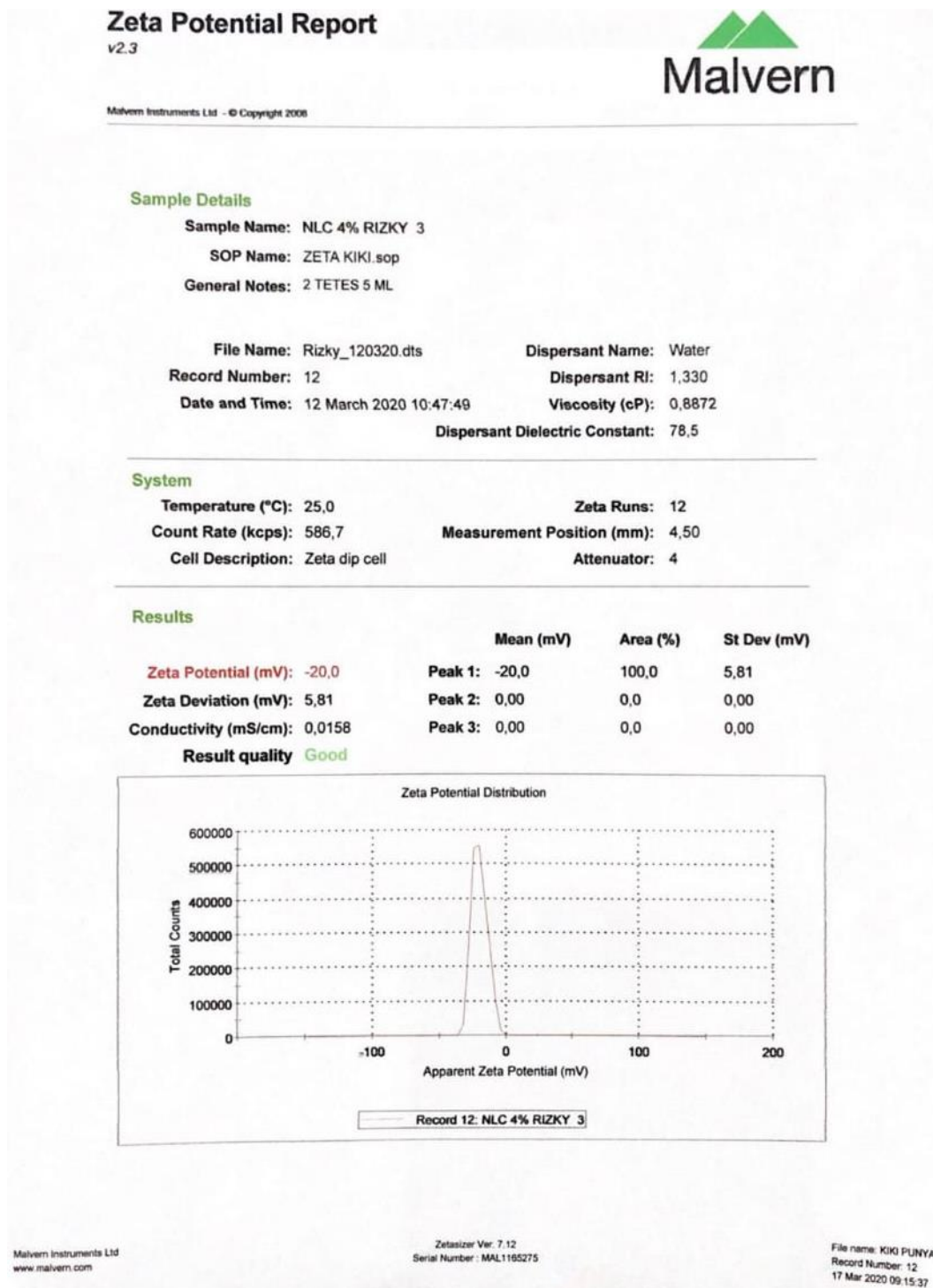
Attenuator: 4

Results

	Mean (mV)	Area (%)	St Dev (mV)
Zeta Potential (mV): -20,6	Peak 1: -20,6	100,0	6,31
Zeta Deviation (mV): 6,31	Peak 2: 0,00	0,0	0,00
Conductivity (mS/cm): 0,0156	Peak 3: 0,00	0,0	0,00
Result quality Good			

