

Universitas
Esa Unggul

LAMPIRAN

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Lampiran 1. Sertifikat Analisis Enzim Tirosinase

SIGMA-ALDRICH sigmaaldrich.com


3050 Spruce Street, Saint Louis, MO 63103 USA
 Website: www.sigmaaldrich.com
 Email USA: techsup@sigma.com
 Outside USA: eurtechsup@sigma.com

Certificate of Analysis

Product Name: Tyrosinase from mushroom lyophilized powder > 1000 unit/mg solid

Product Number: T3824
Batch Number: SLBZ0022
Brand: SIGMA
CAS Number: 9002-10-2
MDL Number: MFCD00082116
Storage Temperature: Store at 20 °C
Quality Release Date: 01 OCT 2018
Recommended Re-test Date: OCT 2021
Storage Temperature: Store at 20 °C
Quality Release Date: 01 OCT 2018
Recommended Re-test Date: OCT 2021

Test	Specification	Result
units/mg Solid Tyrosinase Activity Unit Definition: One unit will cause an increase in A265 of 0.001 per minute at pH 6.5 at 25 deg C in a 3 mL reaction mix containing L-Tyrosine	≥ 1000	7164
units/mg Solid Polyphenol Oxidase Activity Unit Definition: One unit will cause a decrease in A265 of 0.001 per minute at pH 6.5 at 25 deg C in a 3 mL reaction mixture containing L-Beta-3,4-dihydroxyphenylalanine (L-DOPA)	≥ 30000	149192
units/mg Solid Catechol Oxidase Activity Unit Definition: One unit will cause a decrease in A265 of 0.001 per minute at pH 6.5 at 25 deg C in a 3 mL reaction mixture containing catechol and ascorbic acid	≥ 30000	1431095



Rodney Rurbach, Manager
Analytical Services
St. Louis, Missouri US

Sigma-Aldrich warrants, that at the time of the quality release or subsequent retest date this product conformed to the information contained in this publication. The current specification sheet may be available at Sigma-Aldrich.com. For further inquiries, please contact Technical Service. Purchaser must determine the suitability of the product for its particular use. See reverse side of invoice or packing slip for additional terms and conditions of sale.

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Lampiran 2. Sertifikat Analisis Substrat L-DOPA

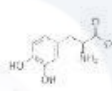
SIGMA-ALDRICH

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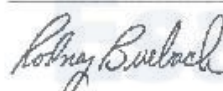
Certificate of Analysis

Product Name:
3,4-Dihydroxy-L-phenylalanine - ≥98% (TLC)

Product Number: D9628
 Batch Number: SLES0617V
 Brand: SIGMA
 CAS Number: 59-92-7
 MDL Number: MFCD00002598
 Formula: C₉H₁₁NO₄
 Formula Weight: 197.19 g/mol
 Quality Release Date: 12 SEP 2018



Test	Specification	Result
Appearance (Color)	White to Off-White	Off-White
Appearance (Form)	Powder	Powder
Solubility (Color)	Colorless to Light Yellow	Faint Yellow
Solubility (Turbidity)	Clear to Slightly Hazy	Clear
50 mg/mL, 0.5 M HCl		
Infrared Spectrum	Conforms to Structure	Conforms
Carbon	53.7 - 55.9 %	54.5 %
Nitrogen	5.6 - 7.4 %	7.1 %
Specific Rotation	-12.5 - -10.5 °	-12.4 °
C = 1 in 1 M HCl, 25 deg C		
Purity (TLC)	≥ 98 %	100 %




Rodney Bueloch, Manager
 Analytical Services
 St. Louis, Missouri, US

Sigma-Aldrich warrants, that at the time of the quality release or subsequent retest date this product conformed to the information contained in this publication. The current Specification sheet may be available at Sigma-Aldrich.com. For further inquiries, please contact Technical Service. Purchaser must determine the suitability of the product for its particular use. See reverse side of invoice or packing slip for additional terms and conditions of sale.

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Lampiran 3. Sertifikat Analisis Asam Kojat



The Power's Question

CERTIFICATE OF ANALYSIS

Catalog Number:	sc-255228
Lot Number:	E1517
Product Name:	Kojic acid
CAS Number:	501-30-4
Molecular Formula:	$C_6H_6O_4$
Molecular Weight:	142.11

Test	Result
Appearance	Pale yellow crystalline powder
Purity	99.8%
Melting Point	152.8 - 154.7 °C
Residue on Ignition	0.04%
Heavy Metals (Pb)	< 20 ppm
Loss on Drying	0.37%
Arsenic	< 2 ppm

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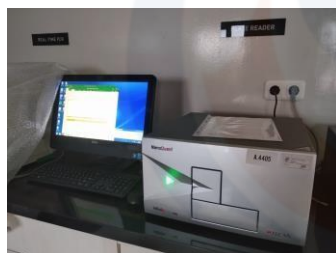
Lampiran 4. Alat dan Bahan



