

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 dengan metode *emulsifikasi*.

Kedua, formula NLC fisetin stabil selama proses penyimpanan.

Ketiga, karakterisasi NLC fisetin menghasilkan ukuran partikel terkecil yaitu 539,33 nm, nilai potensial zeta sebesar -20,25 mV, memiliki efisiensi penyerapan terbesar sebanyak 59,20% dan terdapat aktivitas antioksidan kategori kuat pada fisetin dengan nilai IC50 sebesar 6,07 ppm, sedangkan pada formula NCL fisetin memiliki aktivitas antioksidan yang tergolong aktif.

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 pengujian stabilitas jangka panjang untuk mengetahui kestabilan formula NLC (*Nanostructured Lipid Carriers*) fisetin.

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

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Lampiran 1. Certificate of Analysis (CoA) Fisetin

TOCRIS
a biotechnne brand

Print Date: Jan 14th 2016

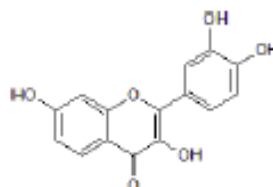
Certificate of Analysis

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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

1. PHYSICAL AND CHEMICAL PROPERTIES

Batch Molecular Formula: $C_{15}H_{10}O_6$
 Batch Molecular Weight: 266.24
 Physical Appearance: Yellow solid
 Solubility: DMSO to 100 mM
 ethanol to 10 mM
 Storage: Store at -20°C
 Batch Molecular Structure:



2. ANALYTICAL DATA

HPLC: Shows 98.1% purity
 $^1\text{H NMR}$: Consistent with structure
 Mass Spectrum: Consistent with structure

Microanalysis:

	Carbon	Hydrogen	Nitrogen
Theoretical	62.94	3.52	
Found	62.81	3.58	

Caution - Not Fully Tested • Research Use Only • Not For Human or Veterinary Use

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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.

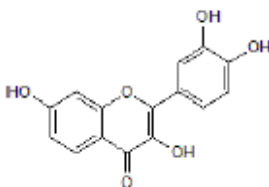
Physical and Chemical Properties:

Batch Molecular Formula: C₁₅H₁₀O₆

Batch Molecular Weight: 286.24

Physical Appearance: Yellow solid

Minimum Purity: >98%

Batch Molecular Structure:

References:

Lu et al (2005) Crystal structure of a human cyclin-dependent kinase 6 complex with a flavonol inhibitor, fisetin. *J.Med.Chem.* **48** 737. PMID: 15689157.

Lee et al (2005) Mechanisms for the inhibition of DNA methyltransferases by tea catechins and bioflavonoids. *Mol.Pharmacol.* **68** 1018. PMID: 16037419.

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Adhami et al (2012) Dietary flavonoid fisetin: a novel dual inhibitor of PI3K/Akt and mTOR for prostate cancer management. *Biochem.Pharmacol.* **84** 1277. PMID: 22842629.

Storage: Store at -20°C

Solubility & Usage Info:

DMSO to 100 mM
ethanol to 10 mM

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.

Caution - Not Fully Tested • Research Use Only • Not For Human or Veterinary Use

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Lampiran 2. Certificate of Analysis (CoA) Compritol



GATTEFOSSE

Certificate of Analysis

Product name : COMPRITOL 888 ATO			
Item N°	: 3123PPD		
Batch N°	: 169430		
Manufacturing date	: 04.2018		
Expiry date	: 04.2021		
CHEMICAL DEFINITION : Glycerol dibehenate EP / Glyceryl dibehenate NF / Glyceryl behenate Ch. P.			
Characteristic	Method	Specification	Result
MANUFACTURING PLANT		Gattefosse : 01 / SIO : 02 / Innovia : 06	02/01
APPEARANCE	MA0193	Fine powder	Conforms
ODOUR	MA0170	Faint	Conforms
COLOUR (Gardner Scale)	MA0214	< = 5.0	0.8
DROP POINT (METTLER)	MA0003	69.0 to 74.0 °C	73.2
MELTING POINT (Cap. tube)	MA0167	65.0 to 77.0 °C	65.0 to 77.0*
ACID VALUE	MA0008	< = 4.00 mgKOH/g	1.66
SAPONIFICATION VALUE	MA0172	145 to 165 mgKOH/g	150
IODINE VALUE	MA0092	< = 3.0 gI2/100g	< = 3.0*
PEROXIDE VALUE	MA0010	< = 6.0 meqO2/kg	< = 6.0*
FREE GLYCEROL CONTENT	MA0006	< = 1.0 %	0.7
1-MONOGLYCERIDES CONTENT	MA0006	12.0 to 18.0 %	16.4
TOTAL MONOGLYCERIDES CONTENT	MA0006	15.0 to 23.0 %	18.2
TOTAL DIESTERS CONTENT	MA0006	40.0 to 60.0 %	50.6
TOTAL TRIESTERS CONTENT	MA0006	21.0 to 35.0 %	29.5
PALMITIC ACID (C16)	MA0002	< = 3.0 %	0.9
STEARIC ACID (C18)	MA0002	< = 5.0 %	1.8
ARACHIDIC ACID (C20)	MA0002	< = 10.0 %	6.8
BEHENIC ACID (C22)	MA0002	> = 83.0 %	86.6
ERUCIC ACID (C22:1)	MA0002	< = 1.0 %	0.1
LIGNOCERIC ACID (C24:0)	MA0002	< = 3.0 %	1.6
RESIDUE ON IGNITION	MA0028	< = 0.1 %	< = 0.1*
TOTAL ASHES CONTENT	MA0028	< = 0.10 %	< = 0.10*
WATER CONTENT	MA0007	< = 1.0 %	< 0.1
NICKEL CONTENT	MA0256	< 1 ppm	Conforms *
HEAVY METALS CONTENT (Pb)	MA0032	< 10 ppm	Conforms *
RESIDUAL SOLVENTS (Eur. Pharm., USP/NF)		Meets the requirements (production without solvent)	Conforms
EUR. PHARM., USP/NF, JSFA		Conforms to the monographs	Conforms *
ARSENIC CONTENT	MA0323	< 2 ppm (expressed in AS2O3)	Conforms *
POLYOXYETHYLENE (JSFA)	MA0334	No change of chloroform layer to blue.	Conforms *
TLC (Id. test N° 1 and 2, JSFA, current ed.)	MA0335	Conforms	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			

Page : 1 / 2

Lampiran 3. 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 I) EP / Glyceryl 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	

Lampiran 4. Foto serbuk fisetin



Lampiran 5. Foto serbuk gliseril monostearat (GMS)



Lampiran 6. Foto serbuk precinol



Lampiran 7. Foto serbuk compritol




Lampiran 8. Foto tween 80



Lampiran 9. Foto isopropil miristat



Lampiran 10. Alat yang digunakan dalam penelitian

Nama Alat	Foto Alat	Kegunaan
<i>Particle Size Analyzer (PSA)</i>		alat yang digunakan untuk mengukur ukuran partikel
Zeta Nanosizer ZS		analisa ukuran zeta potensial menggunakan hamburan cahaya elektroforesis
<i>Spektrofotometri Uv-vis</i>		merupakan spektrofotometer yang digunakan untuk pengukuran serapan cahaya di daerah ultraviolet (200 –350 nm) dan sinar tampak (350 – 800 nm) oleh suatu senyawa.
<i>Magnetic stirrer</i>		berfungsi menghomogenkan suatu larutan dengan pengadukan.

