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## LAMPIRAN

### A. Etik Penelitian

 <p style="text-align: center;">KEMENTERIAN PENDIDIKAN, KEBUDAYAAN, RISET DAN TEKNOLOGI UNIVERSITAS HASANUDDIN FAKULTAS KEDOKTERAN KOMITE ETIK PENELITIAN KESEHATAN RSPTN UNIVERSITAS HASANUDDIN RSUP Dr. WAHIDIN SUDIROHUSODO MAKASSAR Sekretariat : Lantai 2 Gedung Laboratorium Terpadu JL.PERINTIS KEMERDEKAAN KAMPUS TAMALANREA KM.10 MAKASSAR 90245. Contact Person: dr. Agussalim Bukhari.,MMed,PhD.,SpCK TELP. 081241850858, 0411 5780103. Fax : 0411-581431</p> 			
<b>REKOMENDASI PERSETUJUAN ETIK</b>			
Nomor : 371/UN4.6.4.5.31/ PP36/ 2021			
Tanggal: 4 Juni 2021			
Dengan ini Menyatakan bahwa Protokol dan Dokumen yang Berhubungan Dengan Protokol berikut ini telah mendapatkan Persetujuan Etik :			
No Protokol	UH21050342	No Sponsor Protokol	
Peneliti Utama	<b>dr. Pandu Ishaq Nandana, SpU</b>	Sponsor	
Judul Peneliti	Aktivitas Anti-Tumor Senyawa Brucein-D Dari Ekstrak Buah Brucea Javanica Pada Sel Kanker Kandung Kemih T24 Melalui Induksi Ekspresi Gen Pro-Apoptosis Bax, Bak, dan p53serta Hambatan Gen Anti-Apoptosis Bcl-2		
No Versi Protokol	<b>1</b>	Tanggal Versi	<b>28 Mei 2021</b>
No Versi PSP		Tanggal Versi	
Tempat Penelitian	<b>Laboratorium Immuno Biologi Universitas Mataram, Laboratorium Molekuler Fakultas Kedokteran Universitas Mataram</b>		
Jenis Review	<input checked="" type="checkbox"/> Exempted <input type="checkbox"/> Expedited <input type="checkbox"/> Fullboard Tanggal	Masa Berlaku	Frekuensi review lanjutan
		<b>4 Juni 2021</b> sampai <b>4 Juni 2022</b>	
Ketua Komisi Etik Penelitian Kesehatan FKUH	Nama <b>Prof.Dr.dr. Suryani As'ad, M.Sc.,Sp.GK (K)</b>	Tanda tangan	
Sekretaris Komisi Etik Penelitian Kesehatan FKUH	Nama <b>dr. Agussalim Bukhari, M.Med.,Ph.D.,Sp.GK (K)</b>	Tanda tangan	
Kewajiban Peneliti Utama:			
<ul style="list-style-type: none"> <li>• Menyerahkan Amandemen Protokol untuk persetujuan sebelum di implementasikan</li> <li>• Menyerahkan Laporan SAE ke Komisi Etik dalam 24 Jam dan dilengkapi dalam 7 hari dan Laporan SUSAR dalam 72 Jam setelah Peneliti Utama menerima laporan</li> <li>• Menyerahkan Laporan Kemajuan (progress report) setiap 6 bulan untuk penelitian resiko tinggi dan setiap setahun untuk penelitian resiko rendah</li> <li>• Menyerahkan laporan akhir setelah Penelitian berakhir</li> <li>• Melaporkan penyimpangan dari prokol yang disetujui (protocol deviation / violation)</li> <li>• Mematuhi semua peraturan yang ditentukan</li> </ul>			