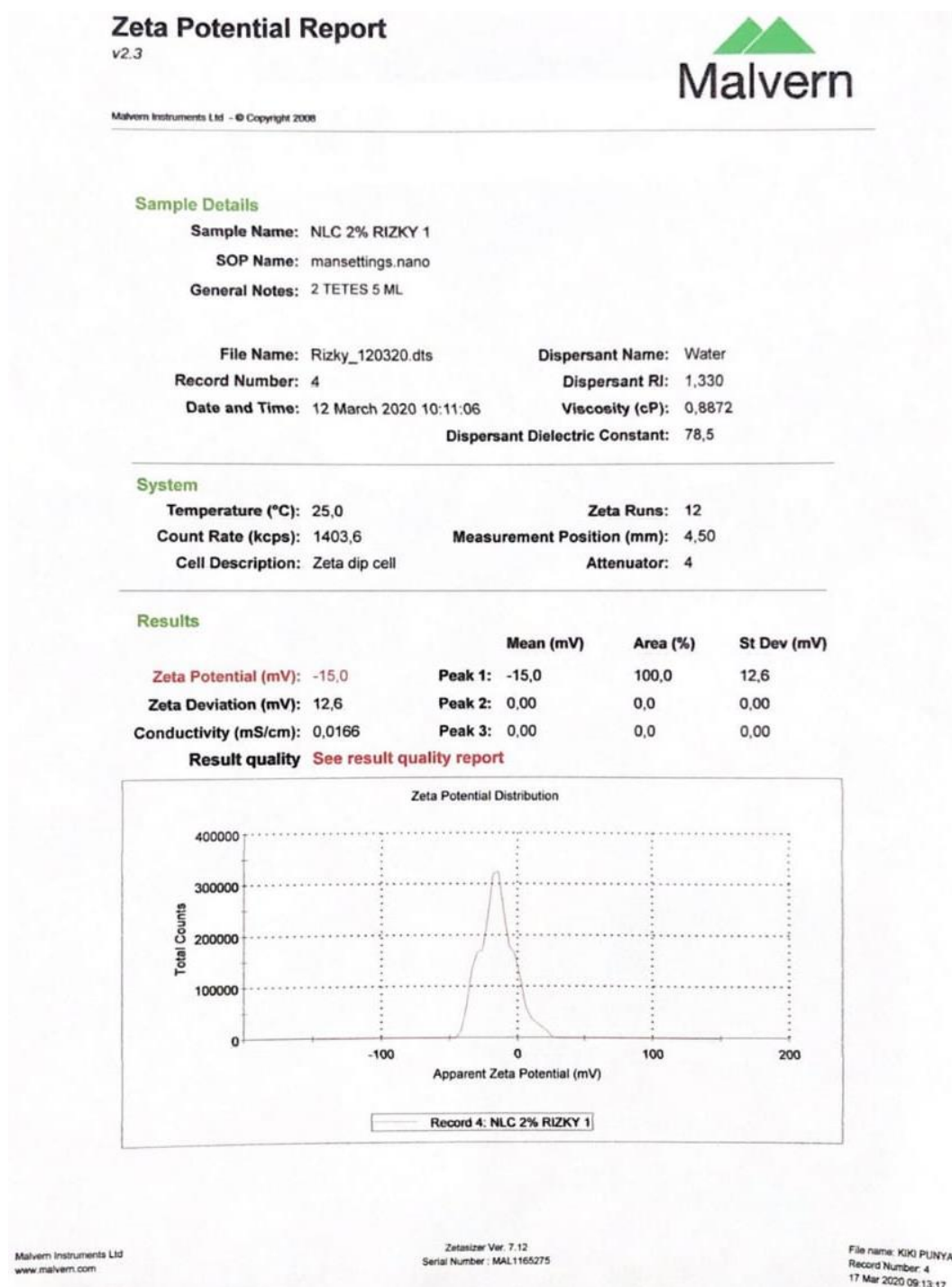


c. Replikasi III



Lampiran 13. Uji zeta potensial formula 3 (Precirol 6%)