Timbangan



Ph Meter



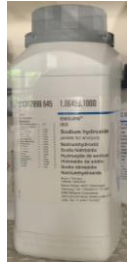
Spektrofotometer UV-Vis



Neraca Analitik



Aquadest



NaOH



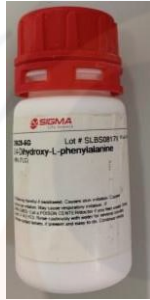
KH₂PO₄



Enzim Tirosinase



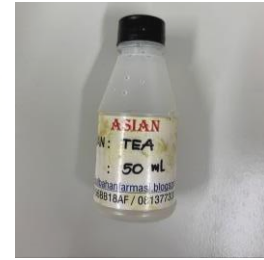
Asam Kojat



L-DOPA



Propilenglikol



TEA

Metil Paraben



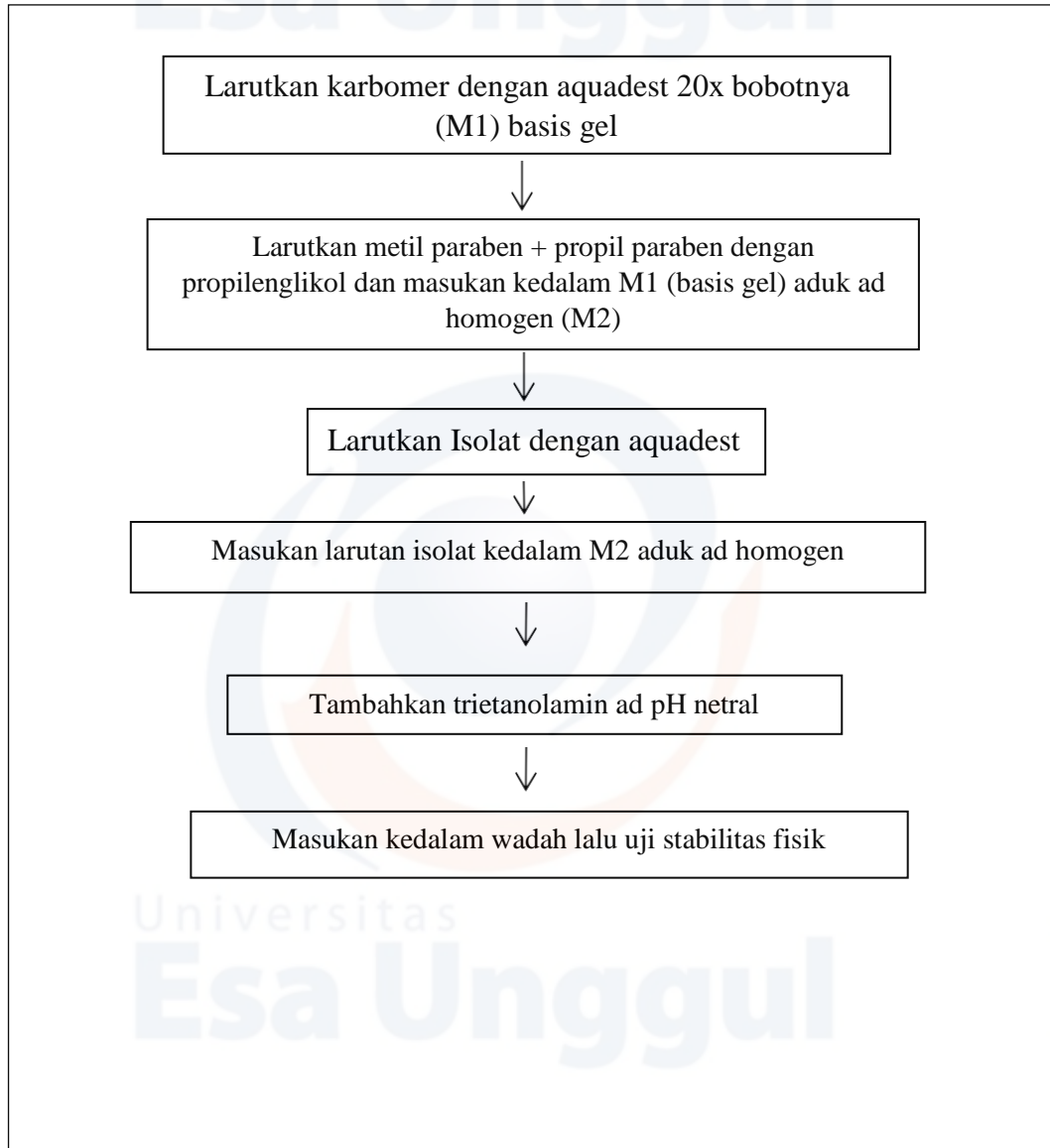
Propilen Paraben



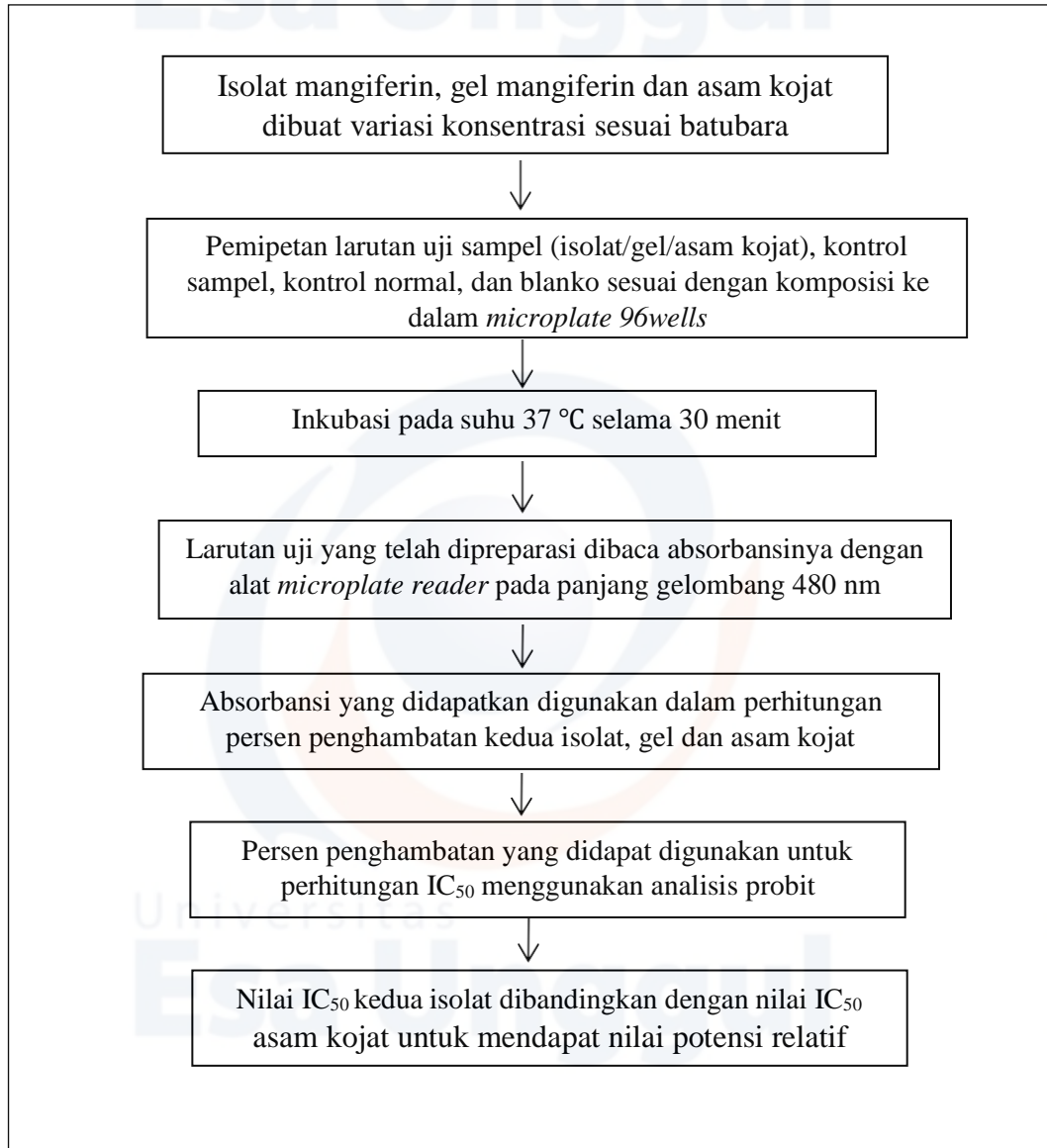
Karbomer



Lampiran 5. Skema Pembuatan Gel Isolat Mangiferin



Lampiran 6. Skema Uji Aktivitas Penghambatan Enzim Tirosinase



Lampiran 7. Perhitungan Sediaan Gel Isolat Mangiferin

Perhitungan bahan:

F1 = Isolat Mangiferin	$= \frac{1,25 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 1,25 \text{ gram}$
Karbomer	$= \frac{1 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 1 \text{ gram}$
Trietanolamin	= ad pH netral
Propilenglikol	$= \frac{5 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 5 \text{ gram}$
Metil paraben	$= \frac{0,18 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 0,18 \text{ gram}$
Propil paraben	$= \frac{0,02 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 0,02 \text{ gram}$
Air suling ad	$= 100 - (1,25+1+0+5+0,18+0,02)$ $= 92,55 \text{ gram}$
F2 = Isolat Mangiferin	$= \frac{2,5 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 2,5 \text{ gram}$
Karbomer	$= \frac{1 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 1 \text{ gram}$
Trietanolamin	= ad pH netral
Propilenglikol	$= \frac{5 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 5 \text{ gram}$
Metil paraben	$= \frac{0,18 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 0,18 \text{ gram}$
Propil paraben	$= \frac{0,02 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 0,02 \text{ gram}$
Air suling ad	$= 100 - (2,5+1+0+5+0,18+0,02)$ $= 91,3 \text{ gram}$

