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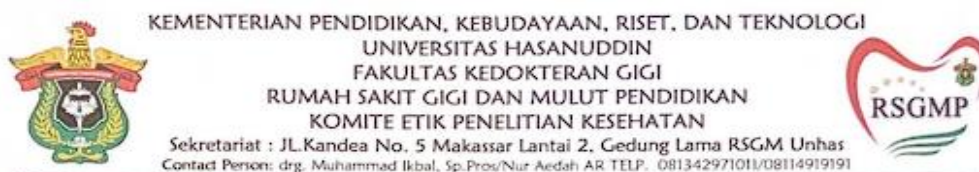
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## LAMPIRAN GAMBAR PENELITIAN

### 1. Lembar Penelitian Etik



#### REKOMENDASI PERSETUJUAN ETIK Nomor: 0125/PL.09/KEPK FKG-RSGM UNHAS/2023

Tanggal: 06 Juli 2023

Dengan ini menyatakan bahwa protokol dan dokumen yang berhubungan dengan protokol berikut ini telah mendapatkan persetujuan etik:

No. Protokol	UH 17120864	No Protokol Sponsor	
Peneliti Utama	drg. Arrang Sestoria	Sponsor	Pribadi
Judul Peneliti	Pengaruh Ekstrak Etanol Daun Kelor ( <i>Moringa oleifera</i> ) terhadap Ekspresi TGF- $\beta$ pada Periodontitis Anak yang di Induksi Bakteri <i>Porphyromonas gingivalis</i> (Studi In Vivo Pada Rattus Novergicus)		
No. Versi Protokol	1	Tanggal Versi	03 Juli 2023
No. Versi Protokol		Tanggal Versi	
Tempat Penelitian	1. Laboratorium Biokimia Politeknik Pangkep. 2. Laboratorium Mikrobiologi Fakultas Kedokteran Unhas. 3. Klinik Hewan Lacoste. 4. Laboratorium Patologi Anatomi RSP Unhas		
Dokumen Lain			
Jenis Review	<input type="checkbox"/> Exempted <input checked="" type="checkbox"/> Expedited <input type="checkbox"/> Fullboard	Masa Berlaku 06 Juli 2023-06 Juli 2024	Frekuensi Review Lanjutan
Ketua Komisi Etik Penelitian	Nama: Dr. drg. Marhamah, M.Kes	Tanda Tangan 	Tanggal
Sekretaris Komisi Etik Penelitian	Nama: drg. Muhammad Iqbal, Sp.Prost	Tanda Tangan 	Tanggal

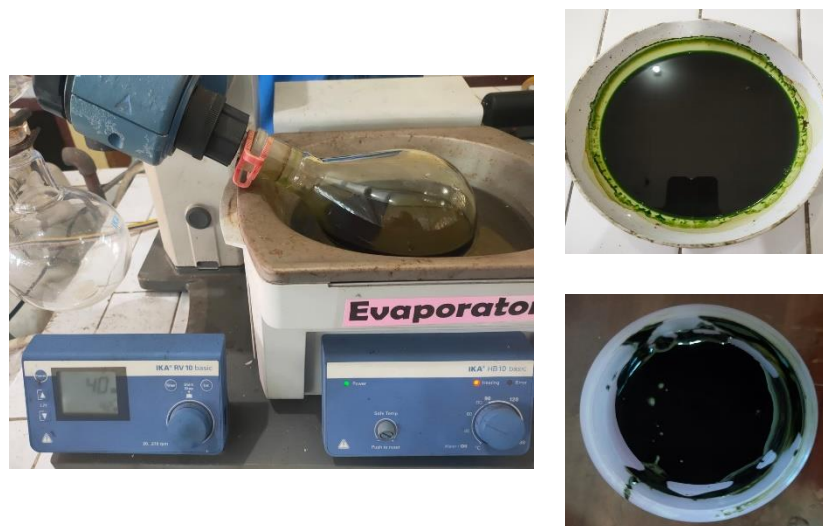
Kewajiban peneliti utama:

- Menyerahkan Amandemen Protokol untuk persetujuan sebelum diimplementasikan
- Menyerahkan laporan SAE ke Komisi Etik dalam 24 Jam dan dilengkapi dalam 7 hari dan lapor SUSAR dalam 72 jam setelah peneliti utama menerima laporan.
- Menyerahkan laporan kemajuan (*progress report*) setiap 6 bulan untuk penelitian resiko tinggi dan setiap setahun untuk penelitian resiko rendah.
- Menyerahkan laporan akhir setelah penelitian berakhir.
- Melaporkan penyimpangan dari protokol yang disetujui (*protocol deviation/violation*)
- Mematuhi semua aturan yang berlaku.