a. Replikasi I



b. Replikasi II

Zeta Potential Report
v2.3

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Sample Details

Sample Name: NLC 6% RIZKY 2

SOP Name: ZETA KIKI.sop

General Notes: 2 TETES 5 ML

File Name: Rizky_120320.dts

Dispersant Name: Water

Record Number: 17

Dispersant RI: 1,330

Date and Time: 12 March 2020 11:14:09

Viscosity (cP): 0,8872

Dispersant Dielectric Constant: 78,5

System

Temperature (°C): 25,0

Zeta Runs: 12

Count Rate (kcps): 271,6

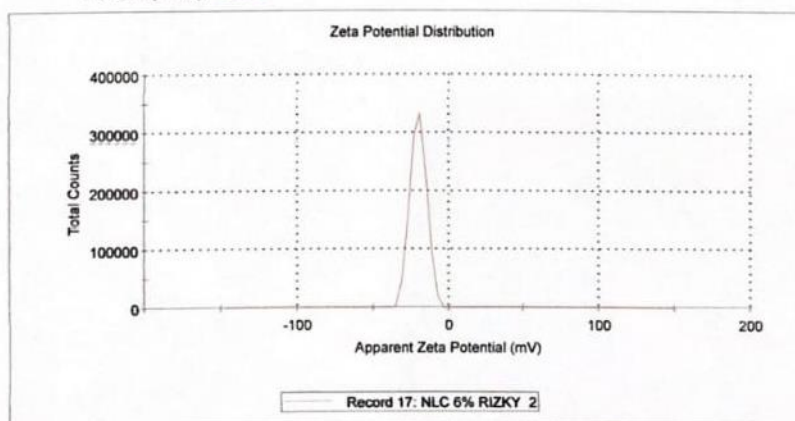
Measurement Position (mm): 4,50

Cell Description: Zeta dip cell

Attenuator: 4

Results

	Mean (mV)	Area (%)	St Dev (mV)
Zeta Potential (mV): -20,2	Peak 1: -20,2	100,0	5,33
Zeta Deviation (mV): 5,33	Peak 2: 0,00	0,0	0,00
Conductivity (mS/cm): 0,0213	Peak 3: 0,00	0,0	0,00
Result quality Good			



c. Replikasi III

Zeta Potential Report

v2.3



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Sample Details

Sample Name: NLC 6% RIZKY 3

SOP Name: ZETA KIKI.sop

General Notes: 2 TETES 5 ML

File Name: Rizky_120320.dts

Dispersant Name: Water

Record Number: 18

Dispersant RI: 1,330

Date and Time: 12 March 2020 11:14:49

Viscosity (cP): 0,8872

Dispersant Dielectric Constant: 78,5

System

Temperature (°C): 25,0

Zeta Runs: 12

Count Rate (kcps): 388,3

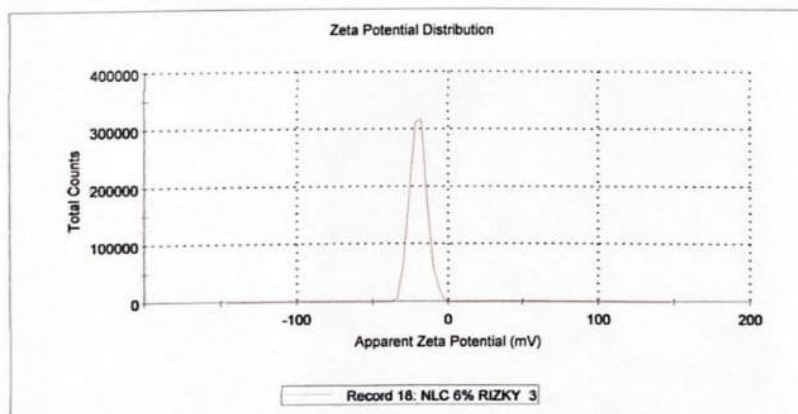
Measurement Position (mm): 4,50

Cell Description: Zeta dip cell

Attenuator: 4

Results

	Mean (mV)	Area (%)	St Dev (mV)
Zeta Potential (mV): -20,0	Peak 1: -20,0	100,0	5,44
Zeta Deviation (mV): 5,44	Peak 2: 0,00	0,0	0,00
Conductivity (mS/cm): 0,0215	Peak 3: 0,00	0,0	0,00
Result quality Good			



Lampiran 14. Foto serum NLC fisetin baru dibuat.



