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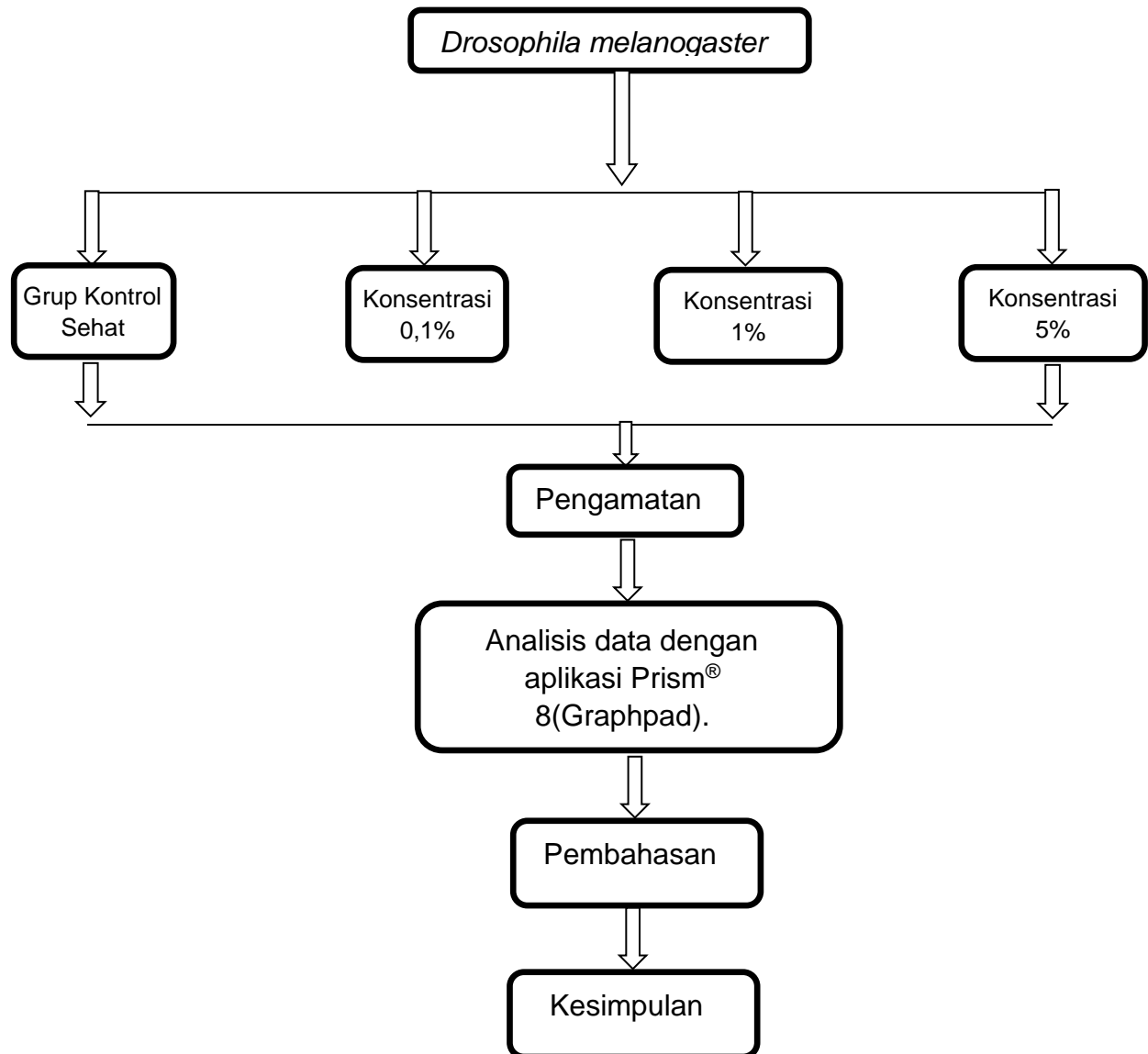