## 2. Persiapan Pembuatan Ekstrak Kelor (*Moringa oleifera*)



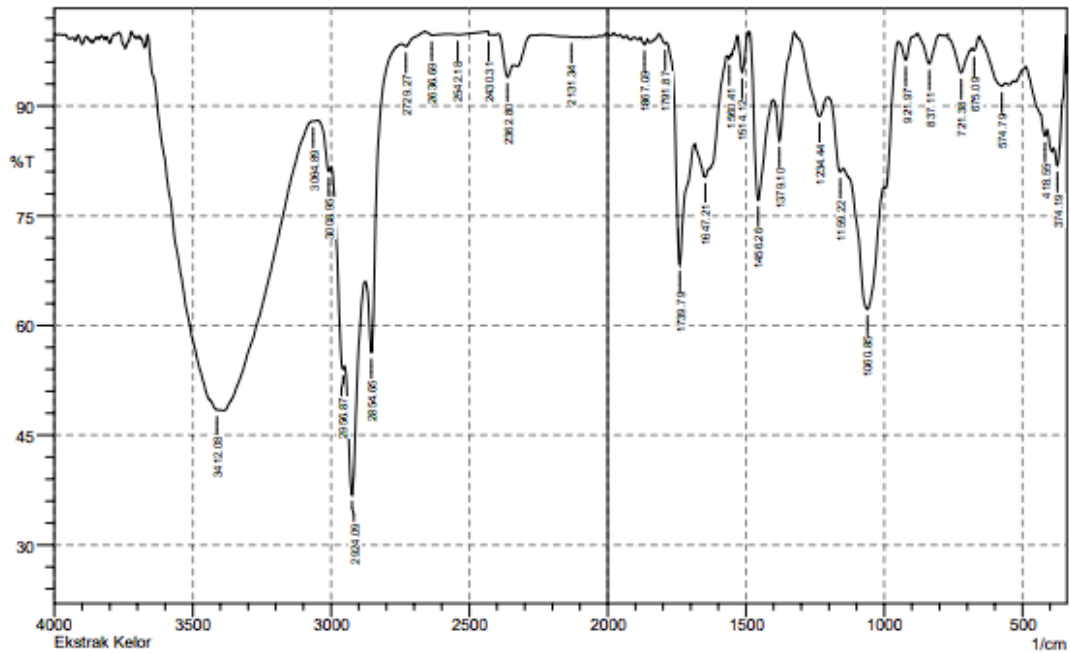
Daun kelor kering dihaluskan sampai berupa bubuk lalu disaring sehingga memperoleh hasil bubuk halus. Tepung Daun Kelor ditimbang dan di maserasi selama 24 jam dengan pelarut Ethanol. Hasil maserasi disaring dan proses ini di ulang hingga 3 kali.



Pemisahan pelarut etanol dengan ekstrak menggunakan alat Rotary Evaporator. Sisa – sisa pelarut diuapkan dalam oven suhu 50°C sampai diperoleh ekstrak berbentuk pasta dengan konsentrasi 100%.