Lampiran 15. Foto serum NLC fisetin setelah 30 hari



Lampiran 16. Data skunder efisiensi penyerapan.

a. Rancangan Formula NLC

Table 3. Composition of modified diflucortolone valerate in Precirol®-based nanostructured lipid carriers using lipid-based surfactants.

Formula ^a	Total lipid (20% w/w/formulation)				Poloxamer [®] 407 concentration (% w/w/formulation)	Lipid based surfactant	
	Solid lipid		Liquid lipid			Type	Concentration (% w/w/formulation)
	Type	Concentration (% w/w/base)	Type	Concentration (% w/w/base)			
NLCM1	Precirol [®]	70	Capryol™	30	10	Labrasol [®]	5
NLCM2		70		30	10		10
NLCM3		70		30	10	Labrafil [®]	5
NLCM4		70		30	10		10
NLCM5	Precirol [®]	50	Capryol™	50	10	Labrasol [®]	5
NLCM6		50		50	10		10
NLCM7		50		50	10	Labrafil [®]	5
NLCM8		50		50	10		10
NLCM9	Precirol [®]	70	IPM	30	10	Labrasol [®]	5
NLCM10		70		30	10		10
NLCM11		70		30	10	Labrafil [®]	5
NLCM12		70		30	10		10
NLCM13	Precirol [®]	50	IPM	50	10	Labrasol [®]	5
NLCM14		50		50	10		10
NLCM15		50		50	10	Labrafil [®]	5
NLCM16		50		50	10		10
NLCM17	Tristearin [®]	70	Capryol™	30	10	Labrasol [®]	5
NLCM18		70		30	10		10
NLCM19		70		30	10	Labrafil [®]	5
NLCM20		70		30	10		10
NLCM21	Tristearin [®]	50	Capryol™	50	10	Labrasol [®]	5
NLCM22		50		50	10		10
NLCM23		50		50	10	Labrafil [®]	5
NLCM24		50		50	10		10
NLCM25	Tristearin [®]	70	IPM	30	10	Labrasol [®]	5
NLCM26		70		30	10		10
NLCM27		70		30	10	Labrafil [®]	5
NLCM28		70		30	10		10
NLCM29	Tristearin [®]	50	IPM	50	10	Labrasol [®]	5
NLCM30		50		50	10		10
NLCM31		50		50	10	Labrafil [®]	5
NLCM32		50		50	10		10

^aFormulation weight was made to 100% with distilled water and contained 0.1% w/w of diflucortolone valerate.

(Abdel-Salam, Mahmoud, Ammar & Elkheshen 2016)

b. Hasil uji ukuran partikel, PDI dan efisiensi penjerapan

Table 4. Mean particle size, polydispersity index, and entrapment efficiency values of modified nanostructured lipid carriers.

