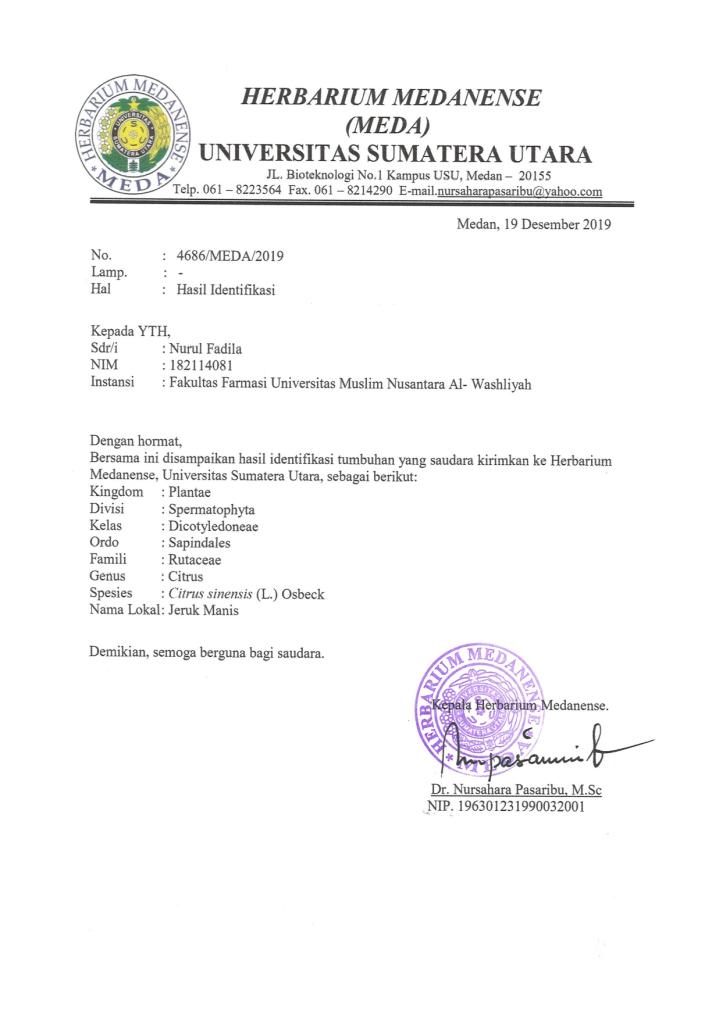
**Lampiran 1.** Surat Hasil Determinasi Tanaman Jeruk Manis (*Citrus sinensis* L.)



**Lampiran 2.** Bagan Alir Prosedur Kerja

Pengumpulan sampel kulit jeruk manis (*Citrus sinensis* L.)

Kulit jeruk manis segar

Disortasi basah

Dicuci dengan air kran mengalir

Ditiriskan

Dirajang tipis-tipis

Ditimbang

Berat basah kulit jeruk manis

Dikeringkan dalam lemari pengering dengan suhu ±40oC

Disortasi kering

Ditimbang kembali

Berat kering simplisia kulit jeruk manis

Dihaluskan menggunakan blender

Disimpan dalam wadah tertutup rapat

Berat serbuk simplisia kulit jeruk manis

Karakterisasi simplisia

**Lampiran 3.** Bagan Alir Karakterisasi Simplisia Kulit Jeruk Manis (*citrus sinensis L.*)

Simplisia kulit jeruk manis segar

Pemeriksaan Makroskopik

Pemeriksaan Mikroskopis

* Kadar air
* Kadar sari larut air
* Kadar sari larut etanol
* Kadar abu total
* Kadar abu tidak larut asam

**Lampiran 4**. Bagan Alir Ekstraksi Serbuk Simplisia Kulit Jeruk Manis (*Citrus sinensis* L.)

Serbuk simplisia kulit jeruk manis

Ditimbang serbuk simplisia 500 gram

Dimasukkan kedalam bejana maserasi

Ditambahkan 75 bagian pelarut yaitu 3750 ml etanol 96%

Didiamkan selama 5 hari sambil diaduk

Disaring

Ampas

Maserat

Ditambahkan 25 bagian pelarut yaitu 1250 ml etanol 96% (sampai 5000 ml)