Perhitungan bahan:

F3 = Isolat mangiferin	$= \frac{5 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 5 \text{ gram}$
Karbomer	$= \frac{1 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 1 \text{ gram}$
Trietanolamin	= ad pH netral
Propilenglikol	$= \frac{5 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 5 \text{ gram}$
Metil paraben	$= \frac{0,18 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 0,18 \text{ gram}$
Propil paraben	$= \frac{0,02 \text{ gr}}{100 \text{ gr}} \times 100 \text{ gr} = 0,02 \text{ gram}$
Air suling ad	$= 100 - (5+1+0+5+0,18+0,02)$ $= 88,8 \text{ gram}$

Lampiran 8. Perhitungan Uji Sineresis Pada Evaluasi Fisik Sediaan Gel

Rumus: Sineresis (%) = $\frac{\text{berat awal} - \text{berat akhir}}{\text{berat awal}} \times 100\%$

24 jam :

$$F1 = \text{Sineresis (\%)} = \frac{55,1836 - 55,1003}{55,1836} \times 100\% = 0,15\%$$

$$F2 = \text{Sineresis (\%)} = \frac{52,8242 - 52,7435}{52,8242} \times 100\% = 0,15\%$$

$$F3 = \text{Sineresis (\%)} = \frac{49,9117 - 49,8828}{49,9117} \times 100\% = 0,05\%$$

48 jam :

$$F1 = \text{Sineresis (\%)} = \frac{55,1003 - 55,0440}{55,1003} \times 100\% = 0,10\%$$

$$F2 = \text{Sineresis (\%)} = \frac{52,7435 - 52,6764}{52,7435} \times 100\% = 0,12\%$$

$$F3 = \text{Sineresis (\%)} = \frac{49,8828 - 49,8011}{49,8828} \times 100\% = 0,16\%$$

72 jam :

$$F1 = \text{Sineresis (\%)} = \frac{55,0440 - 54,8493}{55,0440} \times 100\% = 0,35\%$$

$$F2 = \text{Sineresis (\%)} = \frac{52,6764 - 52,4252}{52,6764} \times 100\% = 0,47\%$$

$$F3 = \text{Sineresis (\%)} = \frac{49,7456 - 49,5363}{49,7456} \times 100\% = 0,42\%$$

Lampiran 9. Perhitungan Larutan Substrat L-DOPA 2 mM

Konsentrasi Substrat L-DOPA 2 mM :

$$M = \frac{\text{gram}}{\text{BM}} \times \frac{1000 \text{ ml}}{V}$$

Diketahui :

M : Molaritas (2 mM)

BM Substrat L-DOPA = 197,19

V = volume yang diinginkan (10 ml)

$$\begin{aligned} \text{Bobot substrat L-DOPA yang harus ditimbang} &= \frac{M \times \text{BM} \times V}{1000 \text{ ml}} = \frac{2 \text{ mM} \times 197,19 \times 10 \text{ ml}}{1000 \text{ ml}} \\ &= 3,94 \text{ mg} \end{aligned}$$

Lampiran 10. Perhitungan Larutan Enzim Tirosinase 33 Unit/ml

- a. Pembuatan larutan induk enzim tirosinase:

Diketahui:

Aktivitas enzim dalam sediaan 7164 Unit/mg

Aktivitas induk yang akan dibuat 716,4 Unit/ml

Volume larutan enzim yang akan dibuat 10 ml

Perhitungan larutan induk enzim tirosinase:

$$\text{Unit/ml} = \frac{\text{bobot (mg)} \times \text{Unit/mg}}{\text{volume (ml)}}$$

$$\text{Bobot enzim yang ditimbang} = \frac{716,4 \frac{\text{Unit}}{\text{ml}} \times 10 \text{ ml}}{7164 \frac{\text{Unit}}{\text{mg}}} = 1 \text{ mg}$$

- b. Pembuatan larutan enzim tirosinase 333 Unit/ml

Diketahui:

Aktivitas induk enzim tirosinase 7164 Unit/ml

Aktivitas enzim yang akan dibuat dalam pengujian 333 Unit/ml

Volume larutan enzim yang akan dibuat dalam pengujian 10 ml

Perhitungan pengenceran larutan enzim tirosinase dalam pengujian:

$$= \frac{333 \frac{\text{Unit}}{\text{ml}}}{7164 \frac{\text{Unit}}{\text{ml}}} \times 10 \text{ ml} = 4,6 \text{ ml (yang dipipet dari larutan baku)}$$

Lampiran 11. Perhitungan Pembuatan Larutan Asam Kojat

1. Pembuatan larutan induk asam kojat 500 µg/ml

Diketahui :