Formulation	Mean particle size (nm) $\pm$ SD	PDI $\pm$ SD	EE% $\pm$ SD
NLCM1	439.1 $\pm$ 24.9	0.44 $\pm$ 0.09	41.12 $\pm$ 4.90
NLCM2	490.1 $\pm$ 21.5	0.44 $\pm$ 0.06	43.06 $\pm$ 4.50
NLCM3	229.0 $\pm$ 25.4	0.23 $\pm$ 0.02	44.82 $\pm$ 3.52
NLCM4	245.1 $\pm$ 14.5	0.28 $\pm$ 0.08	68.00 $\pm$ 7.40
NLCM5	206.1 $\pm$ 20.5	0.24 $\pm$ 0.12	42.06 $\pm$ 5.80
NLCM6	743.7 $\pm$ 20.4	0.52 $\pm$ 0.03	42.02 $\pm$ 4.78
NLCM7	246.3 $\pm$ 17.2	0.14 $\pm$ 0.05	50.20 $\pm$ 5.65
NLCM8	197.7 $\pm$ 7.6	0.15 $\pm$ 0.06	59.59 $\pm$ 5.20
NLCM9	230.9 $\pm$ 15.2	0.39 $\pm$ 0.13	37.38 $\pm$ 5.62
NLCM10	589.2 $\pm$ 22.6	0.52 $\pm$ 0.11	38.38 $\pm$ 4.66
NLCM11	264.1 $\pm$ 33.4	0.24 $\pm$ 0.01	44.56 $\pm$ 5.70
NLCM12	244.5 $\pm$ 30.1	0.25 $\pm$ 0.03	53.18 $\pm$ 2.94
NLCM13	187.5 $\pm$ 11.5	0.22 $\pm$ 0.02	40.28 $\pm$ 3.86
NLCM14	337.2 $\pm$ 21.5	0.53 $\pm$ 0.09	41.77 $\pm$ 3.96
NLCM15	183.5 $\pm$ 14.3	0.23 $\pm$ 0.04	36.18 $\pm$ 5.02
NLCM16	180.8 $\pm$ 18.2	0.37 $\pm$ 0.14	55.61 $\pm$ 8.65
NLCM17	ND	ND	ND
NLCM18	ND	ND	ND
NLCM19	295.0 $\pm$ 13.7	0.23 $\pm$ 0.02	51.34 $\pm$ 4.00
NLCM20	255.1 $\pm$ 11.0	0.23 $\pm$ 0.01	56.95 $\pm$ 6.60
NLCM21	ND	ND	ND
NLCM22	ND	ND	ND
NLCM23	276.9 $\pm$ 19.8	0.27 $\pm$ 0.03	42.29 $\pm$ 3.70
NLCM24	219.7 $\pm$ 6.5	0.15 $\pm$ 0.01	63.90 $\pm$ 4.30
NLCM25	252.6 $\pm$ 25.7	0.24 $\pm$ 0.05	40.75 $\pm$ 5.40
NLCM26	231.3 $\pm$ 20.5	0.25 $\pm$ 0.01	48.25 $\pm$ 1.93
NLCM27	229.5 $\pm$ 9.3	0.25 $\pm$ 0.02	ND
NLCM28	217.7 $\pm$ 8.0	0.28 $\pm$ 0.02	64.59 $\pm$ 1.09
NLCM29	ND	ND	ND
NLCM30	ND	ND	ND
NLCM31	ND	ND	ND

ND, not determined.

(Abdel-Salam, Mahmoud, Ammar & Elkheshen 2016)

c. Hasil uji efisiensi penjerapan (Apriyani 2019).