### 3. Hasil Interpretasi Kandungan Ekstrak kelor dengan alat (FTIR)

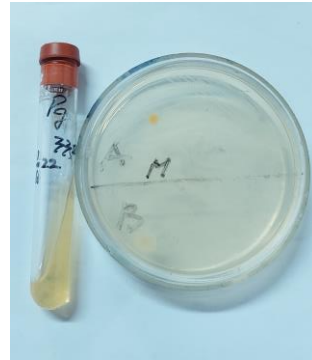


No.	Peak	Intensity	Corr. Intensity	Base (H)	Base (L)	Area	Corr. Area
1	374.19	81.85	6.147	385.76	345.26	2.485	0.81
2	418.55	85.871	1.707	487.99	410.84	3.465	0.253
3	574.79	92.716	1.178	655.8	563.21	1.921	0.356
4	675.09	97.566	0.802	682.8	655.8	0.2	0.048
5	721.38	94.546	3.941	763.81	682.8	1.183	0.651
6	837.11	95.776	4.049	879.54	800.46	0.706	0.647
7	921.97	96.28	2.944	948.98	879.54	0.5	0.303
8	1060.85	62.219	17.752	1147.65	1001.06	21.154	7.091
9	1159.22	80.972	2.236	1203.58	1149.57	3.643	0.242
10	1234.44	88.583	4.758	1325.1	1205.51	3.531	1.155
11	1379.1	85.235	8.03	1398.39	1327.03	2.438	0.982
12	1456.26	77.117	19.47	1485.19	1406.11	5.455	3.738
13	1514.12	94.594	5.229	1531.48	1492.9	0.476	0.451
14	1560.41	96.457	0.446	1564.27	1554.63	0.14	0.01
15	1647.21	80.292	0.26	1649.14	1633.71	1.422	0.012
16	1739.79	68.304	23.96	1786.08	1685.79	8.422	4.485
17	1791.87	98.506	0.405	1815.02	1786.08	0.12	0.028
18	1867.09	98.389	0.838	1882.52	1855.52	0.142	0.053
19	2131.34	99.436	0.009	2150.63	2127.48	0.055	0.001
20	2362.8	93.923	3.401	2393.66	2341.58	0.902	0.374
21	2430.31	99.736	0.37	2432.24	2422.59	0.009	0.008
22	2542.18	99.73	0.184	2573.04	2432.24	0.048	0.057
23	2636.69	99.65	0.434	2661.77	2578.83	0.063	0.068
24	2729.27	98.142	0.639	2744.71	2661.77	0.307	0.062
25	2854.65	56.307	15.027	2875.86	2746.63	10.076	1.82
26	2924.09	36.869	21.805	2951.09	2877.79	21.904	5.691
27	2956.87	53.995	2.598	2999.31	2953.02	8.341	0.379
28	3008.95	81.071	1.614	3047.53	3001.24	3.342	0.104
29	3064.89	87.894	0.023	3066.82	3049.46	0.967	0.002
30	3412.08	48.445	1.175	3662.82	3406.29	45.714	5.2

Comment;  
Ekstrak Kelor

Date/Time; 10/11/2023 12:47:24 PM  
No. of Scans;  
Resolution;  
Apodization;