Diendapkan 2 hari

Maserat

Maserat yang diperoleh di uapkan menggunakan Rotary evavorator

Ekstrak kental kulit jeruk manis

**Lampiran 5**. Bagan Alir Skrining Fitokimia Simplisia dan Ekstrak Kulit Jeruk Manis (*Citrus sinensis* L.)

Ekstrak kulit jeruk manis

Serbuk simplisia kulit jeruk manis

Skrining fitokimia

Golongan tanin

Golongan saponin

Golongan flavonoid

Golongan alkaloid

Golongan glikosida

Golongan triterpenoid/steroid

Golongan glikosida antrakinon

**Lampiran 6**. Bagan Alir Uji Sitotoksisitas Ekstrak Kulit Jeruk Manis (*Citrus sinensis* L.)

50 mg ekstrak kulit jeruk manis

Dilarutkan dengan air garam 10 ml

Konsentrasi 5000 µg/ml (LIB 1)

Dibuat dalam beberapa variasi konsetrasi

100 µg/ml

200 µg/ml

300 µg/ml

400 µg/ml

kontrol

150 µg/ml

500 µg/ml

350 µg/ml

250 µg/ml

Disiapkan vial yang telah berisi 10 ekor larva artemia salina dalam 10 ml ekstrak yang sudah dilarutkan dengan air garam

Masing-masing konsentrasi dibuat dengan 3 kali pengulangan

Mortalitas dihitung setelah 24 jam

% Mortalitas

**Lampiran 7.** Pengelolaan Sampel Kulit Jeruk Manis (*Citrus sinensis* L.)

Perajangan

Pencucian

Sortasi basah

Pengayakan

Penghalusan

Pengeringan

**Lampiran 8**. Perhitungan Susut Pengeringan

Diketahui :

Bobot tumbuhan segar = 10.000 gram

Bobot simplisia = 1200 gram

% susut pengeringan = x 100%

% susut pengeringan = x 100%

= 88 %

**Lampiran 9.** Proses Ekstraksi Kulit Jeruk Manis (*Citrus sinensis* L.)

Penyaringan larutan

Wadah maserasi

Serbuk simplisia kulit jeruk manis

Pengentalan ektrak kulit jeruk manis

Hasil maserat kulit jeruk manis

Ekstrak kental kulit jeruk manis

**Lampiran 10**. Perhitungan Randemen Ekstrak

Diketahui :