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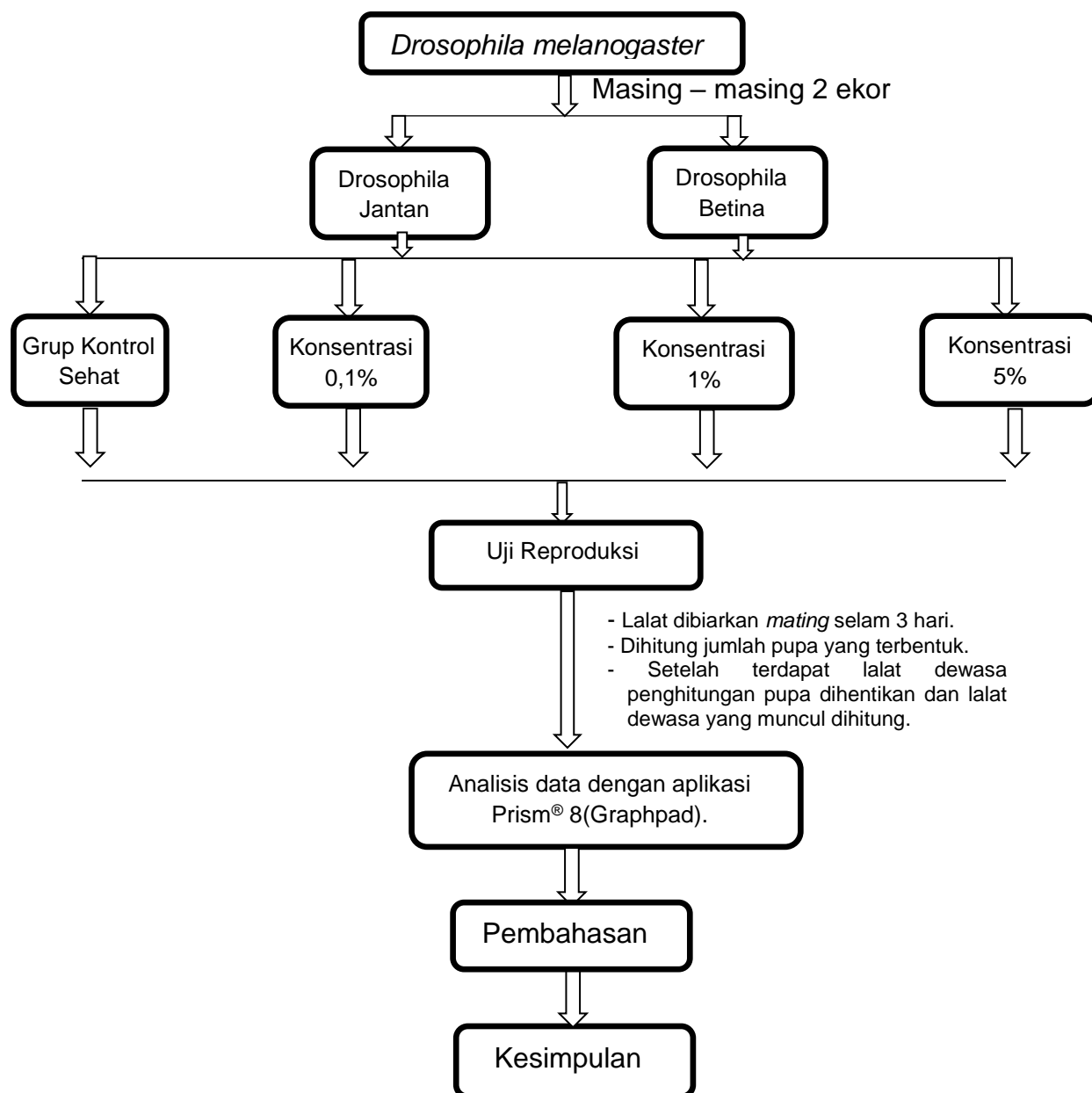