Konsentrasi larutan induk asam kojat yang akan dibuat = 500 µg/ml

Volume larutan induk yang akan dibuat = 10 ml

Perhitungan bobot asam kojat yang ditimbang :

$$\frac{500 \mu\text{g}}{\text{ml}} \times 10 \text{ ml} = 5000 = 5 \text{ mg}$$

2. Pembuatan variasi konsentrasi larutan asam kojat 250, 125, 62,5, 31,25, 15,625 dan 7,8125 µg/ml dalam labu ukur 10 ml

a. $250 \mu\text{g/ml} = \frac{250 \mu\text{g/ml}}{500 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 500 µg/ml

b. $125 \mu\text{g/ml} = \frac{125 \mu\text{g/ml}}{250 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 250 µg/ml

c. $62,5 \mu\text{g/ml} = \frac{62,5 \mu\text{g/ml}}{125 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 125 µg/ml

d. $31,25 \mu\text{g/ml} = \frac{31,25 \mu\text{g/ml}}{62,5 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 62,5

µg/ml

e. $15,625 \mu\text{g/ml} = \frac{15,625 \mu\text{g/ml}}{31,25 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 31,25

µg/ml

f. $7,8125 \mu\text{g/ml} = \frac{7,8125 \mu\text{g/ml}}{15,625 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi

15,625 µg/ml

Seri konsentrasi yang digunakan dalam pengujian adalah konsentrasi 7,8125 – 125 µg/ml

Lampiran 12. Perhitungan Pembuatan Larutan Isolat Mangiferin

1. Pembuatan larutan induk isolat Mangiferin
 Konsentrasi larutan induk isolat Mangiferin yang akan dibuat 2000 $\mu\text{g/ml}$
 Volume larutan induk yang akan dibuat = 10 ml
 Perhitungan seri konsentrasi isolat yang ditimbang

$$\frac{2000 \mu\text{g}}{\text{ml}} \times 10 \text{ ml} = 20000 \mu\text{g} = 20 \text{ mg}$$
2. Pembuatan variasi konsentrasi isolat Mangiferin 2000, 1000, 500, 250 dan 125 $\mu\text{g/ml}$ dalam labu ukur
 - a. 2000 $\mu\text{g/ml}$ = sesuai pada pembuatan larutan induk
 - b. 1000 $\mu\text{g/ml}$ = $\frac{1000 \mu\text{g/ml}}{2000 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 2000 $\mu\text{g/ml}$
 - c. 500 $\mu\text{g/ml}$ = $\frac{500 \mu\text{g/ml}}{1000 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 1000 $\mu\text{g/ml}$
 - d. 250 $\mu\text{g/ml}$ = $\frac{250 \mu\text{g/ml}}{500 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 500 $\mu\text{g/ml}$
 - e. 125 $\mu\text{g/ml}$ = $\frac{125 \mu\text{g/ml}}{250 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 250 $\mu\text{g/ml}$
 - f. 62,5 $\mu\text{g/ml}$ = $\frac{62,5 \mu\text{g/ml}}{125 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 125 $\mu\text{g/ml}$
 - g. 31,25 $\mu\text{g/ml}$ = $\frac{31,25 \mu\text{g/ml}}{62,5 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 62,5 $\mu\text{g/ml}$
 - h. 15,625 $\mu\text{g/ml}$ = $\frac{15,625 \mu\text{g/ml}}{31,25 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 31,25 $\mu\text{g/ml}$
 - i. 7,8125 $\mu\text{g/ml}$ = $\frac{7,8125 \mu\text{g/ml}}{15,625 \mu\text{g/ml}} \times 10 \text{ ml} = 5 \text{ ml}$, diambil dari konsentrasi 15,625 $\mu\text{g/ml}$

Lampiran 13. Pemetaan Sumuran Saat Uji Aktivitas Inhibitor Tirosinase

	1	2	3	4	5	6	7	8	9	10	11	12
A	○	○	○	○	○	○	○	○	○	○	○	○
B	○	○	○	○	○	○	○	○	○	○	○	○
C	○	○	○	○	○	○	○	○	○	○	○	○
D	○	○	○	○	○	○	○	○	○	○	○	○
E	○	○	○	○	○	○	○	○	○	○	○	○
F	○	○	○	○	○	○	○	○	○	○	○	○
G	○	○	○	○	○	○	○	○	○	○	○	○
H	○	○	○	○	○	○	○	○	○	○	○	○

Keterangan :

A9 : Sampel (Ekstrak Buah Mahkota Dewa)

B9 : Kontrol Sampel (Ekstrak)

C9 : Sampel (Gel F1)

D9 : Kontrol Sampel (Gel F1)

E9 : Sampel (Gel F2)

F9 : Kontrol Sampel (Gel F2)

G9: Sampel (Gel F3)

H9 : Kontrol Sampel (Gel F3)