Bobot serbuk simplisia = 500 gram

Volume pelarut = 5000 ml

Bobot ekstrak = 98,594 gram

% Rendemen ekstrak = x 100 %

% Rendemen ekstrak = x 100 %

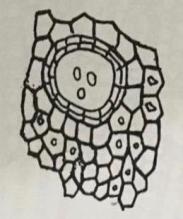
= 19,72 %

**Lampiran 11**. Pemeriksaan Mikroskopik Simplisia Kulit Jeruk Manis (*Citrus sinensis* L.)



1

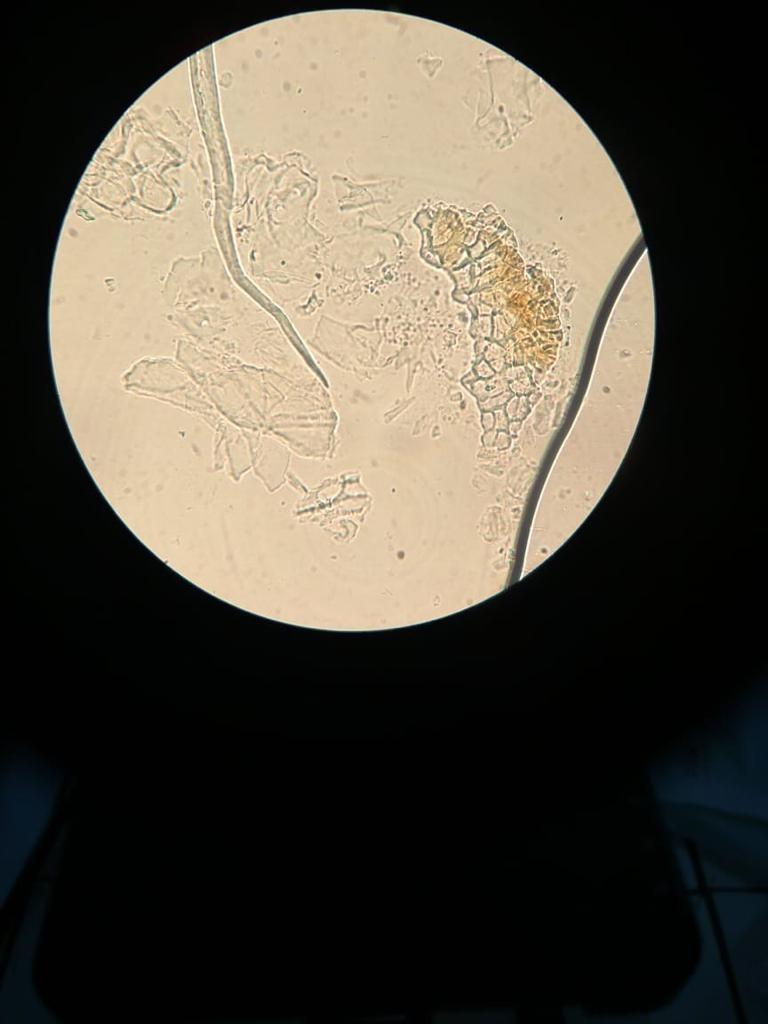
1

2

2

3



3

Keterangan hasil :

1. Minyak atsiri
2. Epidermis
3. Kristal kalsium oksalat berbentuk prisma

**Lampiran 12**. Perhitungan Penetapan Kadar Air Simplisia Kulit Jeruk Manis (*Citrus sinensis* L.)

% kadar air = x 100%

Keterangan : V0 = Volume destilasi dari penjenuhan toluen

V1 = Volume destilasi air dari simplisia

1. Sampel pengulangan I

V0 = 1,2 ml

V1 = 1,6 ml

Berat simplisia = 5 gram

% kadar air = x 100% = 8 %

1. Sampel pengulangan II

V0 = 1,5 ml

V1 = 1,8 ml

Berat simplisia = 5 gram

% kadar air = x 100% = 6 %

1. Sampel pengulangan III

V0 = 1,3 ml

V1 = 1,6 ml

Berat simplisia = 5 gram

% kadar air = x 100% = 6 %

Kadar air rata-rata = = 6,66%

**Lampiran 13**. Perhitungan Penetapan Kadar Sari Larut Air dan Kadar Sari Larut

Etanol Simplisia Kulit Jeruk Manis (*Citrus sinensis* L.)

