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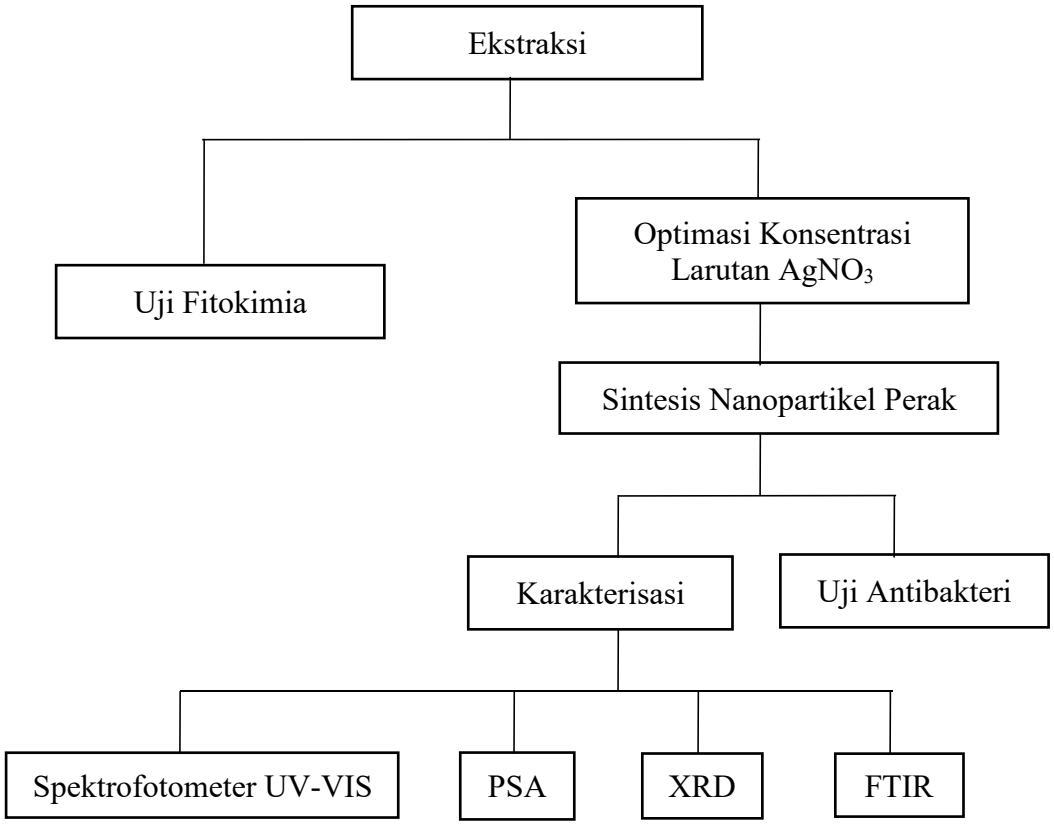