Lampiran 11. Screening lipid

a. Konsentrasi 2%



b. Konsentrasi 4%



c. Konsentrasi 6%



Formula lipid yang terpilih yaitu pada konsentrasi lipid 2% untuk masing-masing lipid yaitu gliseril monostearat, compritol dan precirol dengan pembentukan emulsi yang stabil dan tidak keruh.

Lampiran 12. Penentuan panjang gelombang dan pembuatan kurva baku

a. Penentuan panjang gelombang

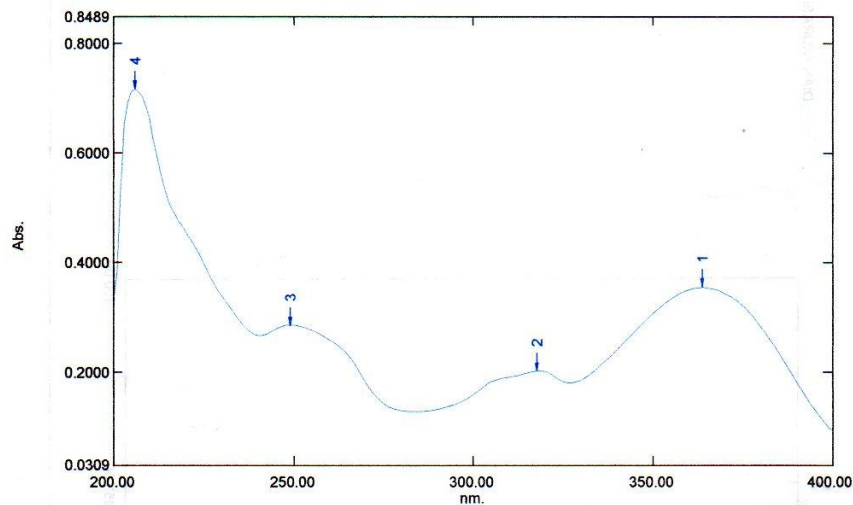
Spectrum Peak Pick Report

16/2019
4

Fisetin

Maks: 364 nm
04/16/2019 08:04:23 AM

Data Set: DIAN AKURASI_080249 - RawData



[Measurement Properties]
 Wavelength Range (nm.): 200.00 to 400.00
 Scan Speed: Medium
 Sampling Interval: 1.0
 Auto Sampling Interval: Disabled
 Scan Mode: Auto

[Instrument Properties]
 Instrument Type: UV-1800 Series
 Measuring Mode: Absorbance
 Slit Width: 1.0 nm
 Light Source Change Wavelength: 340.0 nm
 S/R Exchange: Normal

[Attachment Properties]
 Attachment: None

[Operation]
 Threshold: 0.0010000
 Points: 4
 InterPolate: Disabled
 Average: Disabled

[Sample Preparation Properties]
 Weight:
 Volume:
 Dilution:
 Path Length:
 Additional Information:

No.	P/V	Wavelength	Abs.	Description
1	●	364.00	0.3545	
2	●	318.00	0.2021	
3	●	249.00	0.2859	
4	●	206.00	0.7175	
5	●	327.00	0.1800	
6	●	284.00	0.1279	
7	●	241.00	0.2665	

Panjang gelombang maksimum yang diperoleh dari *scanning* larutan fisetin dalam etanol pro analisis, panjang gelombang maksimum yang diperoleh sebesar 364 nm dengan serapan 0,3545 nm.

b. Penentuan *operating time***Kinetics Data Print Report**

05/15/2019 03:46:21 PM

Time (Minute)	RawData ...
0.000	0.457
1.000	0.458
2.000	0.459
3.000	0.460
4.000	0.459
5.000	0.461
6.000	0.460
7.000	0.461
8.000	0.460
9.000	0.460
10.000	0.462
11.000	0.462
12.000	0.462
13.000	0.462
14.000	0.463
15.000	0.463
16.000	0.464
17.000	0.464
18.000	0.463
19.000	0.464
20.000	0.464
21.000	0.465
22.000	0.465
23.000	0.464
24.000	0.464
25.000	0.466
26.000	0.465
27.000	0.466
28.000	0.466
29.000	0.466
30.000	0.466

c. Kurva kalibrasi (Linieritas)

Penimbangan bahan :

$$\text{Kertas kosong} = 0,2736$$

$$\text{Kertas + bahan} = 0,2786$$

$$\text{Kertas + sisa} = \frac{0,2740}{0,0046} -$$

Perhitungan konsentrasi kurva kalibrasi :

$$1) V_1 \times N_1 = V_2 \times N_2$$

$$46 \text{ ppm} \times 0,8 \text{ ml} = V_2 \times 10 \text{ ml}$$

$$V_2 = 3,68 \text{ ppm}$$

$$2) V_1 \times N_1 = V_2 \times N_2$$

$$46 \text{ ppm} \times 1 \text{ ml} = V_2 \times 10 \text{ ml}$$

$$V_2 = 4,6 \text{ ppm}$$

$$3) V_1 \times N_1 = V_2 \times N_2$$

$$46 \text{ ppm} \times 1,2 \text{ ml} = V_2 \times 10 \text{ ml}$$

$$V_2 = 5,52 \text{ ppm}$$

$$4) V_1 \times N_1 = V_2 \times N_2$$

$$46 \text{ ppm} \times 1,4 \text{ ml} = V_2 \times 10 \text{ ml}$$

$$V_2 = 6,44 \text{ ppm}$$

$$5) V_1 \times N_1 = V_2 \times N_2$$

$$46 \text{ ppm} \times 1,6 \text{ ml} = V_2 \times 10 \text{ ml}$$

$$V_2 = 7,36 \text{ ppm}$$

$$6) V_1 \times N_1 = V_2 \times N_2$$

$$46 \text{ ppm} \times 1,8 \text{ ml} = V_2 \times 10 \text{ ml}$$

$$V_2 = 8,28 \text{ ppm}$$

Konsentrasi (ppm)	Absorbansi
3,68	0,246
4,6	0,305
5,52	0,371
6,44	0,424
7,36	0,478
8,28	0,541

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

$$a = 0,0140$$

$$b = 0,0636$$

$$r = 0,99951$$

$$y = a + bx$$

$$y = 0,0140 + 0,0636x$$

keterangan :

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

y = serapan

Hasil linearitas diperoleh $R = 0,99951$; sehingga dapat disimpulkan bahwa data tersebut linier.

d. Akurasi

Konsentrasi (ppm)	Absorbansi	Recovery (%)	Rata-rata (%)
4,6	0,308	101	
4,6	0,308	101	
4,6	0,305	100	100
5,52	0,366	100	
5,52	0,366	100	
5,52	0,365	100	100
6,44	0,429	100	
6,44	0,429	100	
6,44	0,425	100	100