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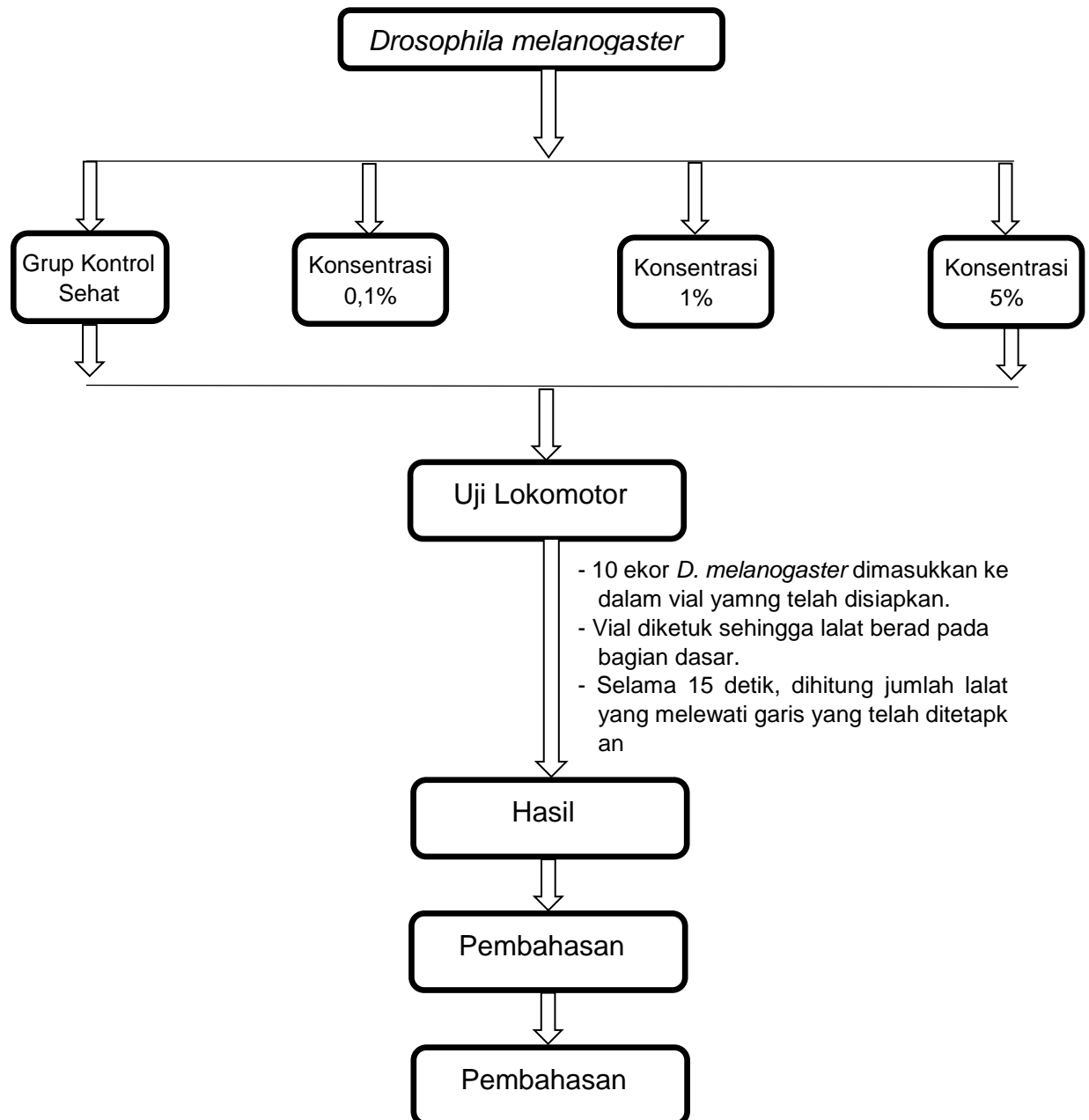
## LAMPIRAN