1. Perhitungan penetapan kadar sari larut air

% kadar = x 100%

1. Pengulangan 1

Berat sampel = 5 gram

Berat cawan kosong = 31,69 gram

B1 = 31,92 gram

B2 = 31,90 gram

B3 = 31,90 gram

Brata-rata = 31,90 gram

% kadar = x 100% = 21 %

1. Pengulangan 2

Berat sampel = 5 gram

Berat cawan kosong = 32,16 gram

B1 = 32,36 gram

B2 = 32,37 gram

B3 = 32,37 gram

Brata-rata = 32,36 gram

% kadar = x 100% = 20 %

**Lampiran 13. (**Lanjutan)

1. Pengulangan 3

Berat sampel = 5 gram

Berat cawan kosong = 32,27 gram

B1 = 32,52 gram

B2 = 32,50 gram

B3 = 32,50 gram

Brata-rata = 32,50 gram

% kadar = x 100% = 23 %

Kadar sari larut air rata-rata = = 21,33 %

1. Perhitungan penetapan kadar sari larut etanol

% kadar = x 100%

1. Pengulangan 1

Berat sampel = 5 gram

Berat cawan kosong = 31,83 gram

B1 = 32,07 gram

B2 = 32,06 gram

B3 = 32,06 gram

Brata-rata = 32,06 gram

% kadar = x 100% = 23 %

**Lampiran 13. (**Lanjutan)

1. Pengulangan 2

Berat sampel = 5 gram

Berat cawan kosong = 32,09 gram

B1 = 32,35 gram

B2 = 32,38 gram

B3 = 32,38 gram

Brata-rata = 32,37 gram

% kadar = x 100% = 28 %

1. Pengulangan 3

Berat sampel = 5 gram

Berat cawan kosong = 31,90 gram

B1 = 32,14 gram

B2 = 32,17 gram

B3 = 32,17 gram

Brata-rata = 32,16 gram

% kadar = x 100% = 26 %

kadar sari larut etanol rata-rata = x 100% = 25,67 %

**Lampiran 14.** Perhitungan Penetapan Kadar Abu Total dan Kadar Abu Tidak Larut Asam Simplisia Kulit Jeruk Manis (*Citrus sinensis* L.)