#### 4. Pembuatan Suspensi Bakteri *Porphyromonas gingivalis*



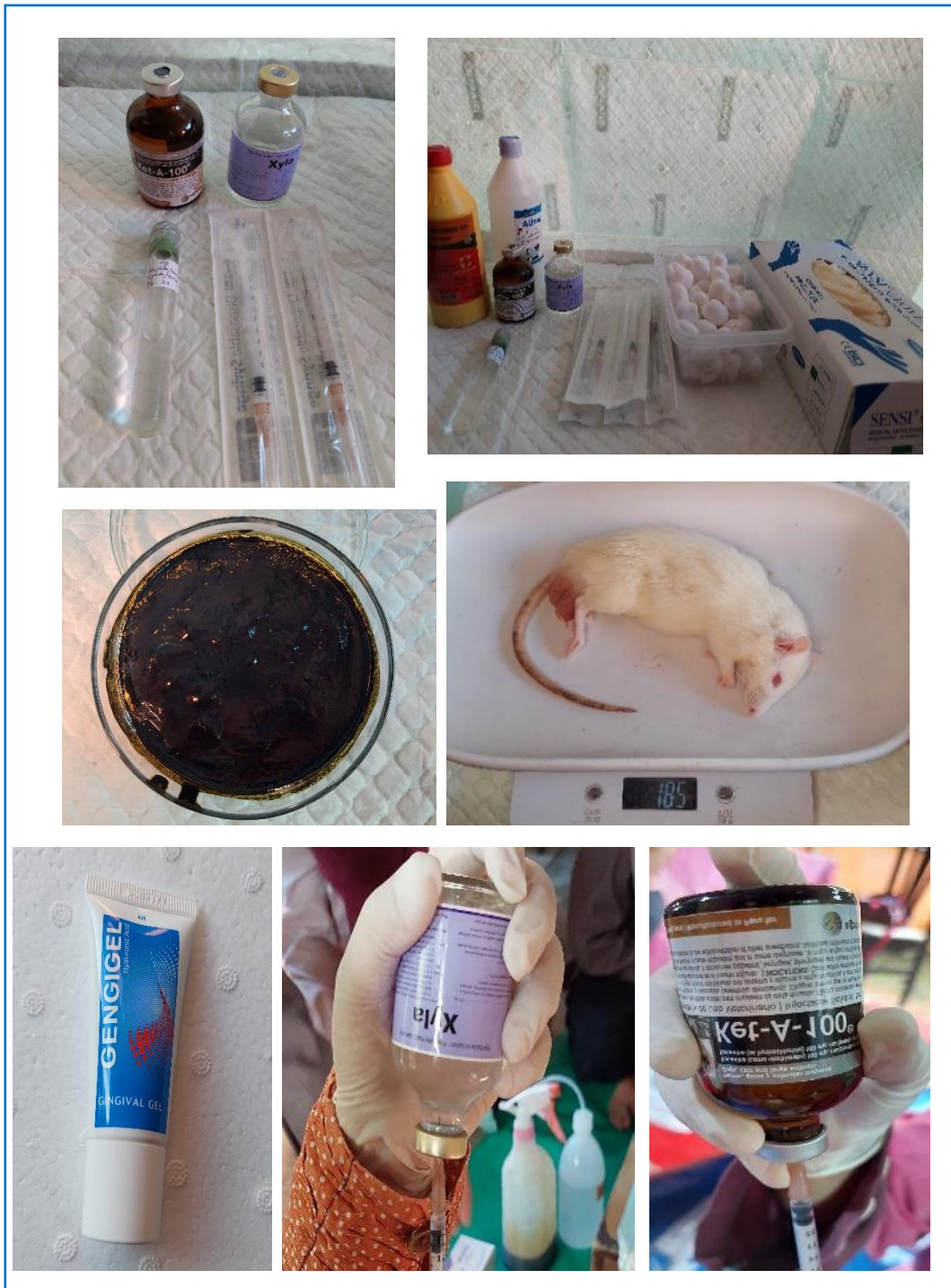
Pembuatan media cair sebanyak 10 ml, yaitu dari 0,37 gram BHI-B, 1  $\mu$ l vitamin K, 5  $\mu$ l hemin serta 50  $\mu$ l ekstrak yeast dan dibagi menjadi 2 bagian @5cc. Tiap bagian diberikan satu ose bakteri yang berasal dari pembiakan di media agar BHI-A. Suspensi bakteri yang didapat lalu dimasukkan desicator dan dinkubasi selama 2x24 jam. Setelah di inkubasi, suspensi bakteri diukur konsentrasinya hingga didapatkan  $1,5 \times 10^6$ .

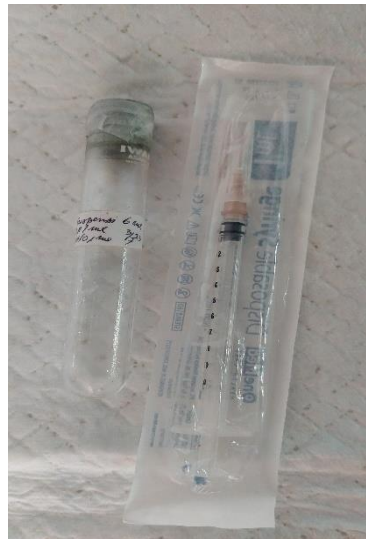
#### 5. Adaptasi Hewan



Tikus wistar dipelihara secara berkelompok (5- 6 ekor per kandang). Adaptasi dilakukan selama 7 hari untuk mengkondisikan hewan dalam keadaan sehat. Makanan berupa kombinasi pellet dan sayuran serta minum yang cukup.