Lampiran 1. Skema Kerja *Survival Assay*

## Lampiran 2. Skema Kerja Reproduksi



### Lampiran 3. Skema Kerja Reproduksi



#### Lampiran 4. Skema Kerja Pembuatan Pakan *Drosophila melanogaster*

1. Disiapkan alat dan bahan yang akan digunakan.
2. Ditimbang semua bahan:
  - a. Tepung jagung sebanyak 37,5 gram.
  - b. Ragi (*yeast*) sebanyak 25 gram.
  - c. Gula sebanyak 22,5 gram.
  - d. Tepung agar sebanyak 4,5 gram
3. Dicampurkan semua bahan dalam gelas Beaker 500mL dan dihomogenkan dengan menggunakan magnetic stirer
4. Dicumukkan dengan *aquadest* yang telah disterilkan sampai 500 mL.
5. Dipanaskan pada suhu 300°C sambil diaduk selama 2 jam.
6. Ditambahkan metil paraben sebanyak 2,1 mL dan asam propionat 15% sebanyak 1,9 mL.
7. Setelah itu ditunggu hingga suhu pakan menjadi hangat dan dituang ke dalam vial yang telah disediakan.



## Lampiran 5. Penimbangan Kapsul Ekstrak *Phyllanthus niruri* Fitofarmaka

Jumlah kapsul yang ditimbang sebanyak 20 kapsul dengan obot sebagai berikut:

1. 0,2514 gram	7. 0,2516 gram	13. 0,2581 gram
2. 0,2490 gram	8. 0,2522 gram	14. 0,2656 gram
3. 0,2473 gram	9. 0,2544 gram	15. 0,2553 gram
4. 0,2565 gram	10. 0,2586 gram	16. 0,2657 gram
5. 0,2580 gram	11. 0,2621 gram	17. 0,2604 gram
6. 0,2471 gram	12. 0,2645 gram	18. 0,2580 gram
19. 0,2526 gram		
20. 0,2711 gram		

Jumlah keseluruhan bobot serbuk sebesar 5,1395 gram.





**Lampiran 6. Pembuatan Pakan Yang Mengandung Ekstrak *Phyllanthus niruri* 0,1%; 1%; dan 5%**

- a. Pembuatan Pakan yang Mengandung Ekstrak *Phyllanthus niruri*  
Konsentrasi 0,1%

Ditimbang sejumlah serbuk setara dengan 2 mg ekstrak *Phyllanthus niruri*:

$$\frac{2 \text{ mg}}{1000 \text{ mg}} \times 5,1395 \text{ gram} = 0,010279 \text{ gram}$$

0,010279 gram serbuk kemudian ditambahkan pakan sebanyak 1.989,7 mg pakan

- b. Pembuatan Pakan yang Mengandung Ekstrak *Phyllanthus niruri*  
Konsentrasi 1%

Ditimbang sejumlah serbuk setara dengan 20 mg ekstrak *Phyllanthus niruri*:

$$\frac{20 \text{ mg}}{1000 \text{ mg}} \times 5,1395 \text{ gram} = 0,10279 \text{ gram}$$

0,10279 gram serbuk kemudian ditambahkan pakan sebanyak 1.989,7 mg pakan.

- c. Pembuatan Pakan yang Mengandung Ekstrak *Phyllanthus niruri*  
Konsentrasi 5%

Ditimbang sejumlah serbuk setara dengan 100 mg ekstrak *Phyllanthus niruri*:

$$\frac{100 \text{ mg}}{1000 \text{ mg}} \times 5,1395 \text{ gram} = 0,51395 \text{ gram}$$