1. Perhitungan penetapan kadar abu total

% kadar = x 100%

1. Pengulangan 1

Berat sampel = 2 gram

Berat cawan kosong = 59,81 gram

B1 = 59,88 gram

B2 = 59,91 gram

B1 = 59,91 gram

Brata-rata = 59,90 gram

% kadar = x 100% = 4,5 %

1. Pengulangan 2

Berat sampel = 2 gram

Berat cawan kosong = 60,53 gram

B1 = 60,63 gram

B2 = 60,65 gram

B1 = 60,65 gram

Brata-rata = 60,64 gram

% kadar = x 100% = 5,5 %

**Lampiran 14.** (Lanjutan)

1. Pengulangan 3

Berat sampel = 2 gram

Berat cawan kosong = 60,77 gram

B1 = 60,87 gram

B2 = 60,84 gram

B1 = 60,84 gram

Brata-rata = 60,85 gram

% kadar = x 100% = 4 %

Kadar abut total rata-rata = = 4,67%

1. Perhitungan penetapan kadar abu tidak larut asam

% kadar = x 100%

1. Pengulangan 1

Berat sampel = 2 gram

Berat cawan kosong = 59,81 gram

B1 = 59,83 gram

B2 = 59,82 gram

B3 = 59,82 gram

Brata-rata = 59,82 gram

% kadar = x 100% = 0,5 %

**Lampiran 14.** (Lampiran)

1. Pengulangan 2

Berat sampel = 2 gram

Berat cawan kosong = 60,53 gram

B1 = 60,53 gram

B2 = 60,56 gram

B3 = 60,56 gram

Brata-rata = 60,55 gram

% kadar = x 100% = 1 %

1. Pengulangan 3

Berat sampel = 2 gram

Berat cawan kosong = 60,77 gram

B1 = 60,77 gram

B2 = 60,79 gram

B3 = 60,79 gram

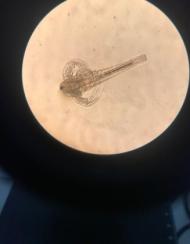
Brata-rata = 60,78 gram

% kadar = x 100% = 0,5 %

% kadar = x 100% = 0,67 %

**Lampiran 15**. Uji Sitotoksisitas Ekstrak Kulit Jeruk Manis (*Citrus sinensis* L.)

1. Penetasan telur artemia

Artemia salina

Proses penetasan

Telur artemia

Air garam buatan

1. Pengenceran ekstrak kulit jeruk manis dalam beberapa konsentrasi

1000 µg/mL

500 µg/mL

400 µg/mL

LIB

250 µg/mL

300 µg/mL

350 µg/mL

200 µg/mL

50 µg/mL

100 µg/mL

150 µg/mL

**Lampiran 15.** (Lanjutan)

1. Pengujian sitotoksisitas ekstrak etanol kulit jeruk manis (*Citrus sinensis* L.) dengan berbagai variasi konsentrasi

200 µg/mL

150 µg/mL

100 µg/mL

50 µg/mL

250 µg/mL

350 µg/mL

400 µg/mL

300 µg/mL

kontrol

1000 µg/mL

500 µg/mL

**Lampiran 16.** Perhitungan Pembuatan Variasi Pengenceran Ekstrak Kulit Jeruk Manis (*Citrus sinensis* L.)

50 mg ekstrak kulit jeruk manis dalam labu tentukur 10 ml

Lib 1 = 50 mg (C = 50.000 µg/10 mL) maka C = 5.000 µg/mL

C1 = 5000 µg/mL X 2 ml = = 1000 µg/mL

C2 = 5000 µg/mL X 1 ml = = 500 µg/mL

C4 = 5000 µg/mL X 0,8 ml = = 400 µg/mL

C5 = 5000 µg/mL X 0,7 ml = = 350 µg/mL

C6 = 5000 µg/mL X 0,6 ml = = 300 µg/mL

C7 = 5000 µg/mL X 0,5 ml = = 250 µg/mL

C8 = 5000 µg/mL X 0,4 ml = = 200 µg/mL

C9 = 5000 µg/mL X 0,3 ml = = 150 µg/mL

C10 = 5000 µg/mL X 0,2 ml = = 100 µg/mL

C11 = 5000 µg/mL X 0,1 ml = = 50 µg/mL

**Lampiran 17**. Perhitungan LC50 Ekstrak Kulit Jeruk Manis (*Citrus sinensis* L.)

% Kematian Larva = x 100%

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| d (C(µg /mL)) | N (jumlah larva) | r (jmlh larva mati) | P (% mortalitas) | X  (log C) | Y (Nilai probit) | XY | X2 |
| 100 | 30 | 7 | 23,33 | 2,0000 | 4,2710 | 8,542 | 4,0000 |
| 150 | 30 | 11 | 36,66 | 2,1761 | 4,6602 | 10,1410 | 4,7354 |
| 200 | 30 | 12 | 40 | 2,3010 | 4,7467 | 10,9221 | 5,2946 |
| 250 | 30 | 16 | 53,33 | 2,3979 | 5,0828 | 12,1880 | 5,7499 |
| 300 | 30 | 19 | 63,33 | 2,4771 | 5,3398 | 13,0670 | 6,1360 |
| 350 | 30 | 21 | 70 | 2,5441 | 5,5244 | 14,0546 | 6,4724 |
| 400 | 30 | 25 | 83,33 | 2,6020 | 5,9661 | 15,5238 | 6,7704 |
|  |  |  |  | ƩX =  16,4982 | ƩY =  35,591 | ƩXY =  84,4385 | ƩX2 =  39,1587 |

Persamaan garis regresi linier : Y = b X + a

Y = konsentrasi kematian

X = log konsentrasi

b =

b =

b =

b =

b = = 2,0216

**Lampiran 17.** (Lanjutan)

a =

a =

a =

a = 0,3197

Nilai LC50 diperoleh dari antilog X, dimana X merupakan logaritma konsentrasi bahan toksik pada Y = 5, yaitu nilai probit 50% hewan uji. Sehingaa diperoleh persamaan regresi Y = 2,0216 X + 0,3197

Jadi, 5 = 2,0216 X + 0,3197

X =

X = 2,31

Maka nilai LC50 antilog 2,31 = 204,17 µg/mL.

**Lampiran 18**. Kurva Hubungan Antara Log Konsentrasi (x) dan Nilai Probit (y) Menggunakan SPSS 20 for windows

|  |  |  |  |
| --- | --- | --- | --- |
| **Variables Entered/Removeda** | | | |
| Model | Variables Entered | Variables Removed | Method |
| 1 | Log Cb | . | Enter |