## B. Statistik

### 1. Oneway ANOVA Jenis dan dosis obat terhadap kematian sel

#### Descriptives

Kematian_sel	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
					Kontrol	3		
0,01Brucein D	3	6.4033	3.15850	1.82356	-1.4429	14.2494	4.08	10.00
0,1 Brucein D	3	26.9549	2.16895	1.25224	21.5669	32.3428	24.64	28.95
1 Brucein D	3	37.8927	.91377	.52757	35.6228	40.1626	36.84	38.50
10 Brucein D	3	55.8798	9.90811	5.72045	31.2667	80.4930	44.44	61.90
100 Brucein D	3	97.1888	1.75771	1.01481	92.8224	101.5552	95.45	98.97
0,01Docetaxel	3	14.4866	3.77592	2.18003	5.1067	23.8666	10.13	16.67
0,1 Docetaxel	3	53.8259	5.97815	3.45149	38.9753	68.6764	47.78	59.73
1 Docetaxel	3	70.4931	6.21968	3.59094	55.0426	85.9437	65.63	77.50
10 Docetaxel	3	85.1594	5.24201	3.02647	72.1376	98.1813	79.38	89.61
0,01 Doxorubicin	3	6.2989	4.85756	2.80451	-5.7680	18.3657	3.33	11.90
0,1 Doxorubicin	3	22.0439	7.30553	4.21785	3.8960	40.1919	15.19	29.73
1 Doxorubicin	3	65.0882	3.33868	1.92759	56.7945	73.3820	62.14	68.72
10 Doxorubicin	3	87.0525	3.00468	1.73475	79.5884	94.5165	83.64	89.29
Total	42	45.6550	31.54616	4.86768	35.8245	55.4855	3.33	98.97

#### Test of Homogeneity of Variances

Kematian_sel			
Levene Statistic	df1	df2	Sig.
2.174	13	28	.042

#### ANOVA

Kematian_sel					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	40101.782	13	3084.752	123.429	.000
Within Groups	699.781	28	24.992		
Total	40801.563	41			



2. Uji Post Hoc Tukey HSD jenis dan dosis obat terhadap kematian sel

Tukey HSD		Kematian_sel							
		Subset for alpha = 0.05							
Dosis_Obat	N	1	2	3	4	5	6	7	8
Dosis 0,01 Doxorubicin	3	6.2989							
Dosis 0,01 Brucein D	3	6.4033							
Kontrol	3	10.4019							
Dosis 0,01 Docetaxel	3	14.4866	14.4866						
Dosis 0,1 Doxorubicin	3		22.0439	22.0439					
Dosis 0,1 Brucein D	3			26.9549					
Dosis 1 Brucein D	3				37.8927				
Dosis 0,1 Docetaxel	3					53.8259			
Dosis 10 Brucein D	3					55.8798	55.8798		
Dosis 1 Doxorubicin	3					65.0882	65.0882		
Dosis 1 Docetaxel	3						70.4931	70.4931	
Dosis 10 Docetaxel	3							85.1594	85.1594
Dosis 10 Doxorubicin	3								87.0525
Dosis 100 Brucein D	3								97.1888
Sig.		.752	.256	.177	.340	.299	.060	.058	.216
Means for groups in homogeneous subsets are displayed.									

### 3. Oneway ANOVA jenis obat terhadap CTCF

Descriptives								
CTCF	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
kontrol	3	10858.8733	683.86200	394.82791	9160.0660	12557.6807	10195.44	11561.48
Brucein D	3	37043.6900	5322.56822	3072.98619	23821.6976	50265.6824	33083.75	43094.15
Docetaxel	3	41198.0367	18413.32342	10630.93723	-4543.1944	86939.2678	21845.02	58499.90
Doxorubicin	3	36413.0133	9204.63999	5314.30138	13547.4200	59278.6067	25879.24	42906.73
Total	12	31378.4033	15462.28196	4463.57632	21554.1381	41202.6686	10195.44	58499.90

Test of Homogeneity of Variances			
CTCF			
Levene Statistic	df1	df2	Sig.
3.191	3	8	.084

ANOVA					
CTCF					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1.725E9	3	5.749E8	5.081	.029
Within Groups	9.051E8	8	1.131E8		
Total	2.630E9	11			

#### 4. Uji Post Hoc Tukey HSD jenis obat terhadap CTCF

CTCF		Multiple Comparisons				
Tukey HSD						
(I) Jenis_obat	(J) Jenis_obat	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
kontrol	Brucein D	-26184.81667	8684.98010	.065	-53997.1801	1627.5468
	Docetaxel	-30339.16333*	8684.98010	.033	-58151.5268	-2526.7999
	Doxorubicin	-25554.14000	8684.98010	.072	-53366.5034	2258.2234
Brucein D	kontrol	26184.81667	8684.98010	.065	-1627.5468	53997.1801
	Docetaxel	-4154.34667	8684.98010	.962	-31966.7101	23658.0168
	Doxorubicin	630.67667	8684.98010	1.000	-27181.6868	28443.0401
Docetaxel	kontrol	30339.16333*	8684.98010	.033	2526.7999	58151.5268
	Brucein D	4154.34667	8684.98010	.962	-23658.0168	31966.7101
	Doxorubicin	4785.02333	8684.98010	.944	-23027.3401	32597.3868
Doxorubicin	kontrol	25554.14000	8684.98010	.072	-2258.2234	53366.5034
	Brucein D	-630.67667	8684.98010	1.000	-28443.0401	27181.6868
	Docetaxel	-4785.02333	8684.98010	.944	-32597.3868	23027.3401

\*. The mean difference is significant at the 0.05 level.