0,51395 gram serbuk kemudian ditambahkan pakan sebanyak 1.486 mg



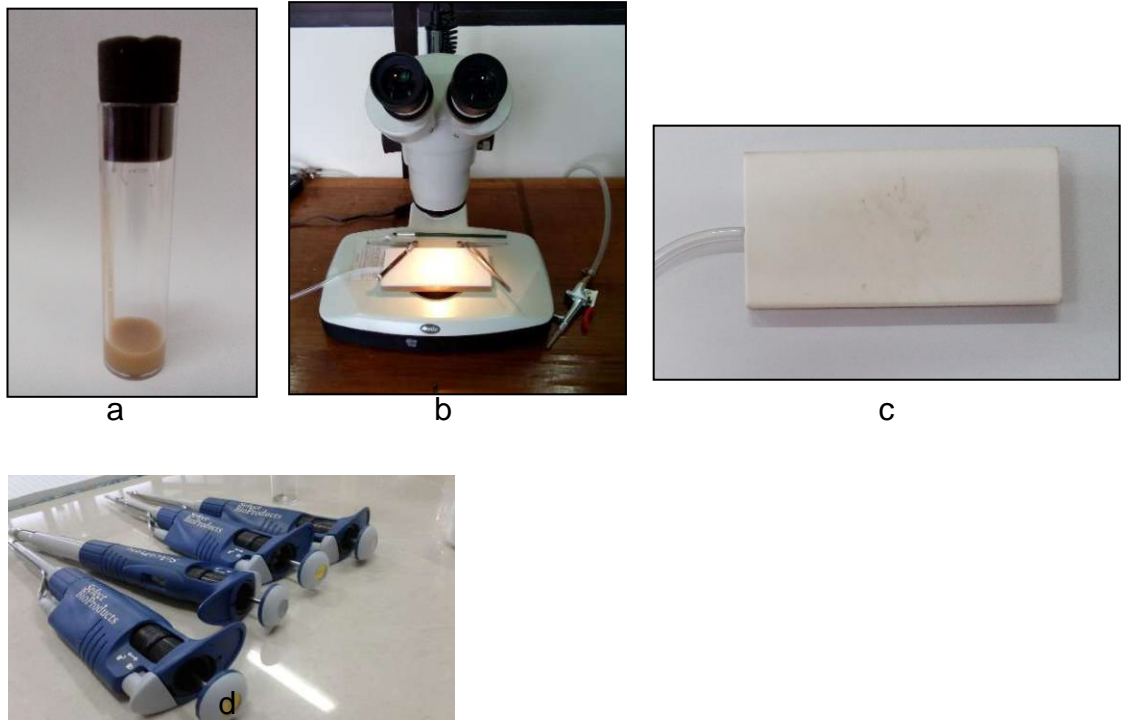
## Lampiran 7. Komposisi Pakan

### 1. Flyfood atau pakan *Drosophila melanogaster*

Komposisi pakan *D. melanogaster* untuk 1 liter :

- Corn meal : 75 g
- Glukosa : 45 g
- Yeast : 25 g
- Agar : 9 g
- Metil paraben : 4,2 ml
- Asam propionat 15%: 3,8 ml
- Air steril : ad 1 liter
- pH : 4-5.



**Lampiran 8. Gambar Alat-Alat Penelitian**

Gambar 7. Gambar alat-alat penelitian: (a) vial *drosophila*; (b) stereo mikroskop; (c) CO<sub>2</sub> stage (d) pipet mikro

**Lampiran****Statistical calc - Kontrol sehat vs Stimuno 0.1%**

## Comparison of Survival Curves

## Log-rank (Mantel-Cox) test

Chi square	4.773
df	1
P value	0.0289
P value summary	*
Are the survival curves sig different?	Yes

## Gehan-Breslow-Wilcoxon test

Chi square	4.713
df	1
P value	0.0299
P value summary	*
Are the survival curves sig different?	Yes

## Median survival

Kontrol Sehat	Undefined
EPn 0,1%	Undefined

Hazard Ratio (Mantel-Haenszel)	A/B	B/A
Ratio (and its reciprocal)	0.1079	9.268
95% CI of ratio	0.01464 to 0.7952	1.257 to 68.31
Hazard Ratio (logrank)	A/B	B/A
Ratio (and its reciprocal)	0.000	
95% CI of ratio	-1.000 to -1.000	-1.000 to -1.000



### Statistical calc - Kontrol sehat vs Stimuno 1%

#### Comparison of Survival Curves

##### Log-rank (Mantel-Cox) test

Chi square	18.08
df	1
P value	<0.0001
P value summary	****
Are the survival curves sig different?	Yes

##### Gehan-Breslow-Wilcoxon test

Chi square	17.37
df	1
P value	<0.0001
P value summary	****
Are the survival curves sig different?	Yes