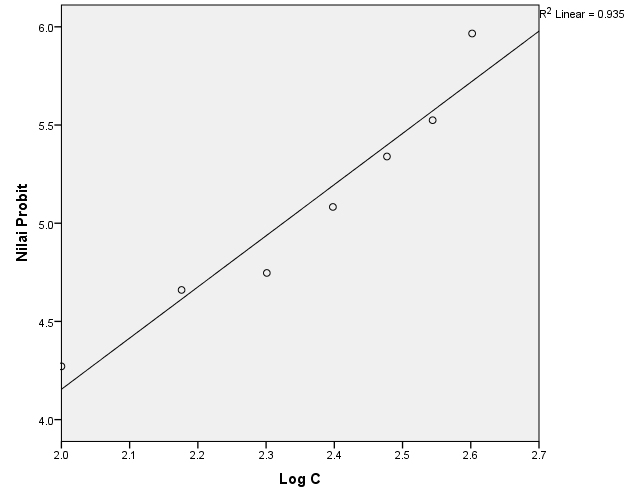
|  |
| --- |
| a. Dependent Variable: Nilai Probit |
| b. All requested variables entered. |

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Model Summary** | | | | |
| Model | R | R Square | Adjusted R Square | Std. Error of the Estimate |
| 1 | .967a | .935 | .922 | .161 |

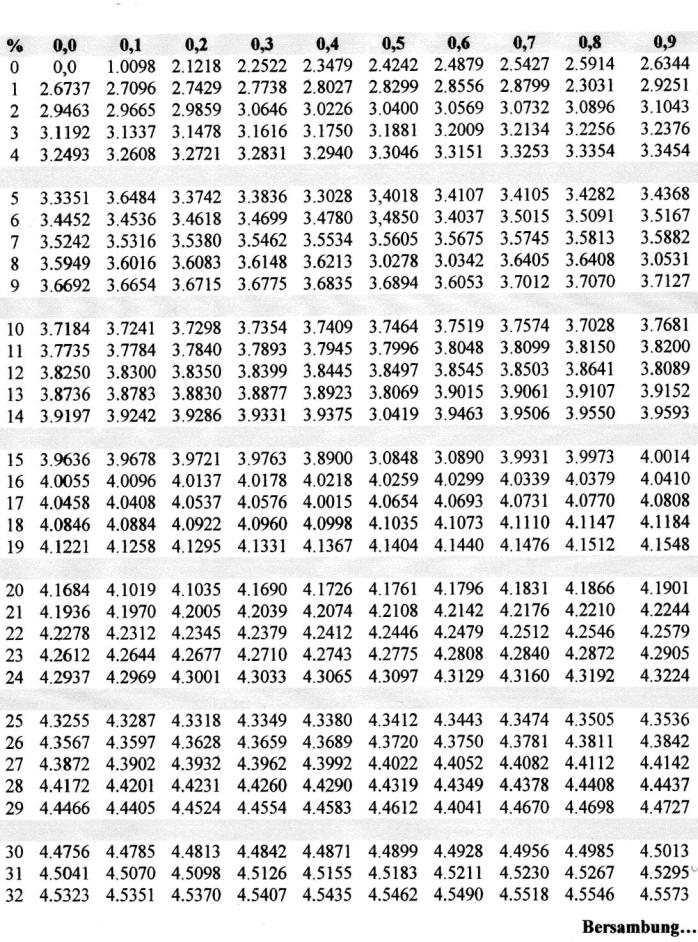
|  |
| --- |
| a. Predictors: (Constant), Log C |

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Coefficientsa** | | | | | | |
| Model | | Unstandardized Coefficients | | Standardized Coefficients | t | Sig. |
| B | Std. Error | Beta |
| 1 | (Constant) | -1.056 | .726 |  | -1.455 | .205 |
| Log C | 2.605 | .307 | .967 | 8.493 | .000 |