Larutan induk $\rightarrow$ 200 mg NLC Fisetin/10 ml etanol p.a = 20.000 ppm

Perhitungan teoritis Fisetin = 10 mg

Eksipien (tween 80 + IPM + GMS) = 11.500 mg %

Kadar fisetin = $\frac{10}{11.500+10} \times 100\% = 0,0869\%$

Kadar dalam 200 mg NLC = $0,0869\% \times 200\text{mg} = 0,1738 \text{ mg}$

Perhitungan kadar fisetin terjerap menggunakan persamaan regresi linier

: y = a + bx

0,449 = 0,014 + ,00636x

0,0636x = 0,435

x = 6,840 ppm

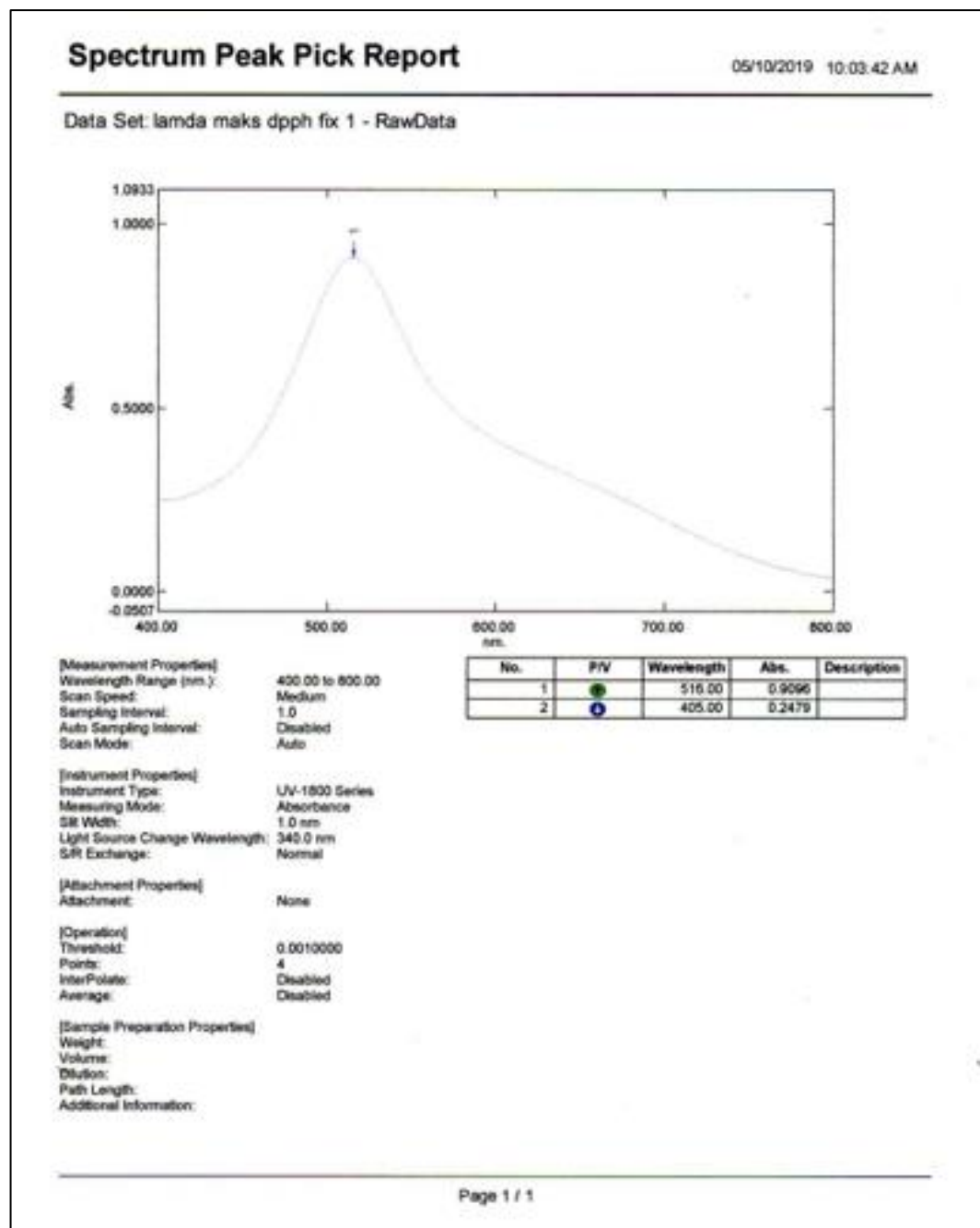
% kadar = $\frac{6840}{20000 \text{ ppm}} \times 100\% = 0,0324\%$

Kadar dalam 200mg NLC fisetin = $0,0324\% \times 200 \text{ mg} = 0,0648 \text{ mg}$

% Efisiensi penjerapan (EE) = $\frac{\text{kadar terjerap}}{\text{kadar teoritis}} \times 100\%$
 $= \frac{0,0648}{0,174} \times 100\%$
 $= 59,20\%$

Lampiran 17. Uji Aktivitas Antioksidan (Apriyani D 2019).

a. Penentuan Lamda max.



b. Penentuan Operating Time

Kinetics Data Print Report

OT Finitio DPPH 4/5/19
60 MEHIT

05/04/2019 12:24:22 PM

Time (Minute)	RawData ...
0.000	0.199
1.000	0.199
2.000	0.198
3.000	0.198
4.000	0.199
5.000	0.198
6.000	0.198
7.000	0.198
8.000	0.198
9.000	0.199
10.000	0.198
11.000	0.199
12.000	0.198
13.000	0.199
14.000	0.198
15.000	0.199
16.000	0.199
17.000	0.199
18.000	0.199
19.000	0.199
20.000	0.199
21.000	0.199
22.000	0.199
23.000	0.199
24.000	0.199
25.000	0.199
26.000	0.199
27.000	0.199
28.000	0.199
29.000	0.199
30.000	0.199
31.000	0.199
32.000	0.199
33.000	0.199
34.000	0.200
35.000	0.200
36.000	0.199
37.000	0.200
38.000	0.200
39.000	0.200
40.000	0.200
41.000	0.200
42.000	0.200
43.000	0.200
44.000	0.200
45.000	0.200
46.000	0.200
47.000	0.200
48.000	0.200
49.000	0.200
50.000	0.200

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Kinetics Data Print Report

05/04/2019 12:24:22 PM

Time (Minute)	RawData ...
51.000	0.201
52.000	0.200
53.000	0.200
54.000	0.201
55.000	0.201
56.000	0.201
57.000	0.201
58.000	0.201
59.000	0.201
60.000	0.201

Lampiran 18. DPPH Fisetin (Apriyani 2019).