A10-G10 : Asam Kojat

A11-G11 : Kontrol Asam Kojat

A12-C12 : Blanko

D12-F12 : Kontrol Blanko

Lampiran 14. Hasil Uji Aktivitas Enzim Tirosinase Asam Kojat

B	KB	(B-KB)	Konsentrasi	S	KS	Rata-Rata (S-KS)	Persen Inhibisi (%)
0,1121	0,0419	0,0702	125	0,0606 0,0607 0,0609	0,0481 0,0479 0,0476	0,0129	88,40%
0,2404	0,0495	0,1945	62,5	0,0668 0,0661 0,0675	0,0517 0,0520 0,0523	0,0148	86,70%
0,1115	0,0422	0,0693	31,25	0,0670 0,0631 0,0695	0,0425 0,0419 0,0418	0,0262	76,46%
			15,625	0,0702 0,0723 0,0731	0,0421 0,0426 0,0423	0,0298	73,22%
			7,8125	0,0946 0,0997 0,1010	0,0482 0,0485 0,0480	0,0497	55,34%
Rata-rata : 0,1113							

Keterangan :

- B : Absorbansi larutan blanko (dengan enzim)
 KB : Absorbansi larutan kontrol blanko (tanpa enzim)
 S : Absorbansi larutan sampel (dengan enzim)
 KS : Absorbansi larutan kontrol sampel (tanpa enzim)

Lampiran 15. Perhitungan Persen Inhibisi dan IC₅₀ Asam Kojat

1. Persen Inhibisi

$$\text{Persen Penghambatan (\%)} = \frac{A1-A2}{A1} \times 100\%$$

- a. Konsentrasi 125 µg/ml = $\frac{0,1113-0,0129}{0,1113} \times 100\% = 88,40\%$
- b. Konsentrasi 62,5 µg/ml = $\frac{0,1113-0,0148}{0,1113} \times 100\% = 86,70\%$
- c. Konsentrasi 31,25 µg/ml = $\frac{0,1113-0,0262}{0,1113} \times 100\% = 76,46\%$
- d. Konsentrasi 15,625 µg/ml = $\frac{0,1113-0,0298}{0,1113} \times 100\% = 73,22\%$
- e. Konsentrasi 7,8125 µg/ml = $\frac{0,1113-0,0497}{0,1113} \times 100\% = 55,34\%$

2. IC₅₀

Konsentrasi (ppm)	X (Log Kons)	Persen Penghambatan (%)	Y (Probit)	IC ₅₀
7,8125	0,8927	55,3459	5,1332	
15,625	1,1938	73,2255	5,6189	
31,25	1,4948	76,4600	5,7192	4,22
62,5	1,7958	86,7026	6,1123	
125	2,0969	88,4097	6,1952	

a = 4,4561
 b = 0,8695
 r = 0,9701

Regresi Linier = Y = a+bx

Y = a + bx

5 = 4,4561 + 0,8695 x

$x = \frac{5-4,4561}{0,8695}$

x = 0,6255

Anti log = 4,2218 IC₅₀

Lampiran 16. Hasil Uji Aktivitas Enzim Isolat Mangiferin

B	KB	(B-KB)	Konsentrasi	S	KS	Rata-Rata (S-KS)	Persen Inhibisi (%)
0,2431	0,0401	0,2030	2000	0,0637	0,0549	0,0115	94,29%
0,2410	0,0403	0,2007		0,0643	0,0500		
0,2418	0,0409	0,2009	1000	0,0711	0,0582	0,0117	94,19%
			500	0,0709	0,0548	0,0306	84,81%
				0,0851	0,0551		
			250	0,0857	0,0545	0,0309	84,66%
				0,0943	0,0640		
			125	0,0951	0,0635	0,0462	77,07%
				0,1158	0,0684		
Rata-rata : 0,2015				0,1132	0,0682		

Keterangan :

- B : Absorbansi larutan blanko (dengan enzim)
 KB : Absorbansi larutan kontrol blanko (tanpa enzim)
 S : Absorbansi larutan sampel (dengan enzim)
 KS : Absorbansi larutan kontrol sampel (tanpa enzim)

Lampiran 17. Perhitungan Persen Inhibisi dan Nilai IC₅₀ Isolat Mangiferin

1. Persen Inhibisi

$$\text{Persen Penghambatan (\%)} = \frac{A1-A2}{A1} \times 100\%$$

a. Konsentrasi 2000 µg/ml = $\frac{0,2015-0,0115}{0,2015} \times 100 \% = 94,29 \%$

b. Konsentrasi 1000 µg/ml = $\frac{0,2015-0,0117}{0,2015} \times 100 \% = 94,19 \%$

c. Konsentrasi 500 µg/ml = $\frac{0,2015-0,0306}{0,2015} \times 100 \% = 84,81 \%$

d. Konsentrasi 250 µg/ml = $\frac{0,2015-0,0309}{0,2015} \times 100 \% = 84,66 \%$

e. Konsentrasi 125 µg/ml = $\frac{0,2015-0,0462}{0,2015} \times 100 \% = 77,07 \%$