Nilai rata-rata recovery dilihat dari data diatas adalah 100%

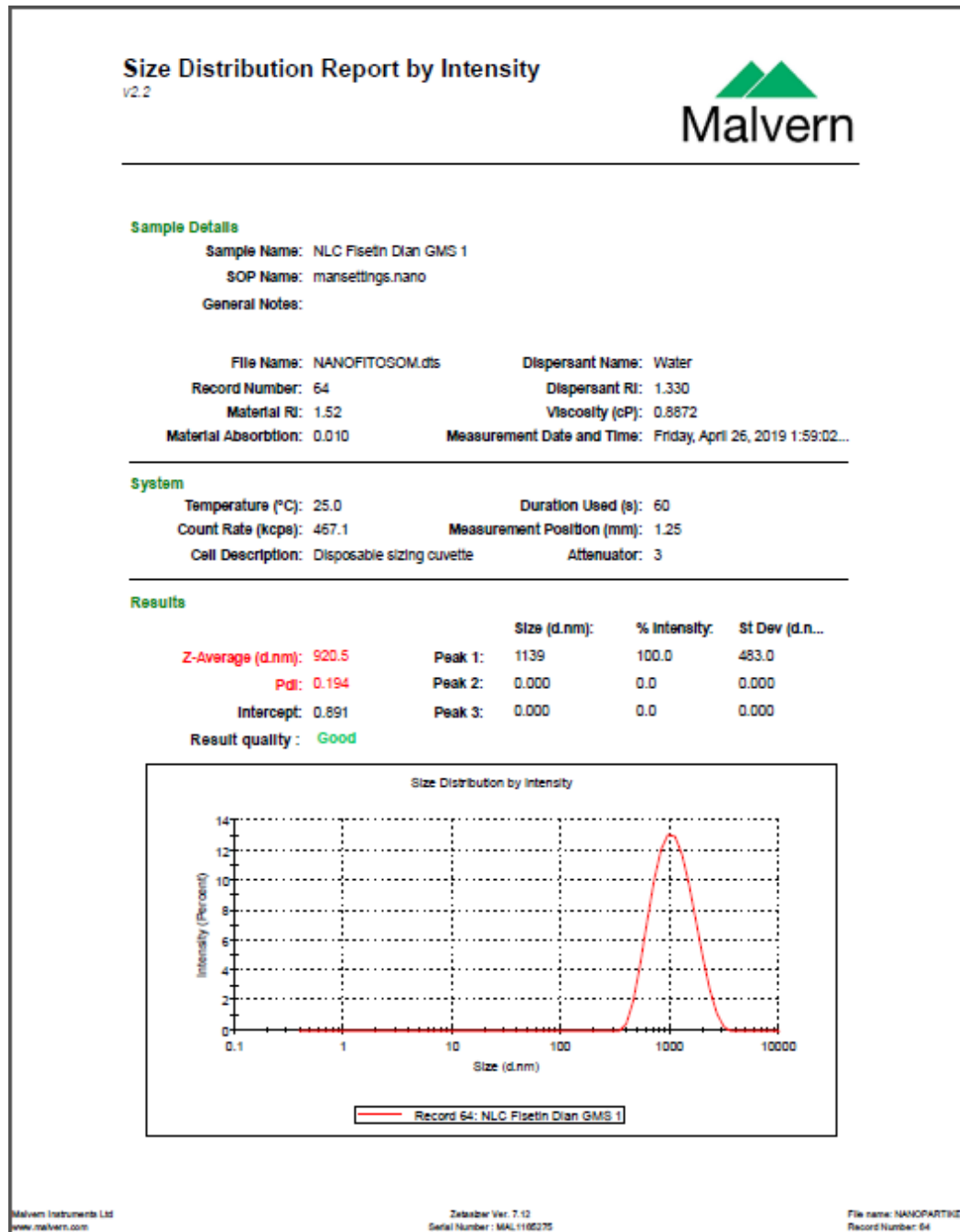
e. Presisi

Konsentrasi (ppm)	Absorbansi
4,6	0,367
4,6	0,365
4,6	0,369
4,6	0,370
4,6	0,366
4,6	0,368
4,6	0,369
4,6	0,371
4,6	0,370
4,6	0,366
Rata-rata	6,149
SD	0,034
CV	1%

Nilai CV dilihat dari data diatas adalah 1%, hasil ini sesuai dengan persyaratan presisi yaitu $\leq 2\%$.

Lampiran 13. Uji ukuran partikel formula I (GMS 2%)

a. Replikasi I



b. Replikasi II

Size Distribution Report by Intensity

v2.2



Sample Details

Sample Name: NLC Fisetin Dian GMS 2

SOP Name: mansettings.nano

General Notes:

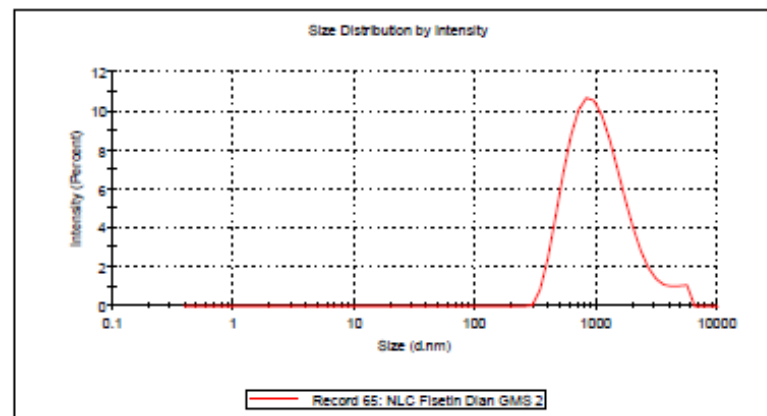
File Name: NANOFITOGOM.dts	Dispersant Name: Water
Record Number: 65	Dispersant RI: 1.330
Material RI: 1.52	Viscosity (cP): 0.8872
Material Absorption: 0.010	Measurement Date and Time: Friday, April 26, 2019 2:01:05...

System

Temperature (°C): 25.0	Duration Used (s): 60
Count Rate (kcps): 461.2	Measurement Position (mm): 1.25
Cell Description: Disposable sizing cuvette	Attenuator: 3

Results

	Size (d.nm):	% Intensity:	St Dev (d.n...)
Z-Average (d.nm): 907.0	Peak 1: 1149	96.9	698.0
PDI: 0.243	Peak 2: 4852	3.1	578.9
Intercept: 0.893	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 Fisetin Dian GMS 3

SOP Name: mansettings.nano

General Notes:

File Name: NANOFITOSOM.dts

Dispersant Name: Water

Record Number: 66

Dispersant RI: 1.330

Material RI: 1.52

Viscosity (cP): 0.8872

Material Absorbtion: 0.010

Measurement Date and Time: Friday, April 26, 2019 2:03:09...

System

Temperature (°C): 25.0

Duration Used (s): 60

Count Rate (kcps): 452.6

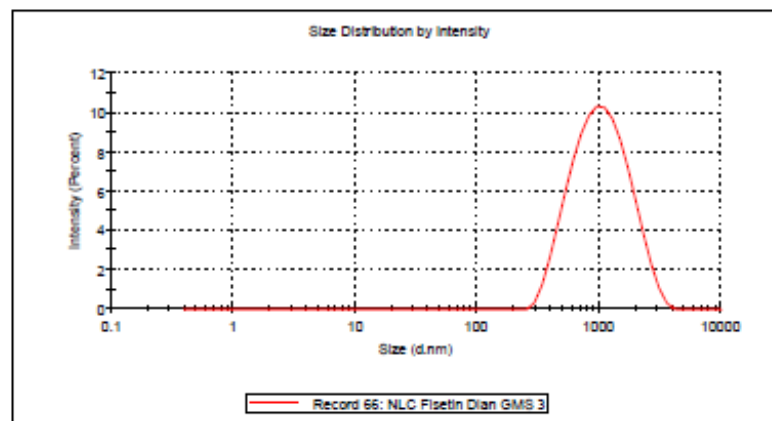
Measurement Position (mm): 1.25

Cell Description: Disposable sizing cuvette

Attenuator: 3

Results

	Size (d.nm):	% Intensity:	St Dev (d.n...)
Z-Average (d.nm): 889.5	Peak 1: 1150	100.0	600.4
Pdi: 0.210	Peak 2: 0.000	0.0	0.000
Intercept: 0.891	Peak 3: 0.000	0.0	0.000
Result quality : Good			



Lampiran 14. Hasil uji ukuran partikel formula II (Precirol 2%)

a. Replikasi I

Size Distribution Report by Intensity v2.2



Sample Details

Sample Name: NLC Fisetin Dian PRESIROL 1

SOP Name: mansettings.nano

General Notes:

File Name: NANOFITOSOM.dfs	Dispersant Name: Water
Record Number: 67	Dispersant RI: 1.330
Material RI: 1.52	Viscosity (cP): 0.8672
Material Absorbtion: 0.010	Measurement Date and Time: Friday, April 26, 2019 2:09:28...