CTCF			
Tukey HSD			
Jenis_obat	N	Subset for alpha = 0.05	
		1	2
kontrol	3	1.08±683.86	
Doxorubicin	3		3.64±9204.63
Brucein D	3		3.70±5322.56
Docetaxel	3		4.11±18413.32
Sig.		.065	.944

Means for groups in homogeneous subsets are displayed.

## 5. Oneway ANOVA jenis obat terhadap Apoptosis

Descriptives								
Apoptosis								
	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
Control	3	9.4259	2.88837	1.66760	2.2507	16.6010	6.77	12.50
Brucein_D	3	56.0482	3.09086	1.78451	48.3700	63.7263	54.17	59.62
Docetaxel	3	74.4272	9.79328	5.65415	50.0994	98.7551	66.10	85.22
Doxorubicin	3	58.9752	12.31312	7.10898	28.3877	89.5627	44.90	67.74
Total	12	49.7191	26.30269	7.59293	33.0072	66.4310	6.77	85.22

Test of Homogeneity of Variances			
Apoptosis			
Levene Statistic	df1	df2	Sig.
3.850	3	8	.057

ANOVA					
Apoptosis					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	7079.309	3	2359.770	35.563	.000
Within Groups	530.835	8	66.354		
Total	7610.144	11			

## 6. Uji Post Hoc Tukey HSD jenis obat terhadap Apoptosis

		Multiple Comparisons				
Apoptosis						
Tukey HSD					95% Confidence Interval	
(I)	(J)	Mean Difference (I-J)	Std. Error	Sig.	Lower	Upper
Jenis_Obat	Jenis_Obat				Bound	Bound
Control	Brucein_D	-46.62230*	6.65103	.001	-67.9212	-25.3234
	Docetaxel	-65.00138*	6.65103	.000	-86.3003	-43.7024
	Doxorubicin	-49.54935*	6.65103	.000	-70.8483	-28.2504
Brucein_D	Control	46.62230*	6.65103	.001	25.3234	67.9212
	Docetaxel	-18.37908	6.65103	.093	-39.6780	2.9199
	Doxorubicin	-2.92705	6.65103	.970	-24.2260	18.3719
Docetaxel	Control	65.00138*	6.65103	.000	43.7024	86.3003
	Brucein_D	18.37908	6.65103	.093	-2.9199	39.6780
	Doxorubicin	15.45203	6.65103	.172	-5.8469	36.7510
Doxorubicin	Control	49.54935*	6.65103	.000	28.2504	70.8483
	Brucein_D	2.92705	6.65103	.970	-18.3719	24.2260
	Docetaxel	-15.45203	6.65103	.172	-36.7510	5.8469

\*. The mean difference is significant at the 0.05 level.

		Apoptosis		
Tukey HSD				
Jenis_Obat	N	Subset for alpha = 0.05		
		1	2	3
Control	3	9.42±2.88		
Brucein_D	3		56.04±3.09	
Doxorubicin	3		58.97±12.31	
Docetaxel	3			74.42±9.79
Sig.		1.000	.672	1.000

Means for groups in homogeneous subsets are displayed.

## C. Dokumentasi

### 1. Brucein D

**COMMERCIAL INVOICE**  
商业发票

TO: dr. Pandu Ishaq Nandana (Mataram University)  
收件人:  
SHIPMENT NO: 77232672422  
运单号:  
CONTACT ADDRESS: Laboratorium Molekuler, Fakultas Kedokteran Univer. JI. Pendidikan No. 37, Mataram, Nusa Tenggara Barat, 83125, Indonesia.