## 6. Perlakuan pada Hewan Coba



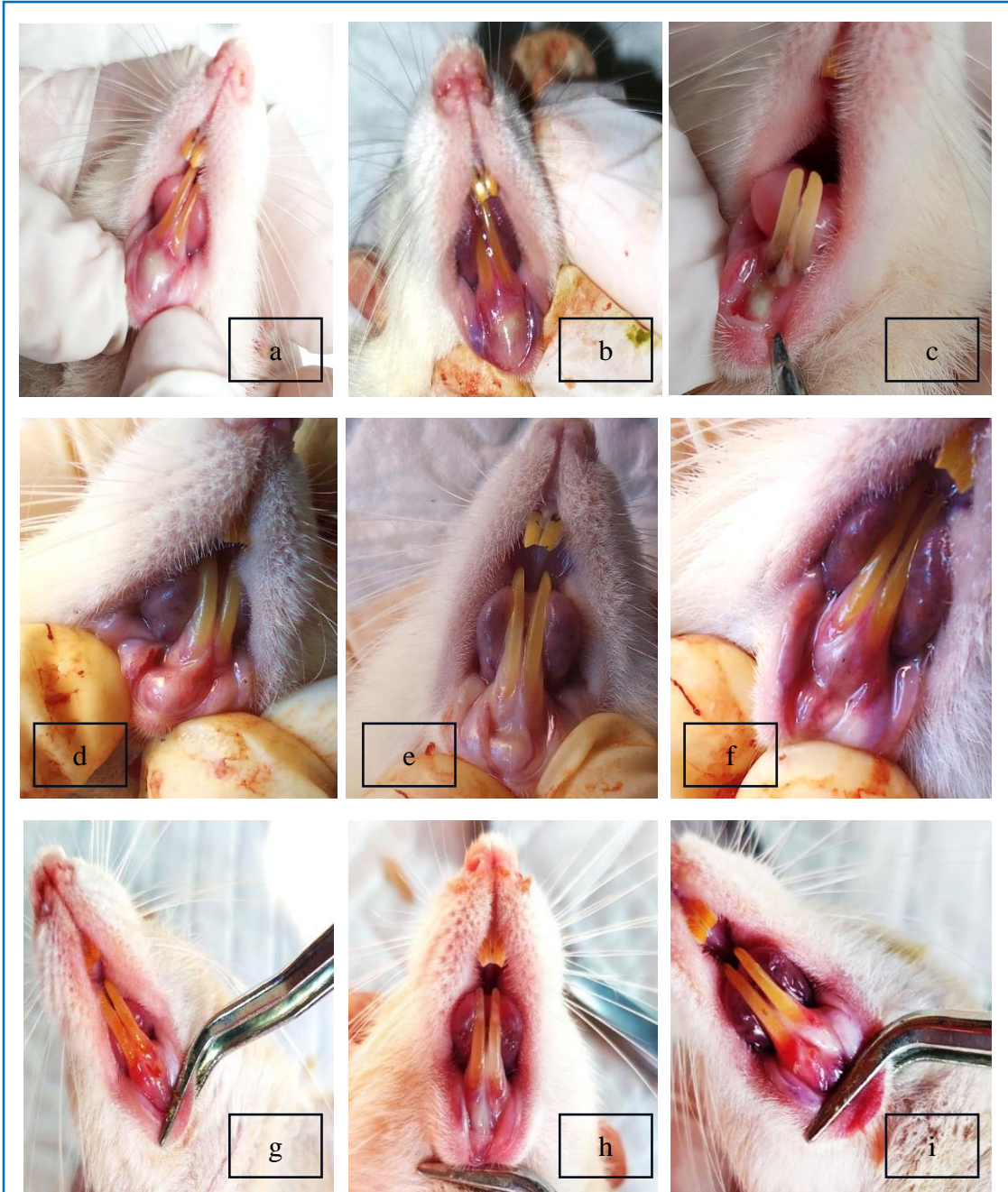


Pertama-tama dilakukan persiapan alat. Menimbang berat badan masing-masing wistar untuk mengukur dosis anestesi. Menyuntikkan anestesi sesuai dosis pada paha tikus yang sudah di sterilkan dengan alkohol. Suspensi bakteri diambil memalui spuit 1 cc steril. Menginduksi bakteri *Porphyromonas gingivalis* pada jaringan periodontal gigi insisivus mandibula



Penampakan klinis jaringan periodontal 7 hari post induksi bakteri *Porphyromonas gingivalis* pada kelompok a) perlakuan b) kontrol (+) c) kontrol (-). Pemberian ekstrak kelor pada kelompok d) perlakuan , e) pemberian gengigel pada kelompok kontrol (+) dan f) irigasi aquades pada kelompok kontrol (-).