#### Median survival

Kontrol Sehat	Undefined
EPn 1%	12.00

Hazard Ratio (Mantel-Haenszel)	A/C	C/A
Ratio (and its reciprocal)	0.04131	24.21
95% CI of ratio	0.009508 to 0.1795	5.572 to 105.2
Hazard Ratio (logrank)	A/C	C/A
Ratio (and its reciprocal)	0.000	
95% CI of ratio	-1.000 to -1.000	-1.000 to -1.000



### Statistical calc - Kontrol sehat vs Stimuno 5%

#### Comparison of Survival Curves

##### Log-rank (Mantel-Cox) test

Chi square	18.69
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df	1
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P value	<0.0001
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P value summary	****
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Are the survival curves sig different?	Yes
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##### Gehan-Breslow-Wilcoxon test

Chi square	17.26
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df	1
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P value	<0.0001
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P value summary	****
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Are the survival curves sig different?	Yes
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##### Median survival

Kontrol Sehat	Undefined
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EPn 5%	6.500
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Hazard Ratio (Mantel-Haenszel)	A/D	D/A
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Ratio (and its reciprocal)	0.04590	21.79
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95% CI of ratio	0.01135 to 0.1856	5.389 to 88.09
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Hazard Ratio (logrank)	A/D	D/A
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Ratio (and its reciprocal)	0.000
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95% CI of ratio	-1.000 to -1.000	-1.000 to -1.000
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### Statistical calc - Stimuno 0.1% vs Stimuno 1%

#### Comparison of Survival Curves

##### Log-rank (Mantel-Cox) test

Chi square	4.544
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df	1
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P value	0.0330
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P value summary	*
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Are the survival curves sig different?	Yes
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##### Gehan-Breslow-Wilcoxon test

Chi square	4.079
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df	1
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P value	0.0434
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P value summary	*
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Are the survival curves sig different?	Yes
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##### Median survival

EPn 0,1%	Undefined
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EPn 1%	12.00
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##### Hazard Ratio (Mantel-Haenszel)

B/C

C/B

Ratio (and its reciprocal)

0.2487

4.021

95% CI of ratio

0.06919 to 0.8940

1.119 to 14.45

##### Hazard Ratio (logrank)

B/C

C/B

Ratio (and its reciprocal)

0.3708

2.697

95% CI of ratio

0.1300 to 1.058

0.9453 to 7.695



### Statistical calc - Stimuno 1% vs Stimuno 5%

#### Comparison of Survival Curves

##### Log-rank (Mantel-Cox) test

Chi square	6.975
df	1
P value	0.0083
P value summary	**
Are the survival curves sig different?	Yes

##### Gehan-Breslow-Wilcoxon test

Chi square	5.487
df	1
P value	0.0192
P value summary	*
Are the survival curves sig different?	Yes

#### Median survival

EPn 1%	12.00
EPn 5%	6.500
Ratio (and its reciprocal)	1.846      0.5417
95% CI of ratio	0.7684 to 4.436    0.2255 to 1.301

#### Hazard Ratio (Mantel-Haenszel)

	C/D	D/C
Ratio (and its reciprocal)	0.1937	5.164
95% CI of ratio	0.05727 to 0.6549	1.527 to 17.46

#### Hazard Ratio (logrank)

	C/D	D/C
Ratio (and its reciprocal)	0.4036	2.477
95% CI of ratio	0.1533 to 1.063	0.9408 to 6.524





### Statistical calc - Stimuno 0.1% vs Stimuno 5%

#### Comparison of Survival Curves

##### Log-rank (Mantel-Cox) test

Chi square	18.69
df	1
P value	<0.0001
P value summary	****
Are the survival curves sig different?	Yes

##### Gehan-Breslow-Wilcoxon test

Chi square	17.26
df	1
P value	<0.0001
P value summary	****
Are the survival curves sig different?	Yes