Perhitungan bahan fisetin : $50\text{mg}/100\text{ml} = 500\text{mg}/1000\text{ml} = 500 \text{ ppm}$

Perhitungan konsentrasi (ppm) :

- 1) $V_1 \times N_1 = V_2 \times N_2$
 $500 \text{ ppm} \times 0,3112 \text{ ml} = 10\text{ml} \times N_2$
 $N_2 = 15,56 \text{ ppm}$
- 2) $V_1 \times N_1 = V_2 \times N_2$
 $500 \text{ ppm} \times 0,1556 \text{ ml} = 10 \text{ ml} \times N_2$
 $N_2 = 7,78 \text{ ppm}$
- 3) $V_1 \times N_1 = V_2 \times N_2$
 $500 \text{ ppm} \times 0,0778 \text{ ml} = 10\text{ml} \times N_2$
 $N_2 = 3,89 \text{ ppm}$
- 4) $V_1 \times N_1 = V_2 \times N_2$
 $500 \text{ ppm} \times 0,039 \text{ ml} = 10\text{ml} \times N_2$
 $N_2 = 1,95 \text{ ppm}$
- 5) $V_1 \times N_1 = V_2 \times N_2$
 $500 \text{ ppm} \times 0,0194 \text{ ml} = 10\text{ml} \times N_2$
 $N_2 = 0,97 \text{ ppm}$

a. Replikasi I

Konsentrasi (ppm)	Absorbansi	% Inhibisi
15,56	0,212	76,7
7,78	0,405	55,5
3,89	0,507	44,3
1,95	0,555	39,0
0,97	0,578	36,5

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 33,6727$$

$$b = 2,7696$$

$$r = 0,99994$$

$$y = a + bx \quad y = 33,6727 + 2,7696x$$

keterangan :

x = konsentrasi (ppm)

$$y = 5$$

Perhitungan IC₅₀ fisetin

$$50 = a + bx$$

$$50 = 33,6727 + 2,7696x$$

$$33,6272x = 16,3273 \quad x$$

$$= 5,89 \text{ ppm}$$

b. Replikasi II

Konsentrasi (ppm)	Absorbansi	% Inhibisi
15,56	0,221	75,7
7,78	0,416	54,3
3,89	0,509	44,0
1,95	0,557	38,8
0,97	0,578	36,5

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 33,5812$$

$$b = 2,6973$$

$$r = 0,9999$$

$$y = a + bx$$

$$y = 33,58124 + 2,6973x$$

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC₅₀ fisetin

$$50 = a + bx$$

$$50 = 33,5812 + 2,6973x$$

$$33,5812x = 16,4188$$

$$x = 6,09 \text{ ppm}$$

c. Replikasi III

Konsentrasi (ppm)	Absorbansi	% Inhibisi
15,56	0,224	75,4
7,78	0,418	54,0
3,89	0,511	43,8
1,95	0,559	38,5
0,97	0,588	35,4

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 33,0177$$

$$b = 2,7215$$

$$r = 0,9999$$

$$y = a + bx$$

$$y = 33,0177 + 2,7215x$$

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC₅₀ fisetin

$$50 = a + bx$$

$$50 = 33,0177 + 2,7215x$$

$$33,0177x = 16,9823$$

$$x = 6,24 \text{ ppm}$$

Rata-rata IC₅₀ Fisetin :

$$\left. \begin{array}{l} \text{Replikasi I} = 5,89 \text{ ppm} \\ \text{Replikasi II} = 6,09 \text{ ppm} \\ \text{Replikasi III} = 6,24 \text{ ppm} \end{array} \right\} 6,07 \text{ ppm}$$