## 7. Pengambilan Sampel Jaringan



Klinis Hari ke-3 post perlakuan pada kelompok a) Perlakuan (MO) b) kontrol (+) c) Kontrol (-). Klinis hari ke 7 kelompok d) perlakuan (MO) e) kontrol (+) f) kontrol (-). Klinis hari ke 14 kelompok g) perlakuan (MO) h) kontrol (+) i) kontrol (-).



Euthanasia tikus wistar spesimen dilakukan dengan menggunakan alat bedah minor steril. Jaringan tulang ditempatkan dalam pot steril yang berisi formalin 10 % dan diberi label.

## 8. Pembuatan Slide Jaringan



Jaringan yang telah dipotong dimasukkan ke dalam kaset dan diproses di dalam mesin prosesing jaringan (Tissue Automatics Processor). Proses Embedding (jaringan yang telah diproses dalam mesin prosesing diblok menggunakan parafin cair). Potong jaringan dalam blok paraffin menggunakan mikrotom dengan ketebalan 3-4 $\mu$ m. Pita jaringan yang terbentuk dicelupkan ke dalam Waterbath. Ambil potongan jaringan dengan slide lalu tiriskan. Tuliskan kode pada slide sesuai dengan kode yang tertera pada blok paraffin menggunakan pensil.



## 9. Analisis Data

### Kelompok Perlakuan



Kelompok Perlakuan		Descriptives		Statistic	Std. Error
TGFB1	P (Moringa oleifera)	Mean		10.3333	.70711
		95% Confidence Interval for Mean	Lower Bound	8.7027	
			Upper Bound	11.9639	
		5% Trimmed Mean		10.3704	
		Median		10.0000	
		Variance		4.500	
		Std. Deviation		2.12132	
		Minimum		7.00	
		Maximum		13.00	
		Range		6.00	
		Interquartile Range		4.00	
		Skewness		-.157	.717
		Kurtosis		-1.062	1.400
		K+ (Gengigel)		Mean	
95% Confidence Interval for Mean	Lower Bound			6.9914	
	Upper Bound			10.3419	
5% Trimmed Mean				8.6852	
Median				9.0000	
Variance				4.750	
Std. Deviation				2.17945	
Minimum				5.00	
Maximum				12.00	
Range				7.00	
Interquartile Range				3.50	
Skewness				-.103	.717
Kurtosis				-.385	1.400
k- (Aquades)				Mean	
		95% Confidence Interval for Mean	Lower Bound	3.4164	
			Upper Bound	6.8059	
		5% Trimmed Mean		5.0679	
		Median		5.0000	
		Variance		4.861	
		Std. Deviation		2.20479	
		Minimum		2.00	
		Maximum		9.00	
		Range		7.00	
		Interquartile Range		3.50	
		Skewness		.270	.717
		Kurtosis		-.308	1.400

### Tests of Normality

Kelompok Perlakuan	Kolmogorov-Smirnov <sup>a</sup>			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
TGFb1 P (Moringa oleifera)	.118	9	.200*	.948	9	.672
K+ (Gengigel)	.116	9	.200*	.982	9	.972
k- (Aquadex)	.164	9	.200*	.959	9	.789

\*. This is a lower bound of the true significance.

a. Lilliefors Significance Correction

### Oneway (P)

#### Descriptives

TGFb1

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
Hari ke-3	3	8.3333	1.52753	.88192	4.5388	12.1279	7.00	10.00
Hari ke-7	3	10.3333	1.52753	.88192	6.5388	14.1279	9.00	12.00
Hari ke-14	3	12.3333	1.15470	.66667	9.4649	15.2018	11.00	13.00
Total	9	10.3333	2.12132	.70711	8.7027	11.9639	7.00	13.00