联系人地址:  
TEL: +62-81703380891  
电话:  
FROM: Akin Chemicals, Inc.  
寄件人:  
CONTACT ADDRESS: Yongfeng Plaza, No.52, Yongfeng Road, Hi-tech Zone, Chengdu 610041 P.R.China  
联系人地址:  
TEL: 86-28-85159085

DESCRIPTION OF GOODS 品名	QUANTITY 数量	WEIGHT 重量	UNIT PRICE 单价	VALUE 金额
Brucein D (HS code: 2906199090 )		20mg		USD10.00
SAMPLE ONLY NO COMMERCIAL VALUE. DECLARED VALUE FOR CUSTOMS USE ONLY				
TOTAL 合计				USD10.00

COUNTRY OF ORIGIN : CHENGDU  
原产地:  
REASON FOR EXPORT: SAMPLE  
发件用途:

SIGNATURE:   
寄件人签章

DATE:  
日期: 2020-12-14

### 2. Sel T24

**PT. ELO KARSA UTAMA**  
ELO KARSA UTAMA  
SERVING YOU BETTER

JL. RAYA KE BAYURGAN LAMA 34 E JOMBANG 63122-INDONESIA Phone : +62 21 720 1815, 720 2056, Fax : +62 251 1726 9272 E-mail : info@elokarsa.com

**ORDER CONFIRMATION**

Kepada :	Laboratorium PUSB	No. :	120494 (02/10/2020)
:	Universitas Mataram	Date :	23 Maret 2020
:	Mataram - NTB	Salah :	1 60 Mataram
Attn :	Bapak Dka	No. :	08104723146
No. Telep :	087852006139	Email :	info@elokarsa.com

NO	CAT-PACK	NAMA ALAT/ BAHAN	BRAND	QTY	HARGA @ IDR		
1	92101201-1VL	B16-F1	ECACC	1	9,974,000	14,974,000	
2	83061107-1VL	T24/83	ECACC	1	9,974,000	14,974,000	
						SubTotal 1	29,948,000
						PPN	2,994,800
						<b>TOTAL</b>	<b>32,942,800</b>