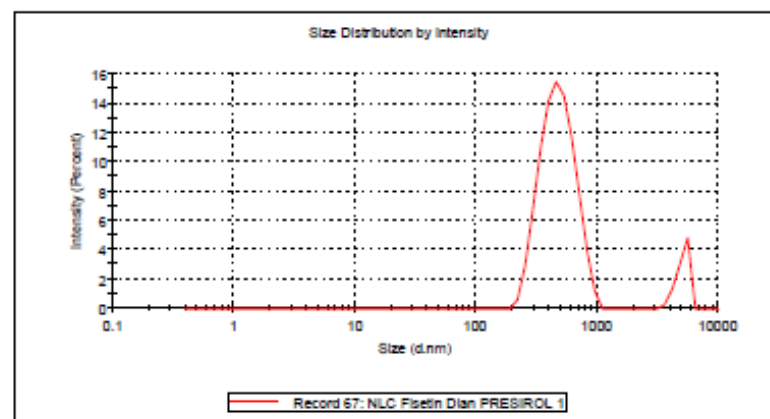
System

Temperature (°C): 25.0	Duration Used (s): 60
Count Rate (kcps): 382.4	Measurement Position (mm): 1.05
Cell Description: Disposable sizing cuvette	Attenuator: 3

Results

	Size (d.nm):	% Intensity:	St Dev (d.n...
Z-Average (d.nm): 546.7	Peak 1: 490.8	90.4	154.6
Pdl: 0.382	Peak 2: 5054	9.6	578.1
Intercept: 0.899	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 Fisetin Dian PRESIROL 2

SOP Name: mansettings.nano

General Notes:

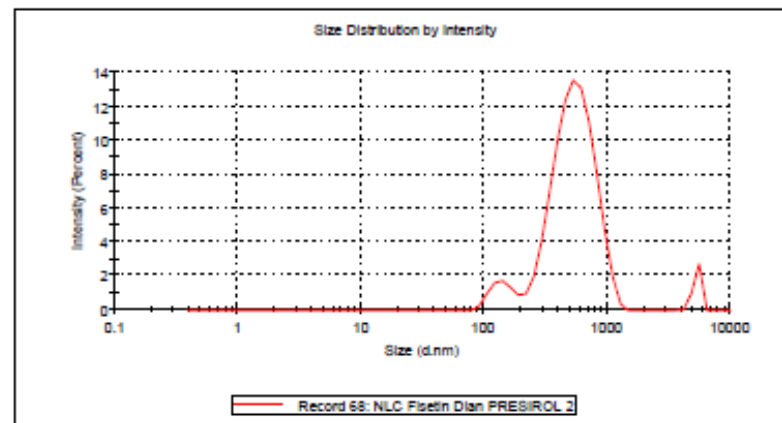
File Name: NANOFITOSOM.dts	Dispersant Name: Water
Record Number: 68	Dispersant RI: 1.330
Material RI: 1.52	Viscosity (cP): 0.8872
Material Absorbtion: 0.010	Measurement Date and Time: Friday, April 26, 2019 2:11:31...

System

Temperature (°C): 25.0	Duration Used (s): 60
Count Rate (kcps): 381.4	Measurement Position (mm): 1.05
Cell Description: Disposable sizing cuvette	Attenuator: 3

Results

	Size(d.nm):	% Intensity:	St Dev (d.n...
Z-Average (d.nm): 539.9	Peak 1: 567.0	89.5	206.7
PdI: 0.375	Peak 2: 141.0	6.8	27.86
Intercept: 0.896	Peak 3: 5344	3.7	357.8
Result quality : Good			



c. Replikasi III

Size Distribution Report by Intensity

v2.2



Sample Details

Sample Name: NLC Fisetin Dian PRESIROL 3

SOP Name: mansettings.nano

General Notes:

File Name: NANOFITOSOM.dts

Dispersant Name: Water

Record Number: 69

Dispersant RI: 1.330

Material RI: 1.52

Viscosity (cP): 0.8872

Material Absorbtion: 0.010

Measurement Date and Time: Friday, April 26, 2019 2:13:35...

System

Temperature (°C): 25.0

Duration Used (s): 60

Count Rate (kcps): 422.7

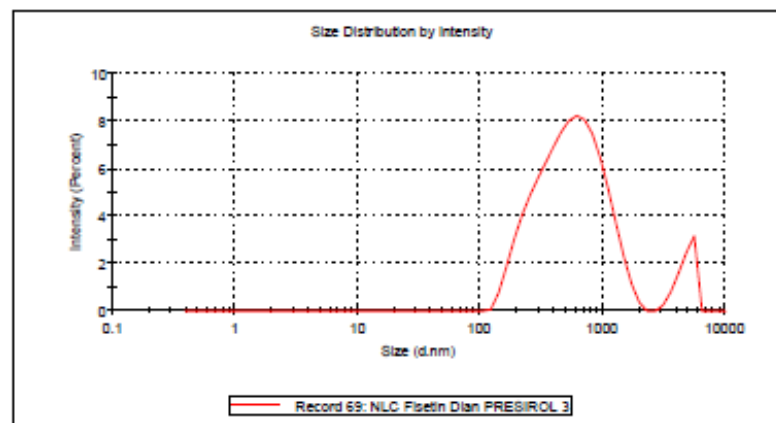
Measurement Position (mm): 1.05

Cell Description: Disposable sizing cuvette

Attenuator: 3

Results

	Size (d.nm):	% Intensity:	St Dev (d.n...
Z-Average (d.nm): 533.6	Peak 1: 621.2	91.7	358.6
Pdi: 0.406	Peak 2: 4791	8.3	732.9
Intercept: 0.891	Peak 3: 0.000	0.0	0.000
Result quality : Good			



Lampiran 15. Hasil uji ukuran partikel formula III (Compritol 2%)

a. Replikasi I

Size Distribution Report by Intensity

v2.2



Sample Details

Sample Name: NLC Fisetin Dian COMPRITOL 1

SOP Name: mansettings.nano

General Notes:

File Name: NANOFITOSOM.dts	Dispersant Name: Water
Record Number: 70	Dispersant RI: 1.330
Material RI: 1.52	Viscosity (cP): 0.8872
Material Absorbtion: 0.010	Measurement Date and Time: Friday, April 26, 2019 2:19:49...