#### ANOVA

TGFb1

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	24.000	2	12.000	6.000	.037
Within Groups	12.000	6	2.000		
Total	36.000	8			

### Post Hoc Tests

#### Multiple Comparisons

Dependent Variable: TGFb1

LSD

(I) Waktu Pengamatan	(J) Waktu Pengamatan	Mean Difference			95% Confidence Interval	
		(I-J)	Std. Error	Sig.	Lower Bound	Upper Bound
Hari ke-3	Hari ke-7	-2.00000	1.15470	.134	-4.8255	.8255
	Hari ke-14	-4.00000	1.15470	.013	-6.8255	-1.1745
Hari ke-7	Hari ke-3	2.00000	1.15470	.134	-.8255	4.8255
	Hari ke-14	-2.00000	1.15470	.134	-4.8255	.8255
Hari ke-14	Hari ke-3	4.00000	1.15470	.013	1.1745	6.8255
	Hari ke-7	2.00000	1.15470	.134	-.8255	4.8255

## Oneway (K+)

### Descriptives

TGFb1

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
Hari ke-3	3	7.0000	2.00000	1.15470	2.0317	11.9683	5.00	9.00
Hari ke-7	3	8.6667	2.08167	1.20185	3.4955	13.8378	7.00	11.00
Hari ke-14	3	10.3333	1.52753	.88192	6.5388	14.1279	9.00	12.00
Total	9	8.6667	2.17945	.72648	6.9914	10.3419	5.00	12.00

### ANOVA

TGFb1

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	16.667	2	8.333	2.344	.177
Within Groups	21.333	6	3.556		
Total	38.000	8			

## Post Hoc Tests

### Multiple Comparisons

Dependent Variable: TGFb1

LSD

(I) Waktu Pengamatan	(J) Waktu Pengamatan	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
Hari ke-3	Hari ke-7	-1.66667	1.53960	.321	-5.4339	2.1006
	Hari ke-14	-3.33333	1.53960	.074	-7.1006	.4339
Hari ke-7	Hari ke-3	1.66667	1.53960	.321	-2.1006	5.4339
	Hari ke-14	-1.66667	1.53960	.321	-5.4339	2.1006
Hari ke-14	Hari ke-3	3.33333	1.53960	.074	-.4339	7.1006
	Hari ke-7	1.66667	1.53960	.321	-2.1006	5.4339

## Oneway (K-)

### Descriptives

TGFb1

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
Hari ke-3	3	7.0000	2.00000	1.15470	2.0317	11.9683	5.00	9.00
Hari ke-7	3	8.6667	2.08167	1.20185	3.4955	13.8378	7.00	11.00
Hari ke-14	3	10.3333	1.52753	.88192	6.5388	14.1279	9.00	12.00
Total	9	8.6667	2.17945	.72648	6.9914	10.3419	5.00	12.00

### ANOVA

TGFb1

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	16.667	2	8.333	2.344	.177
Within Groups	21.333	6	3.556		
Total	38.000	8			

## Post Hoc Tests

### Multiple Comparisons

Dependent Variable: TGFb1

LSD

(I) Waktu Pengamatan	(J) Waktu Pengamatan	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
Hari ke-3	Hari ke-7	-1.66667	1.53960	.321	-5.4339	2.1006
	Hari ke-14	-3.33333	1.53960	.074	-7.1006	.4339
Hari ke-7	Hari ke-3	1.66667	1.53960	.321	-2.1006	5.4339
	Hari ke-14	-1.66667	1.53960	.321	-5.4339	2.1006
Hari ke-14	Hari ke-3	3.33333	1.53960	.074	-.4339	7.1006
	Hari ke-7	1.66667	1.53960	.321	-2.1006	5.4339

### Oneway (hari ke-3)

#### Descriptives

TGFb1

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
P (Moringa oleifera)	3	8.3333	1.52753	.88192	4.5388	12.1279	7.00	10.00
K+ (Gengigel)	3	7.0000	2.00000	1.15470	2.0317	11.9683	5.00	9.00
k- (Aquades)	3	3.3333	1.52753	.88192	-.4612	7.1279	2.00	5.00
Total	9	6.2222	2.68225	.89408	4.1605	8.2840	2.00	10.00