2. IC₅₀

Konsentrasi (ppm)	Log Kons (X)	Penghambatan inhibisi %	Y (probit)	IC ₅₀
2000	1,9744	92,29 %	6,5805	6,5147
1000	1,9740	94,19 %	6,5718	
500	1,9284	84,81 %	6,0279	
250	1,9276	84,66 %	6,0237	
125	1,8868	77,07 %	5,7424	

a = 13,1

b = 9,9519

r = 0,9755

Regresi Linier = Y = a+bx

Y = a + bx

5 = 13,1 + 9,9519 x

x = $\frac{5-13,1}{9,9519}$

x = 0,8139

Anti log = 6,5147IC₅₀

Lampiran 18. Hasil Uji Aktivitas Enzim Tirosinase Formula I

B	KB	(B-KB)	Konsentrasi	S	KS	Rata-Rata (S-KS)	Persen Inhibisi (%)
0,2431	0,0401	0,2030	2000	0,0696	0,0501	0,0189	90,62%
0,2410	0,0403	0,2007		0,0695	0,0511		
0,2418	0,0409	0,2009	1000	0,0703	0,0457	0,0260	87,09%
			500	0,0700	0,0453	0,0296	85,31%
				0,0712	0,0418		
			250	0,0717	0,0419	0,0313	84,46%
				0,0715	0,0397		
			125	0,0703	0,0395	0,0339	83,17%
				0,0742	0,0402		
				0,0740	0,0401		
Rata-rata : 0,2015							

Keterangan :

- B : Absorbansi larutan blanko (dengan enzim)
 KB : Absorbansi larutan kontrol blanko (tanpa enzim)
 S : Absorbansi larutan sampel (dengan enzim)
 KS : Absorbansi larutan kontrol sampel (tanpa enzim)

Lampiran 19. Perhitungan Persen Inhibisi dan Nilai IC₅₀ Formula I

1. Persen Inhibisi

Persen Penghambatan :

$$\text{Persen Penghambatan (\%)} = \frac{A1-A2}{A1} \times 100\%$$

a. Konsentrasi 15,625 µg/ml = $\frac{0,2015-0,0189}{0,2015} \times 100\% = 90,62\%$

b. Konsentrasi 31,25 µg/ml = $\frac{0,2015-0,0260}{0,2015} \times 100\% = 87,09\%$

c. Konsentrasi 62,5 µg/ml = $\frac{0,2015-0,0296}{0,2015} \times 100\% = 85,31\%$

d. Konsentrasi 125 µg/ml = $\frac{0,2015-0,0313}{0,2015} \times 100\% = 84,46\%$

e. Konsentrasi 250 µg/ml = $\frac{0,2015-0,0339}{0,2015} \times 100\% = 83,17\%$

2. Nilai IC₅₀

Konsentrasi (ppm)	Log Kons (X)	Persen Penghambatan	Probit (Y)	IC ₅₀
2000	1,9572	90,62 %	6,3165	5,8979
1000	1,9399	87,09 %	6,1264	
500	1,9309	85,31 %	6,0494	
250	1,9266	84,46 %	6,0152	
125	1,9199	83,17 %	5,9624	

a: 12,3445

b: 9,5294

r: 0,9933

$$Y = a + bx$$

$$5 = 12,3445 + 9,5294 x$$

$$x = \frac{5-12,3445}{9,5294}$$

$$x = 0,7707$$

$$\text{Anti log} = 5,8979 \text{ IC}_{50}$$

Lampiran 20. Hasil Uji Aktivitas Enzim Tirosinase Formula II

B	KB	(B-KB)	Konsentrasi	S	KS	Rata-Rata (S-KS)	Persen Inhibisi (%)
0,2431	0,0401	0,2030	2000	0,0602	0,0407	0,0203	89,92%
0,2410	0,0403	0,2007		0,0616	0,0405		
0,2418	0,0409	0,2009	1000	0,0695	0,0471	0,0224	88,88%
			500	0,0692	0,0468	0,0232	88,48%
				0,0675	0,0450		
			250	0,0682	0,0443	0,0285	85,85%
				0,0712	0,0422		
			125	0,0701	0,0421	0,0300	85,11%
				0,0717	0,0414		
Rata-rata : 0,2015				0,0707	0,0409		

Keterangan :

- B : Absorbansi larutan blanko (dengan enzim)
 KB : Absorbansi larutan kontrol blanko (tanpa enzim)
 S : Absorbansi larutan sampel (dengan enzim)
 KS : Absorbansi larutan kontrol sampel (tanpa enzim)