\*Sharing yang sudah di order tidak dapat direset

Direktori dan Vertrieb, Health Care Corporation

Divisi Vertrieb

PT. ELO KARSA UTAMA

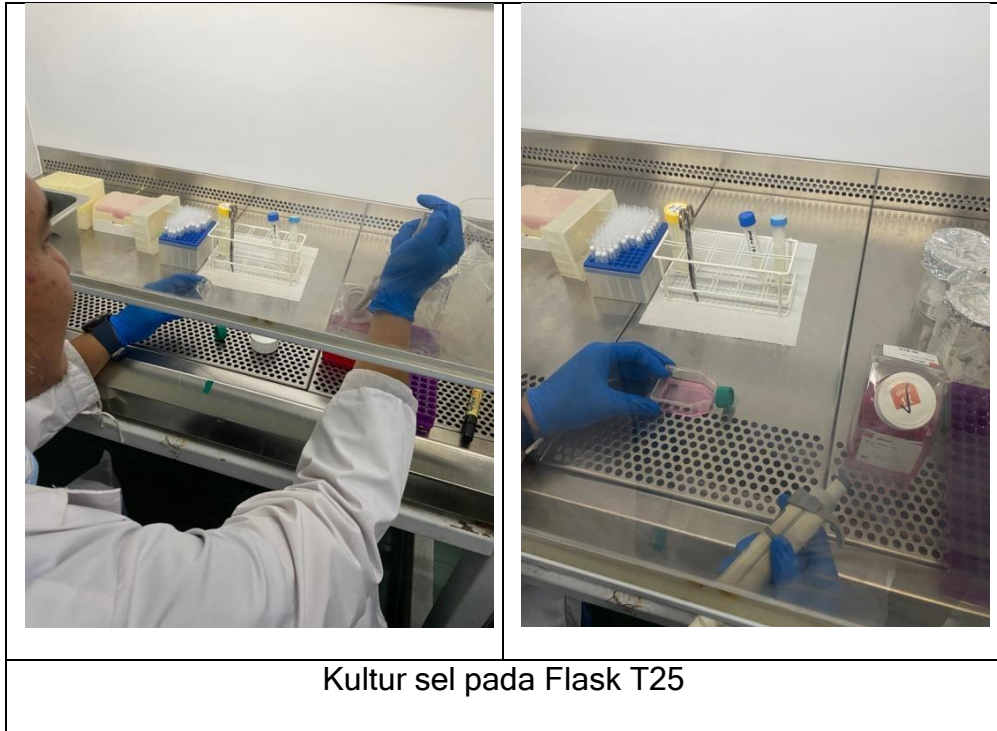
All Indonesia

Jogja Jateng Representative

Invoice Sel T24

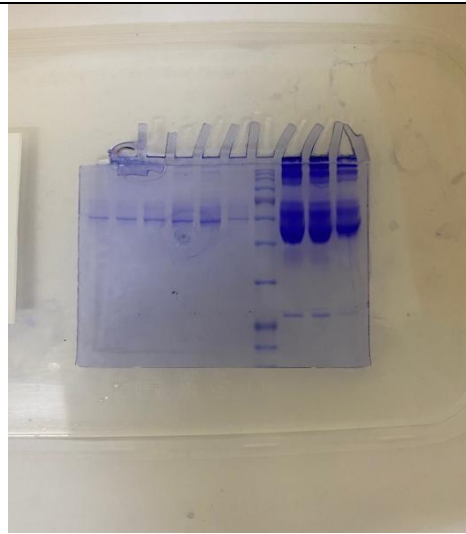
Sel T24

### 3. Dokumentasi Penelitian





Analisis Fragmentasi DNA



Agarose gel electrophoresis



Analisis PCR (Thermal cycler)



Sensoquest Labcycler

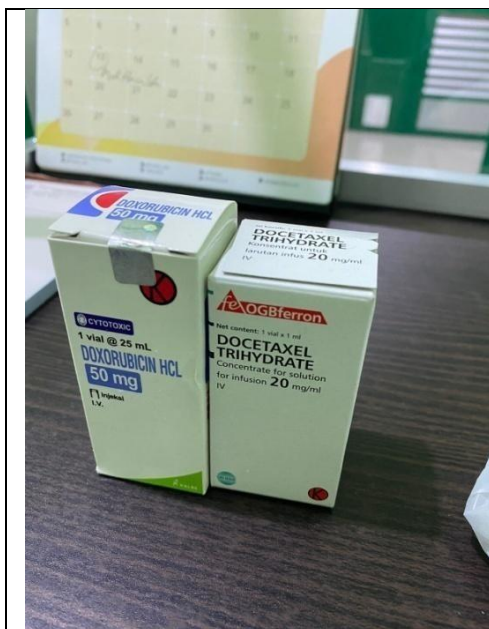




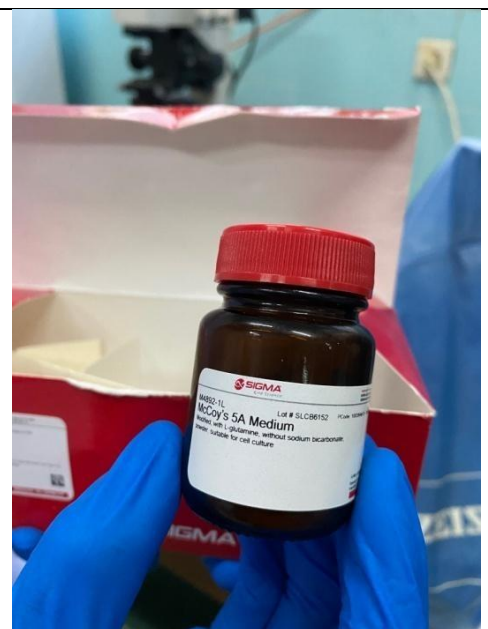
Inkubasi sel T24



Humidified Inkubator



Doxorubicin dan Docetaxel



McCoy Medium



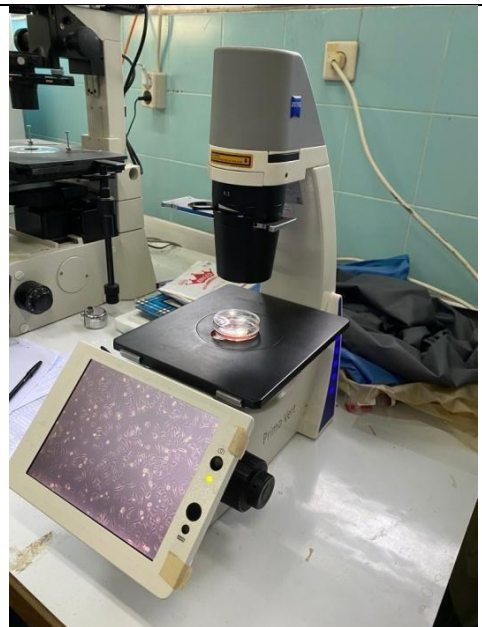
Calcein AM



Hoechst33342



6x DNA Loading Dye



Brightfield Microscope