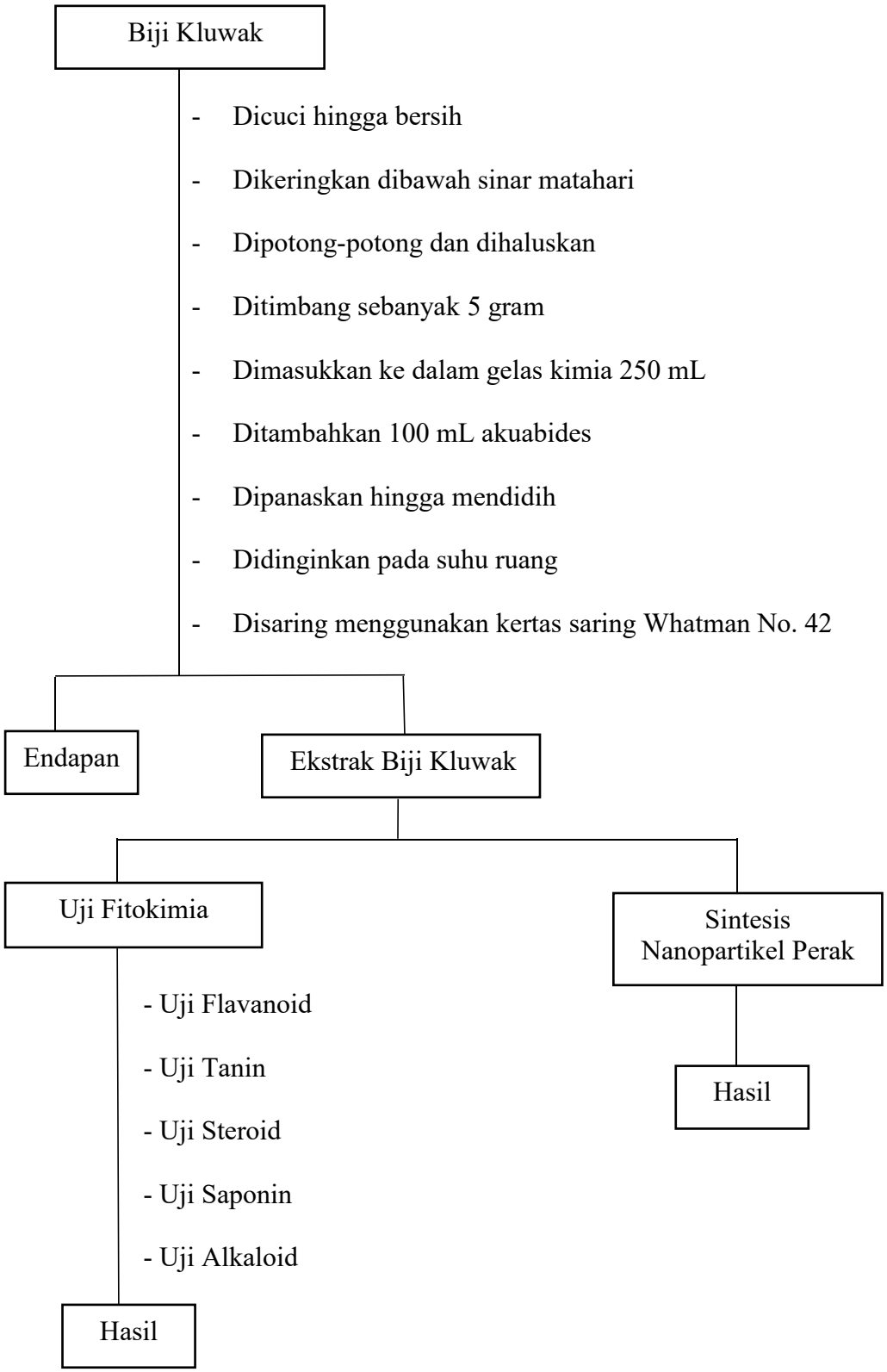
Lampiran 1. Diagram Alur Penelitian