Lampiran 21. Perhitungan Persen Inhibisi dan Nilai IC₅₀ Formula II

1. Persen Inhibisi

Persen Penghambatan :

$$\text{Persen Penghambatan (\%)} = \frac{A1-A2}{A1} \times 100\%$$

a. Konsentrasi 2000 µg/ml = $\frac{0,2015-0,0203}{0,2015} \times 100 \% = 89,92 \%$

b. Konsentrasi 1000 µg/ml = $\frac{0,2015-0,0224}{0,2015} \times 100 \% = 88,88 \%$

c. Konsentrasi 500 µg/ml = $\frac{0,2015-0,0232}{0,2015} \times 100 \% = 88,84 \%$

d. Konsentrasi 250 µg/ml = $\frac{0,2015-0,0285}{0,2015} \times 100 \% = 85,85 \%$

e. Konsentrasi 125 µg/ml = $\frac{0,2015-0,0300}{0,2015} \times 100 \% = 85,11 \%$

2. Nilai IC₅₀

Konsentrasi (ppm)	Log Kons (X)	Persen Penghambatan	Probit (Y)	IC ₅₀
2000	1,9538	89,92 %	6,2750	
1000	1,9488	88,88 %	6,2212	
500	1,9468	88,48 %	6,2004	6,2230
250	1,9337	85,85 %	6,0758	
125	1,9299	85,11 %	6,0407	

a = 12,7175

b = 9,7189

r = 0,9993

Y = a + bx

5 = 12,7175 + 9,7189 x

x = $\frac{5-12,7175}{9,7189}$

x = 0,7940

Anti log = 6,2230 IC₅₀

Lampiran 22. Hasil Uji Aktivitas Enzim Tirosinase Formula III

B	KB	(B-KB)	Konsentrasi	S	KS	Rata-Rata (S-KS)	Persen Inhibisi (%)
0,2431	0,0401	0,2030	2000	0,0602	0,0413	0,0196	90,27%
0,2410	0,0403	0,2007		0,0616	0,0412		
0,2418	0,0409	0,2009	1000	0,0681	0,0444	0,0243	87,94%
			500	0,0692	0,0443	0,0256	87,29%
				0,0822	0,0537		
			250	0,0761	0,0533	0,0266	86,79%
				0,0741	0,0475		
			125	0,0741	0,0474	0,0291	85,55%
				0,0709	0,0419		
				0,0709	0,0416		
Rata-rata : 0,2015							

Keterangan :

- B : Absorbansi larutan blanko (dengan enzim)
 KB : Absorbansi larutan kontrol blanko (tanpa enzim)
 S : Absorbansi larutan sampel (dengan enzim)
 KS : Absorbansi larutan kontrol sampel (tanpa enzim)

Lampiran 23. Perhitungan Persen Inhibisi dan Nilai IC₅₀ Formula III

1. Persen Inhibisi

Persen Penghambatan :

$$\text{Persen Penghambatan (\%)} = \frac{A1-A2}{A1} \times 100\%$$

a. Konsentrasi 2000 µg/ml = $\frac{0,2015-0,0196}{0,2015} \times 100\% = 90,27\%$

b. Konsentrasi 1000 µg/ml = $\frac{0,2015-0,0243}{0,2015} \times 100\% = 87,94\%$

c. Konsentrasi 500 µg/ml = $\frac{0,2015-0,0256}{0,2015} \times 100\% = 87,29\%$

d. Konsentrasi 250 µg/ml = $\frac{0,2015-0,0266}{0,2015} \times 100\% = 86,79\%$

e. Konsentrasi 125 µg/ml = $\frac{0,2015-0,0291}{0,2015} \times 100\% = 85,55\%$

2. IC₅₀

Konsentrasi (ppm)	Log Kons (X)	Penghambatan inhibisi %	Y (probit)	IC ₅₀
2000	1,9555	90,27 %	6,2988	7,0696
1000	1,9441	87,94 %	6,1700	
500	1,9409	87,29 %	6,1407	
250	1,9384	86,79 %	6,1170	
125	1,9322	85,55 %	6,0625	

a = 13,6734

b = 10,2106

r = 0,9953

Y = a + bx

5 = 13,6734 + 10,2106 x

$$x = \frac{5-13,6734}{10,2106}$$

x = 0,8494

Anti log = 7,0696 IC₅₀

Lampiran 24. Potensi Relatif

$$\text{Potensi Relatif} = \frac{\text{Nilai IC}_{50} \text{ asam Kojat}}{\text{Nilai IC}_{50} \text{ isolat mangiferin}}$$

Diketahui :

$$\text{Nilai IC}_{50} \text{ asam kojat} = 4,22 \mu\text{g/ml}$$

$$\text{Nilai IC}_{50} \text{ isolat mangiferin} = 6,5147 \mu\text{g/ml}$$

Potensi Relatif isolat mangiferin

$$= \frac{4,22 \mu\text{g/ml}}{6,5147 \mu\text{g/ml}}$$

$$= 0,6477 \mu\text{g/ml}$$

Lampiran 25. Dokumentasi