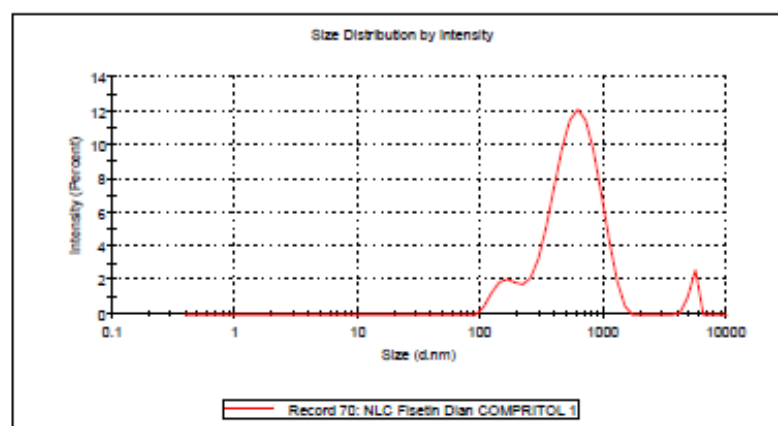
System

Temperature (°C): 25.0	Duration Used (s): 60
Count Rate (kcps): 388.0	Measurement Position (mm): 1.05
Cell Description: Disposable sizing cuvette	Attenuator: 3

Results

	Size (d.nm):	% Intensity:	St Dev (d.n...
Z-Average (d.nm): 550.1	Peak 1: 631.2	87.0	252.5
Pdl: 0.439	Peak 2: 166.9	9.3	35.06
Intercept: 0.895	Peak 3: 5311	3.7	392.7

Result quality : **Good**



b. Replikasi II

Size Distribution Report by Intensity

v2.2



Sample Details

Sample Name: NLC Fisethn Dian COMPRITOL 2

SOP Name: mansettings.nano

General Notes:

File Name: NANOFITOSOM.dts

Dispersant Name: Water

Record Number: 71

Dispersant RI: 1.330

Material RI: 1.52

Viscosity (cP): 0.8872

Material Absorbtion: 0.010

Measurement Date and Time: Friday, April 26, 2019 2:21:52...

System

Temperature (°C): 25.0

Duration Used (s): 60

Count Rate (kcps): 374.5

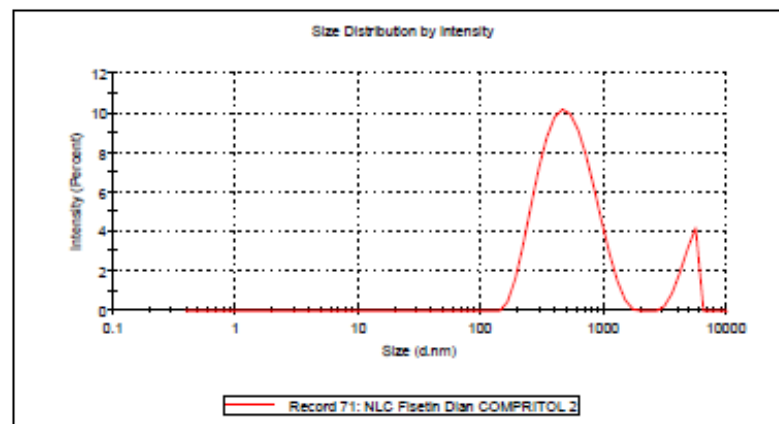
Measurement Position (mm): 1.05

Cell Description: Disposable sizing cuvette

Attenuator: 3

Results

	Size (d.nm):	% Intensity:	St Dev (d.n...
Z-Average (d.nm): 547.6	Peak 1: 539.8	89.5	259.7
Pdl: 0.362	Peak 2: 4836	10.5	712.3
Intercept: 0.896	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 Fisetin Dian COMPRITOL 3

SOP Name: mansettings.nano

General Notes:

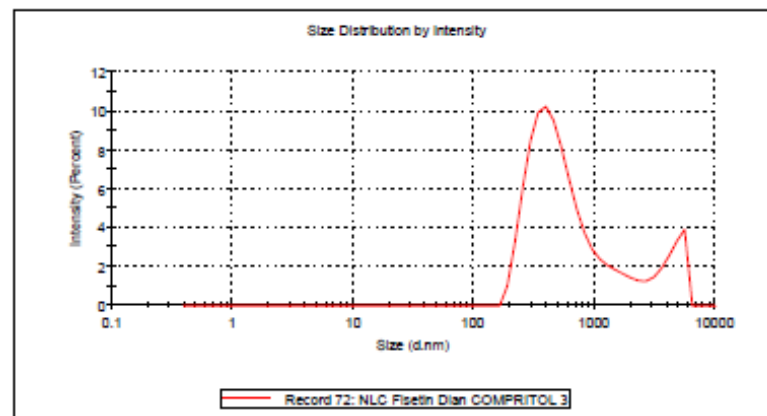
File Name: NANOFITOSOM.dts	Dispersant Name: Water
Record Number: 72	Dispersant RI: 1.330
Material RI: 1.52	Viscosity (cP): 0.8872
Material Absorbtion: 0.010	Measurement Date and Time: Friday, April 26, 2019 2:23:56...

System

Temperature (°C): 25.0	Duration Used (s): 60
Count Rate (kcps): 407.2	Measurement Position (mm): 1.05
Cell Description: Disposable sizing cuvette	Attenuator: 3

Results

	Size (d.nm):	% Intensity:	St Dev (d.nm):
Z-Average (d.nm): 557.0	Peak 1: 629.7	85.7	492.8
Pdi: 0.436	Peak 2: 4365	14.3	968.4
Intercept: 0.899	Peak 3: 0.000	0.0	0.000
Result quality : Good			



Lampiran 16. Hasil uji zeta potensial formula II (Precirol 2%)

Instrument

Serial Number: 3214-DMP
 Model: DelsaMax Pro
 Pals Firmware Version: 1.1.0.6
 DLS Firmware Version: 2.3.1.0
 Assist Firmware Version: 1.0.0.9
 Instrument Name: BCI-3214-DMP
 Laser Wavelength (nm): 532.0
 Has DLS: Yes
 DLS Detector Angle (degrees): 163.5
 Minimum Temperature (C): 3.5
 Minimum Temperature without N2 (C): 20
 Maximum Temperature (C): 70
 Minimum Ramp Rate (C/min): 0
 Maximum Ramp Rate (C/min): 1.5

Instrument Parameters: Measurements

Collect Data: DLS and Pals (Simultaneous)
 Acq Time (s): 20
 Read Interval (s): 1
 Number Acq: 3
 Electric Field Frequency (Hz): 10.0
 Voltage Amplitude (V): 2.5
 Collection Period (s): 15.0
 Auto-attenuation: Yes
 Attenuation Level (%): 0
 Auto-attenuation Time Limit(s): 0
 Laser Mode: Normal
 Set Temp On Connection: No
 Set Temp (C): 20
 Temp Ramp Enabled: Yes
 Temp Ramp Rate (C/min): 1

Datalog Table: Measurements

Item	Zeta Potential	Radius	Diameter	
	(mV)	(nm)	(nm)	
1	Meas 1	-13.91	216.3	432.6
2	Meas 2	-13.12	215.3	430.7
3	Meas 3	-15.00	216.7	433.4
Mean		-14.01	216.1	432.2
S		0.94	0.7	1.4
%S		6.73	0.3	0.3
S ²		0.89	0.5	1.9
Min		-15.00	215.3	430.7
Max		-13.12	216.7	433.4

Lampiran 17. Hasil uji zeta potensial formula III (compritol 2%)