##### Median survival

EPn 0,1%	Undefined
EPn 5%	6.500

Hazard Ratio (Mantel-Haenszel)	B/D	D/B
Ratio (and its reciprocal)	0.04590	21.79
95% CI of ratio	0.01135 to 0.1856	5.389 to 88.09
Hazard Ratio (logrank)	B/D	D/B
Ratio (and its reciprocal)	0.1564	6.395
95% CI of ratio	0.04876 to 0.5015	1.994 to 20.51



## Reproduksi

Number of families	1
Number of comparisons per family	6
Alpha	0.05

Tukey's multiple comparisons test	Mean Diff.	95.00% CI of diff.	Significant?	Summary	Adjusted P Value	
PN + PN vs. Stimuno 0,1% + PN	42.00	-324.7 to 240.7	No	ns	0.3415	A-B
PN + PN vs. PN + Stimuno 0,1%	28.00	-115.0 to 58.98	No	ns	0.1641	A-C
PN + PN vs. Stimuno 0,1% + Stimuno 0,1%	18.00	-257.2 to 221.2	No	ns	0.5938	A-D
Stimuno 0,1% + PN vs. PN + Stimuno 0,1%	14.00	-355.6 to 383.6	No	ns	0.8479	B-C
Stimuno 0,1% + PN vs. Stimuno 0,1% + Stimuno 0,1%	24.00	-19.49 to 67.49	No	ns	0.0964	B-D
PN + Stimuno 0,1% vs. Stimuno 0,1% + Stimuno 0,1%	10.00	-316.2 to 336.2	No	ns	0.9004	C-D

Test details	Mean 1	Mean 2	Mean Diff.	SE of diff.	n1	n2	q	D F
PN + PN vs. Stimuno 0,1% + PN	55.00	97.00	-42.00	13.00	2	2	4.569	1
PN + PN vs. PN + Stimuno 0,1%	55.00	83.00	-28.00	4.000	2	2	9.899	1
PN + PN vs. Stimuno 0,1% + Stimuno 0,1%	55.00	73.00	-18.00	11.00	2	2	2.314	1
Stimuno 0,1% + PN vs. PN + Stimuno 0,1%	97.00	83.00	14.00	17.00	2	2	1.165	1
Stimuno 0,1% + PN vs. Stimuno 0,1% + Stimuno 0,1%	97.00	73.00	24.00	2.000	2	2	16.97	1
PN + Stimuno 0,1% vs. Stimuno 0,1% + Stimuno 0,1%	83.00	73.00	10.00	15.00	2	2	0.9428	1



**Lokomotor**

Number of families			1					
Number of comparisons per family			6					
Alpha			0.05					
Tukey's multiple comparisons test	Mean Diff.	95.00% CI of diff.	Significant?	Summary	Adjusted P Value			
PN vs. EPn 0,1%	-20.00	-96.16 to 56.16	No	ns	0.7242	A-B		
PN vs. EPn 1%	10.00	-66.16 to 86.16	No	ns	0.9461	A-C		
PN vs. EPn 5%	0.000	-76.16 to 76.16	No	ns	>0.9999	A-D		
EPn 0,1% vs. EPn 1%	30.00	-46.16 to 106.2	No	ns	0.4676	B-C		
EPn 0,1% vs. EPn 5%	20.00	-56.16 to 96.16	No	ns	0.7242	B-D		
EPn 1% vs. EPn 5%	-10.00	-86.16 to 66.16	No	ns	0.9461	C-D		
Test details	Mean 1	Mean 2	Mean Diff.	SE of diff.	n1	n2	q	DF
PN vs. EPn 0,1%	50.00	70.00	-20.00	18.71	2	2	1.512	4
PN vs. EPn 1%	50.00	40.00	10.00	18.71	2	2	0.7559	4
PN vs. EPn 5%	50.00	50.00	0.000	18.71	2	2	0.000	4
EPn 0,1% vs. EPn 1%	70.00	40.00	30.00	18.71	2	2	2.268	4
EPn 0,1% vs. EPn 5%	70.00	50.00	20.00	18.71	2	2	1.512	4
EPn 1% vs. EPn 5%	40.00	50.00	-10.00	18.71	2	2	0.7559	4