#### ANOVA

TGFb1

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	40.222	2	20.111	6.962	.027
Within Groups	17.333	6	2.889		
Total	57.556	8			

### Post Hoc Tests

#### Multiple Comparisons

Dependent Variable: TGFb1

LSD

(I) Kelompok Perlakuan	(J) Kelompok Perlakuan	Mean Difference			95% Confidence Interval	
		(I-J)	Std. Error	Sig.	Lower Bound	Upper Bound
P (Moringa oleifera)	K+ (Gengigel)	1.33333	1.38778	.374	-2.0624	4.7291
	k- (Aquades)	5.00000	1.38778	.011	1.6042	8.3958
K+ (Gengigel)	P (Moringa oleifera)	-1.33333	1.38778	.374	-4.7291	2.0624
	k- (Aquades)	3.66667	1.38778	.038	.2709	7.0624
k- (Aquades)	P (Moringa oleifera)	-5.00000	1.38778	.011	-8.3958	-1.6042
	K+ (Gengigel)	-3.66667	1.38778	.038	-7.0624	-.2709

### Oneway (hari ke-7)

#### Descriptives

TGFb1

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
P (Moringa oleifera)	3	10.3333	1.52753	.88192	6.5388	14.1279	9.00	12.00
K+ (Gengigel)	3	8.6667	2.08167	1.20185	3.4955	13.8378	7.00	11.00
k- (Aquades)	3	4.6667	1.52753	.88192	.8721	8.4612	3.00	6.00
Total	9	7.8889	2.93447	.97816	5.6333	10.1445	3.00	12.00

#### ANOVA

TGFb1

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	50.889	2	25.444	8.481	.018
Within Groups	18.000	6	3.000		
Total	68.889	8			

### Post Hoc Tests

#### Multiple Comparisons

Dependent Variable: TGFb1

LSD

(I) Kelompok Perlakuan	(J) Kelompok Perlakuan	Mean Difference			95% Confidence Interval	
		(I-J)	Std. Error	Sig.	Lower Bound	Upper Bound
P (Moringa oleifera)	K+ (Gengigel)	1.66667	1.41421	.283	-1.7938	5.1271
	k- (Aquades)	5.66667	1.41421	.007	2.2062	9.1271
K+ (Gengigel)	P (Moringa oleifera)	-1.66667	1.41421	.283	-5.1271	1.7938
	k- (Aquades)	4.00000	1.41421	.030	.5395	7.4605
k- (Aquades)	P (Moringa oleifera)	-5.66667	1.41421	.007	-9.1271	-2.2062
	K+ (Gengigel)	-4.00000	1.41421	.030	-7.4605	-.5395

## Oneway (Hari 14)

### Descriptives

TGFb1

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
P (Moringa oleifera)	3	12.3333	1.15470	.66667	9.4649	15.2018	11.00	13.00
K+ (Gengigel)	3	10.3333	1.52753	.88192	6.5388	14.1279	9.00	12.00
k- (Aquades)	3	7.3333	1.52753	.88192	3.5388	11.1279	6.00	9.00
Total	9	10.0000	2.50000	.83333	8.0783	11.9217	6.00	13.00

### ANOVA

TGFb1

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	38.000	2	19.000	9.500	.014
Within Groups	12.000	6	2.000		
Total	50.000	8			

## Post Hoc Tests

### Multiple Comparisons

Dependent Variable: TGFb1

LSD

(I) Kelompok Perlakuan	(J) Kelompok Perlakuan	Mean Difference (I-J)		Sig.	95% Confidence Interval	
			Std. Error		Lower Bound	Upper Bound
P (Moringa oleifera)	K+ (Gengigel)	2.00000	1.15470	.134	-.8255	4.8255
	k- (Aquades)	5.00000	1.15470	.005	2.1745	7.8255
K+ (Gengigel)	P (Moringa oleifera)	-2.00000	1.15470	.134	-4.8255	.8255
	k- (Aquades)	3.00000	1.15470	.041	.1745	5.8255
k- (Aquades)	P (Moringa oleifera)	-5.00000	1.15470	.005	-7.8255	-2.1745
	K+ (Gengigel)	-3.00000	1.15470	.041	-5.8255	-.1745