Instrument

Serial Number: 3214-DMP
 Model: DelsaMax Pro
 Pals Firmware Version: 1.1.0.6
 DLS Firmware Version: 2.3.1.0
 Assist Firmware Version: 1.0.0.9
 Instrument Name: BCI-3214-DMP
 Laser Wavelength (nm): 532.0
 Has DLS: Yes
 DLS Detector Angle (degrees): 163.5
 Minimum Temperature (C): 3.5
 Minimum Temperature without N2 (C): 20
 Maximum Temperature (C): 70
 Minimum Ramp Rate (C/min): 0
 Maximum Ramp Rate (C/min): 1.5

Instrument Parameters: Measurements

Collect Data: DLS and Pals (Simultaneous)
 Acq Time (s): 20
 Read Interval (s): 1
 Number Acq: 3
 Electric Field Frequency (Hz): 10.0
 Voltage Amplitude (V): 2.5
 Collection Period (s): 15.0
 Auto-attenuation: Yes
 Attenuation Level (%): 0
 Auto-attenuation Time Limit(s): 0
 Laser Mode: Normal
 Set Temp On Connection: No
 Set Temp (C): 20
 Temp Ramp Enabled: Yes
 Temp Ramp Rate (C/min): 1

Datalog Table: Measurements

	Item	Zeta Potential (mV)	Radius (nm)	Diameter (nm)
1	Meas 1	-19.22	109.2	218.3
2	Meas 2	-21.00	138.6	277.1
3	Meas 3	-20.51	138.7	277.5
	Mean	-20.25	128.8	257.6
	S	0.92	17.0	34.1
	%S	4.55	13.2	13.2
	S ²	0.85	289.9	1159.5
	Min	-21.00	109.2	218.3
	Max	-19.22	138.7	277.5

Lampiran 18. Perhitungan efisiensi penjerapan NLC Fisetin

a. Formula I (GMS 2%)

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

Perhitungan teoritis

$$\text{Fisetin} = 10 \text{ mg}$$

$$\text{Eksipien (tween 80 + IPM + GMS)} = 11.500 \text{ mg}$$

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

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

Perhitungan kadar fisetin terjerap menggunakan persamaan regresi linier :

$$y = a + bx$$

$$0,271 = 0,014 + ,00636x$$

$$0,0636x = 0,257$$

$$X = 4,041 \text{ ppm}$$

$$\% \text{ kadar} = \frac{4,041 \text{ ppm}}{20.000 \text{ ppm}} \times 100\% = 0,02007\%$$

$$\text{Kadar dalam 200mg NLC fisetin} = 0,02007\% \times 200 \text{ mg} = 0,040 \text{ mg}$$

$$\% \text{ Efisiensi penjerapan (EE)} = \frac{\text{kadar terjerap}}{\text{kadar teoritis}} \times 100\%$$

$$= \frac{0,040 \text{ mg}}{0,174 \text{ mg}} \times 100\%$$

$$= 22,99\%$$

b. Formula II (Precirol 2%)

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

Perhitungan teoritis

$$\text{Fisetin} = 10 \text{ mg}$$

$$\text{Eksipien (tween 80 + IPM + GMS)} = 11.500 \text{ mg}$$

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

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

Perhitungan kadar fisetin terjerap menggunakan persamaan regresi linier :

$$y = a + bx$$

$$0,449 = 0,014 + ,00636x$$

$$0,0636x = 0,435$$

$$\begin{aligned}
 x &= 6,840 \text{ ppm} \\
 \% \text{ kadar} &= \frac{6,840 \text{ ppm}}{20.000 \text{ ppm}} \times 100\% = 0,0324\% \\
 \text{Kadar dalam 200mg NLC fisetin} &= 0,0324\% \times 200 \text{ mg} = 0,0648 \text{ mg} \\
 \% \text{ Efisiensi penyerapan (EE)} &= \frac{\text{kadar terjerap}}{\text{kadar teoritis}} \times 100\% \\
 &= \frac{0,0648 \text{ mg}}{0,174 \text{ mg}} \times 100\% \\
 &= 59,20\%
 \end{aligned}$$

c. Formula III (Compritol 2%)

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

Perhitungan teoritis

$$\begin{aligned}
 \text{Fisetin} &= 10 \text{ mg} \\
 \text{Eksipien (tween 80 + IPM + GMS)} &= 11.500 \text{ mg} \\
 \% \text{ kadar fisetin} &= \frac{10}{11.500+10} \times 100\% = 0,0869\% \\
 \text{Kadar dalam 200 mg NLC} &= 0,0869\% \times 200\text{mg} = 0,1738 \text{ mg}
 \end{aligned}$$

Perhitungan kadar fisetin terjerap menggunakan persamaan regresi linier :

$$\begin{aligned}
 y &= a + bx \\
 0,670 &= 0,014 + ,00636x \\
 0,0636x &= 0,656 \\
 x &= 10,314 \text{ ppm} \\
 \% \text{ kadar} &= \frac{10,314 \text{ ppm}}{20.000 \text{ ppm}} \times 100\% = 0,05157\% \\
 \text{Kadar dalam 200mg NLC fisetin} &= 0,05157\% \times 200 \text{ mg} = 0,103 \text{ mg} \\
 \% \text{ Efisiensi penyerapan (EE)} &= \frac{\text{kadar terjerap}}{\text{kadar teoritis}} \times 100\% \\
 &= \frac{0,103 \text{ mg}}{0,174 \text{ mg}} \times 100\% \\
 &= 37,24\%
 \end{aligned}$$

Lampiran 19. Uji stabilitas NLC fisetin setelah penyimpanan secara visual**Pengamatan secara visual**

Minggu Ke-1



Minggu Ke-2



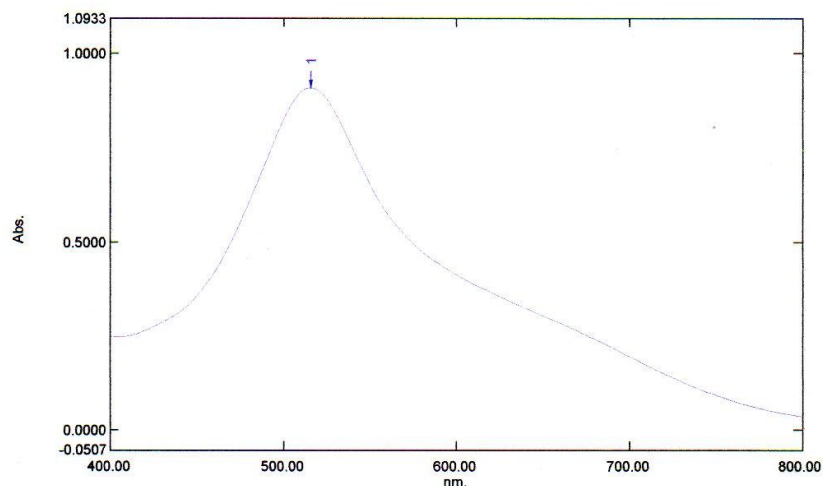
Lampiran 20. Uji Aktifitas Antioksidan

a. Penentuan panjang gelombang maks

Spectrum Peak Pick Report

05/10/2019 10:03:42 AM

Data Set: lamda maks dpph fix 1 - RawData



[Measurement Properties]
 Wavelength Range (nm.): 400.00 to 800.00
 Scan Speed: Medium
 Sampling Interval: 1.0
 Auto Sampling Interval: Disabled
 Scan Mode: Auto

No.	P/V	Wavelength	Abs.	Description
1	●	516.00	0.9096	
2	⬇	405.00	0.2479	

[Instrument Properties]
 Instrument Type: UV-1800 Series
 Measuring Mode: Absorbance
 Slit Width: 1.0 nm
 Light Source Change Wavelength: 340.0 nm
 S/R Exchange: Normal