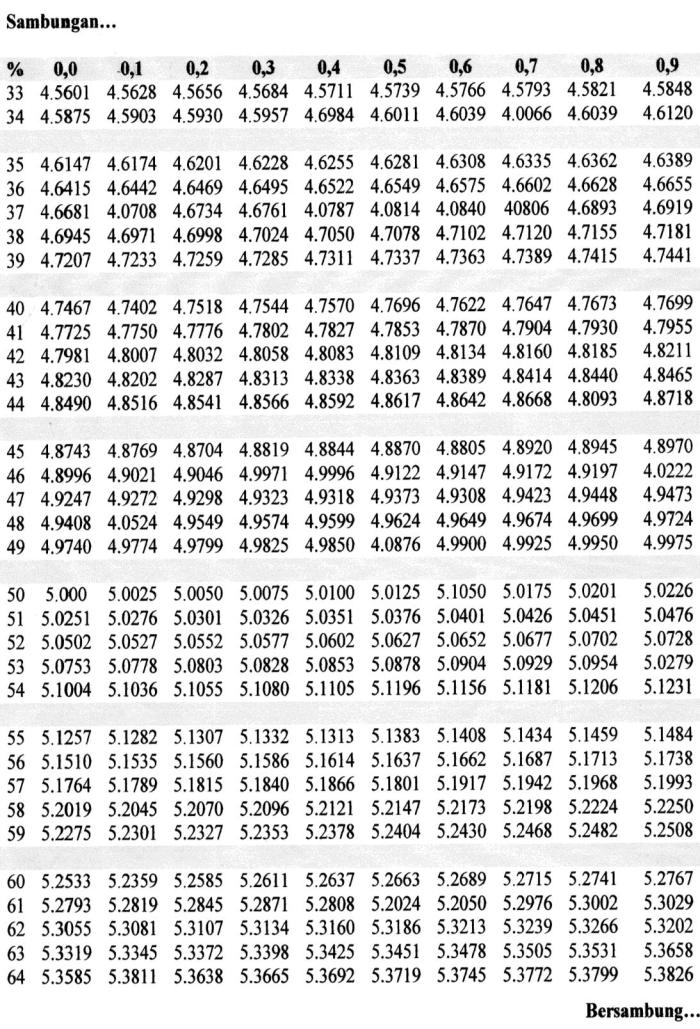
|  |
| --- |
| 1. Dependent Variable: Nilai Probit |
|  |



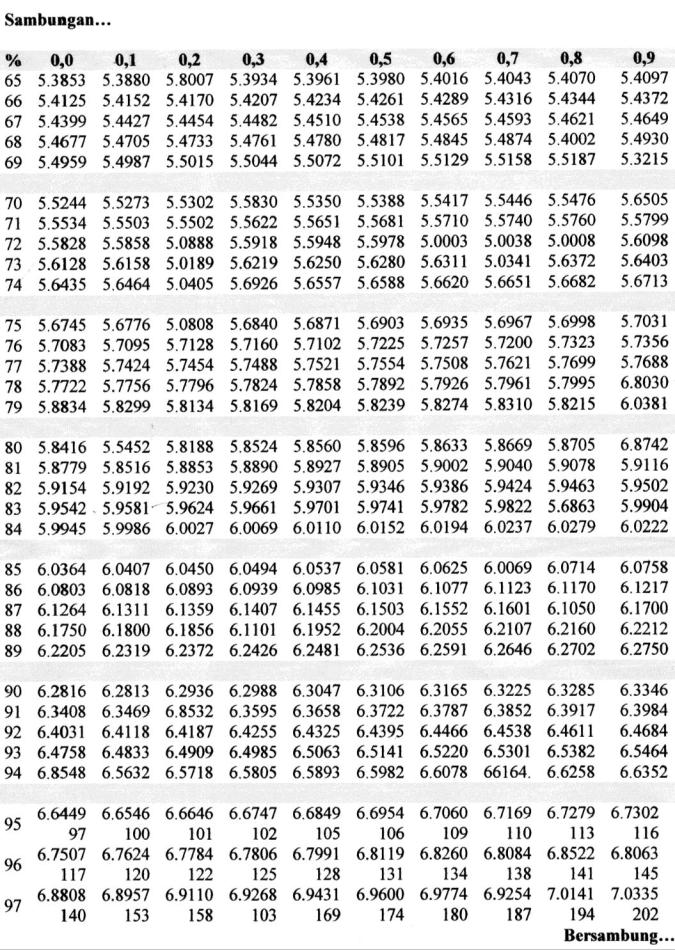
**Lampiran 19**. Tabel Probit

Nilai Probit

**Lampiran 19.** (Lanjutan)



**Lampiran 19.** (Lanjutan)



**Lampiran 19.** (Lanjutan)