[Attachment Properties]
 Attachment: None

[Operation]
 Threshold: 0.0010000
 Points: 4
 InterPolate: Disabled
 Average: Disabled

[Sample Preparation Properties]
 Weight:
 Volume:
 Dilution:
 Path Length:
 Additional Information:

b. Penentuan *operating time*OT Firetin DPPH 4/5/19
EO MEHIT

Kinetics Data Print Report

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

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 21. DPPH fisetin

Perhitungan bahan fisetin :

$$50\text{mg}/100\text{ml} = 500\text{mg}/1000\text{ml} = 500 \text{ ppm}$$

Perhitungan konsentrasi (ppm) :

$$1) \quad V_1 \times N_1 = V_2 \times N_2$$

$$500 \text{ ppm} \times 0,3112\text{ml} = V_2 \times 10\text{ml}$$

$$V_2 = 15,56 \text{ ppm}$$

$$2) \quad V_1 \times N_1 = V_2 \times N_2$$

$$500 \text{ ppm} \times 0,1556\text{ml} = V_2 \times 10\text{ml}$$

$$V_2 = 7,78 \text{ ppm}$$

$$3) \quad V_1 \times N_1 = V_2 \times N_2$$

$$500 \text{ ppm} \times 0,0778\text{ml} = V_2 \times 10\text{ml}$$

$$V_2 = 3,89 \text{ ppm}$$

$$4) \quad V_1 \times N_1 = V_2 \times N_2$$

$$500 \text{ ppm} \times 0,039\text{ml} = V_2 \times 10\text{ml}$$

$$V_2 = 1,95 \text{ ppm}$$

$$5) \quad V_1 \times N_1 = V_2 \times N_2$$

$$500 \text{ ppm} \times 0,0194\text{ml} = V_2 \times 10\text{ml}$$

$$V_2 = 0,97 \text{ ppm}$$

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 :

$$a = 33,6727$$

$$b = 2,7696$$

$$r = 0,99994$$

$$y = a + bx$$

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

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC50 fisetin

$$50 = a + bx$$

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

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

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

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 :

Replikasi I	= 5,89 ppm	} 6,07 ppm
Replikasi II	= 6,09 ppm	
Replikasi III	= 6,24 ppm	

Lampiran 22. DPPH formula I (Gliseril monostearat 2%)

Perhitungan fisetin dalam formula :

$$10\text{mg}/50\text{ ml} = 200\text{mg}/1000\text{ml} = 200\text{ppm}$$

Perhitungan konsentrasi (ppm) :

$$1) \quad V_1 \times N_1 = V_2 \times N_2$$

$$200 \text{ ppm} \times 2,5\text{ml} = V_2 \times 10\text{ml}$$

$$V_2 = 50 \text{ ppm}$$

$$2) \quad V_1 \times N_1 = V_2 \times N_2$$

$$200 \text{ ppm} \times 1,25\text{ml} = V_2 \times 10\text{ml}$$

$$V_2 = 25 \text{ ppm}$$

$$3) \quad V_1 \times N_1 = V_2 \times N_2$$

$$200 \text{ ppm} \times 0,625\text{ml} = V_2 \times 10\text{ml}$$

$$V_2 = 12,5 \text{ ppm}$$

$$4) \quad V_1 \times N_1 = V_2 \times N_2$$

$$200 \text{ ppm} \times 0,3125\text{ml} = V_2 \times 10\text{ml}$$

$$V_2 = 6,25 \text{ ppm}$$

$$5) \quad V_1 \times N_1 = V_2 \times N_2$$

$$200 \text{ ppm} \times 0,156\text{ml} = V_2 \times 10\text{ml}$$

$$V_2 = 3,125 \text{ ppm}$$

a. Replikasi I

Konsentrasi (ppm)	Absorbansi	% Inhibisi
50	0,467	48,7
25	0,604	33,6
12,5	0,675	25,8
6,25	0,708	22,2
3,125	0,729	19,9

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 18,1692$$

$$b = 0,6113$$

$$r = 0,999901$$

$$y = a + bx$$

$$y = 18,1692 + 0,6113x$$

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC50

$$50 = a + bx$$

$$50 = 18,1692 + 0,6113x$$

$$0,6113x = 31,8308$$

$$x = 52,07 \text{ ppm}$$

b. Replikasi II

Konsentrasi (ppm)	Absorbansi	% Inhibisi
50	0,459	49,5
25	0,604	33,6
12,5	0,673	26,0
6,25	0,710	21,9
3,125	0,726	20,2

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 18,1050$$

$$b = 0,6271$$

$$r = 0,999942$$

$$y = a + bx$$

$$y = 18,1050 + 0,6271x$$

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC50

$$50 = a + bx$$

$$50 = 18,1050 + 0,6271x$$

$$0,6271x = 31,895$$

$$x = 50,86 \text{ ppm}$$

c. Replikasi III

Konsentrasi (ppm)	Absorbansi	% Inhibisi
50	0,456	48,4
25	0,599	33,3
12,5	0,668	26,0
6,25	0,702	21,9
3,125	0,719	20,2

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 18,9159$$

$$b = 0,6170$$

$$r = 0,999939$$

$$y = a + bx$$

$$y = 18,9159 + 0,6170x$$

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC₅₀

$$50 = a + bx$$

$$50 = 18,9159 + 0,6170x$$

$$0,6170x = 31,0841$$

$$x = 50,38 \text{ ppm}$$

Rata-rata IC₅₀ Formula I :

Replikasi I	= 52,07 ppm	} 51,10 ppm
Replikasi II	= 50,86 ppm	
Replikasi III	= 50,38 ppm	

Lampiran 23. Data statistik DPPH formula I

→ T-Test

[DataSet0]

One-Sample Statistics

	N	Mean	Std. Deviation	Std. Error Mean
IC50	3	51,1033	,87088	,50280

One-Sample Test

	Test Value = 6.07					
	t	df	Sig. (2-tailed)	Mean Difference	95% Confidence Interval of the Difference	
					Lower	Upper
IC50	89,565	2	,000	45,03333	42,8699	47,1967

Kesimpulan : Nilai sig. (2-tailed) yaitu $0,000 < 0,05$ yang berarti **terdapat perbedaan yang bermakna** antara nilai IC50 fisetin hasil praktikum dengan nilai IC50 formula I.

Lampiran 24. DPPH formula II (Precirol 2%)

Perhitungan fisetin dalam formula :

$$10\text{mg}/50\text{ ml} = 200\text{mg}/1000\text{ml} = 200\text{ppm}$$

Perhitungan konsentrasi (ppm) :

- 1) $V_1 \times N_1 = V_2 \times N_2$
 $200\text{ ppm} \times 2,5\text{ml} = V_2 \times 10\text{ml}$
 $V_2 = 50\text{ ppm}$
- 2) $V_1 \times N_1 = V_2 \times N_2$
 $200\text{ ppm} \times 1,25\text{ml} = V_2 \times 10\text{ml}$
 $V_2 = 25\text{ ppm}$
- 3) $V_1 \times N_1 = V_2 \times N_2$
 $200\text{ ppm} \times 0,625\text{ml} = V_2 \times 10\text{ml}$
 $V_2 = 12,5\text{ ppm}$
- 4) $V_1 \times N_1 = V_2 \times N_2$
 $200\text{ ppm} \times 0,3125\text{ml} = V_2 \times 10\text{ml}$
 $V_2 = 6,25\text{ ppm}$
- 5) $V_1 \times N_1 = V_2 \times N_2$
 $200\text{ ppm} \times 0,156\text{ml} = V_2 \times 10\text{ml}$
 $V_2 = 3,125\text{ ppm}$

a. Replikasi I

Konsentrasi (ppm)	Absorbansi	% Inhibisi
50	0,468	48,5
25	0,607	33,1
12,5	0,675	25,8
6,25	0,709	22,1
3,125	0,728	20,0

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 18,1554$$

$$b = 0,6075$$

$$r = 0,99997$$

$$y = a + bx$$

$$y = 18,1554 + 0,6075x$$

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC50

$$50 = a + bx$$

$$50 = 18,1554 + 0,6075x$$

$$0,6075x = 31,8446$$

$$x = 52,42 \text{ ppm}$$

b. Replikasi II

Konsentrasi (ppm)	Absorbansi	% Inhibisi
50	0,459	49,5
25	0,599	34,1
12,5	0,669	26,5
6,25	0,700	23,0
3,125	0,718	21,1

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 19,0807$$

$$b = 0,6074$$

$$r = 0,9999$$

$$y = a + bx$$

$$y = 19,0807 + 0,6074x$$

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC50

$$50 = a + bx$$

$$50 = 19,0807 + 0,6074x$$

$$0,6074x = 31,9193$$

$$x = 50,91 \text{ ppm}$$

c. Replikasi III

Konsentrasi (ppm)	Absorbansi	% Inhibisi
50	0,469	48,4
25	0,607	33,3
12,5	0,673	26,0
6,25	0,710	21,9
3,125	0,726	20,2

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 18,2929$$

$$b = 0,6027$$

$$r = 0,99994$$

$$y = a + bx$$

$$y = 18,2929 + 0,6027x$$

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC₅₀

$$50 = a + bx$$

$$50 = 18,2929 + 0,6027x$$

$$0,6027x = 31,7071$$

$$x = 52,61 \text{ ppm}$$

Rata-rata IC₅₀ Formula II :

Replikasi I	= 52,42 ppm	} 51,98 ppm
Replikasi II	= 50,91 ppm	
Replikasi III	= 52,61 ppm	

Lampiran 25. Data statistik DPPH formula II (Precirol 2%)

T-Test

[DataSet0]

One-Sample Statistics

	N	Mean	Std. Deviation	Std. Error Mean
IC50	3	51,9800	,93150	,53780

One-Sample Test

Test Value = 6.07						
	t	df	Sig. (2-tailed)	Mean Difference	95% Confidence Interval of the Difference	
					Lower	Upper
IC50	85,366	2	,000	45,91000	43,5960	48,2240

Kesimpulan : Nilai sig. (2-tailed) yaitu $0,000 < 0,05$ yang berarti **terdapat perbedaan yang bermakna** antara nilai IC50 fisetin hasil praktikum dengan nilai IC50 formula II.

Lampiran 26. DPPH formula III (Compritol 2%)

Perhitungan fisetin dalam formula :

$$10\text{mg}/50\text{ ml} = 200\text{mg}/1000\text{ml} = 200\text{ppm}$$

Perhitungan konsentrasi (ppm) :

- 1) $V_1 \times N_1 = V_2 \times N_2$
 $200\text{ ppm} \times 2,5\text{ml} = V_2 \times 10\text{ml}$
 $V_2 = 50\text{ ppm}$
- 2) $V_1 \times N_1 = V_2 \times N_2$
 $200\text{ ppm} \times 1,25\text{ml} = V_2 \times 10\text{ml}$
 $V_2 = 25\text{ ppm}$
- 3) $V_1 \times N_1 = V_2 \times N_2$
 $200\text{ ppm} \times 0,625\text{ml} = V_2 \times 10\text{ml}$
 $V_2 = 12,5\text{ ppm}$
- 4) $V_1 \times N_1 = V_2 \times N_2$
 $200\text{ ppm} \times 0,156\text{ml} = V_2 \times 10\text{ml}$
 $V_2 = 3,125\text{ ppm}$
- 5) $V_1 \times N_1 = V_2 \times N_2$
 $200\text{ ppm} \times 0,078\text{ml} = V_2 \times 10\text{ml}$
 $V_2 = 1,56\text{ ppm}$

a. Replikasi I

Konsentrasi (ppm)	Absorbansi	% Inhibisi
50	0,488	46,4
25	0,627	31,1
12,5	0,700	23,0
3,13	0,749	17,7
1,56	0,759	16,6

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 15,5943$$

$$b = 0,6151$$

$$r = 0,99992$$

$$y = a + bx$$

$$y = 15,5943 + 0,6151x$$

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC50

$$50 = a + bx$$

$$50 = 15,5943 + 0,6151x$$

$$0,6151x = 34,4057$$

$$x = 55,94 \text{ ppm}$$

b. Replikasi II

Konsentrasi (ppm)	Absorbansi	% Inhibisi
50	0,479	47,3
25	0,627	31,1
12,5	0,700	23,0
3,13	0,759	16,6
1,56	0,767	15,7

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 14,6687$$

$$b = 0,6545$$

$$r = 0,99995$$

$$y = a + bx$$

$$y = 14,6687 + 0,6545x$$

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC50

$$50 = a + bx$$

$$50 = 14,6687 + 0,6545x$$

$$0,6545x = 35,3313$$

$$x = 53,98 \text{ ppm}$$

c. Replikasi III

Konsentrasi (ppm)	Absorbansi	% Inhibisi
50	0,489	46,2
25	0,628	31,0
12,5	0,700	23,0
3,13	0,749	17,7
1,56	0,759	16,6

Persamaan regresi linier antara absorbansi dan % inhibisi diperoleh nilai :

$$a = 15,5989$$

$$b = 0,6124$$

$$r = 0,99994$$

$$y = a + bx$$

$$y = 15,5989 + 0,6124x$$

keterangan :

x = konsentrasi (ppm)

$$y = 50$$

Perhitungan IC₅₀

$$50 = a + bx$$

$$50 = 15,5989 + 0,6124x$$

$$0,6124x = 34,4011$$

$$x = 56,17 \text{ ppm}$$

Rata-rata IC₅₀ Formula III :

$$\text{Replikasi I} = 55,94 \text{ ppm}$$

$$\text{Replikasi II} = 53,98 \text{ ppm}$$

$$\text{Replikasi III} = 56,17 \text{ ppm}$$

$$\left. \begin{array}{l} \\ \\ \end{array} \right\} 55,36 \text{ ppm}$$

Lampiran 27. Data statistik DPPH formula III (Compritol 2%)

```
T-TEST
  /TESTVAL=6.07
  /MISSING=ANALYSIS
  /VARIABLES=IC50
  /CRITERIA=CI (.95) .
```

→ T-Test

[DataSet0]

One-Sample Statistics

	N	Mean	Std. Deviation	Std. Error Mean
IC50	3	55,3633	1,20351	,69485

One-Sample Test

	Test Value = 6.07					
	t	df	Sig. (2-tailed)	Mean Difference	95% Confidence Interval of the Difference	
					Lower	Upper
IC50	70,941	2	,000	49,29333	46,3037	52,2830

Kesimpulan : Nilai sig. (2-tailed) yaitu $0,000 < 0,05$ yang berarti **terdapat perbedaan yang bermakna** antara nilai IC50 fisetin hasil praktikum dengan nilai IC50 formula III.