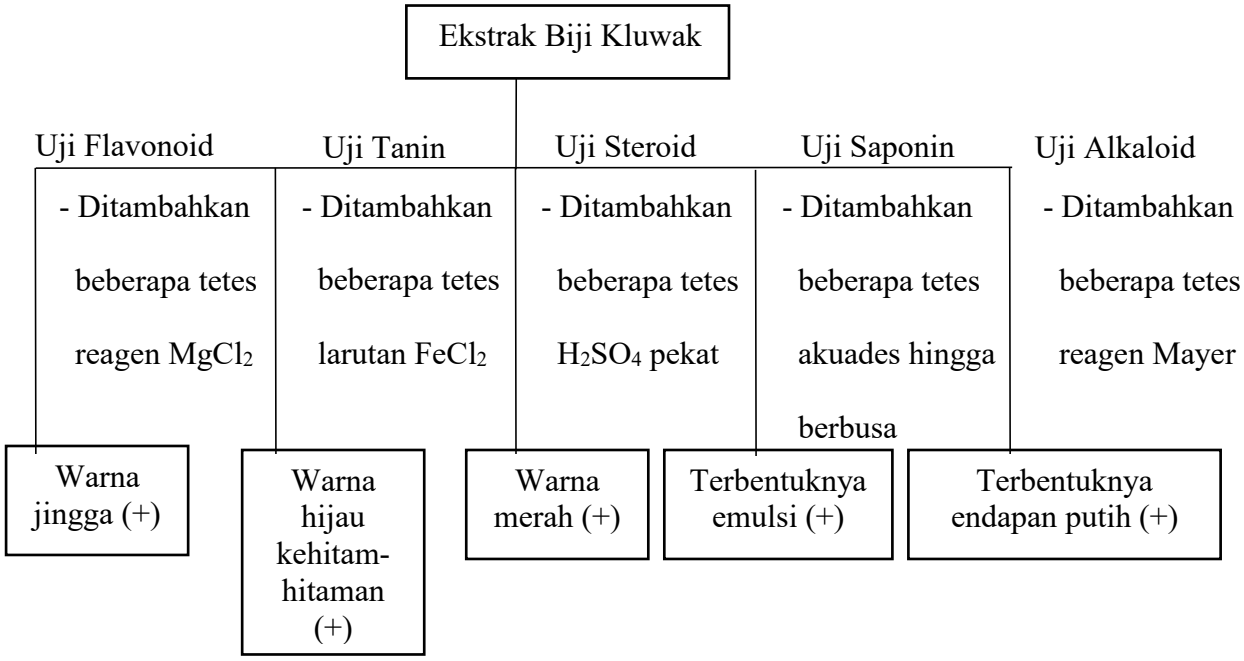


Lampiran 2. Bagan Kerja

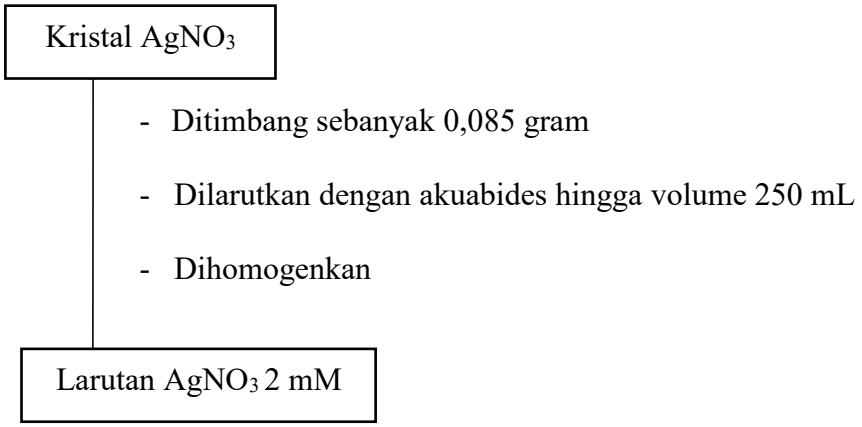
a. Pembuatan Ekstrak Biji Kluwak



b. Uji Fitokimia



c. Pembuatan Larutan $AgNO_3$ 2 mM



d. Pembuatan Larutan $AgNO_3$ 1,5 mM, 1 mM dan 0,5 mM

Larutan $AgNO_3$ 2 mM

- Dipipet masing-masing 37,5 mL, 25 mL, dan 12,5 mL
- Dimasukkan ke dalam labu ukur 50 mL
- Ditambahkan akuabides hingga tanda batas
- Dihomogenkan

Larutan $AgNO_3$ 1,5 mM, 1 mM dan 0,5 mM

e. Optimasi Konsentrasi Larutan $AgNO_3$

Larutan $AgNO_3$ 2 mM, 1,5 mM, 1 mM dan 0,5 mM

Dipipet masing-masing sebanyak 40 mL ke dalam Erlenmeyer 250 mL

- Ditambahkan 1 mL ekstrak biji kluwak
- Diaduk dengan menggunakan *magnetic stirrer* selam 30 menit
- Dianalisis menggunakan Spektrofotometer UV-VIS

Data

7. Sintesis Nanopartikel Perak

40 mL Larutan $AgNO_3$ 2 mM

- Dimasukkan ke daldam Erlenmeyer 125 mL
- Ditambahkan 1 mL ekstrak biji kluwak
- Diaduk menggunakan magnetic stirrer selama 30 menit
- Larutan diaduk hingga berwarna menjadi kuning kecoklatan
- Campuran disentrifugasi pada 1000rpm selama 30 menit

Larutan

Endapan

- Dikeringkan dengan metode *freeze dried*
- Dikarakterisasi mengggunakan FTIR, XRD dan PSA

Data

f. Skema Kerja Pengujian Aktivitas Antibakteri

1) Pembuatan Media Nutrient Agar

Nutrient agar 20 g (5 g pepton, 3 g ekstrak daging, 12 g serbuk agar)

- Ditambahkan akuades 1000ml, dipanaskan lalu diaduk dengan magnetic stirrer

Larutan Nutrien agar

- Dimasukkan dalam erlenmeyer tahan panas ditutup dengan kapas dan kertas sampul coklat

- Disterilkan dalam autoklaf

Media Nutrien agar steril

2) Pembuatan Media Nutrient Broth

Nutrient broth 8 gram (5gr pepton,3 gr ekstrak daging)

- Ditambahkan akuades 1000ml, dipanaskan diaduk dengan magnetic stirrer

Larutan Nutrien broth

- Dimasukkan dalam erlenmeyer tahan panas ditutup dengan kapas dan kertas sampul coklat
- Disterilkan dalam autoklaf

Media Nutrien Broth steril

3) Penyediaan Bakteri dan Suspensi Bakteri

Bakteri Uji

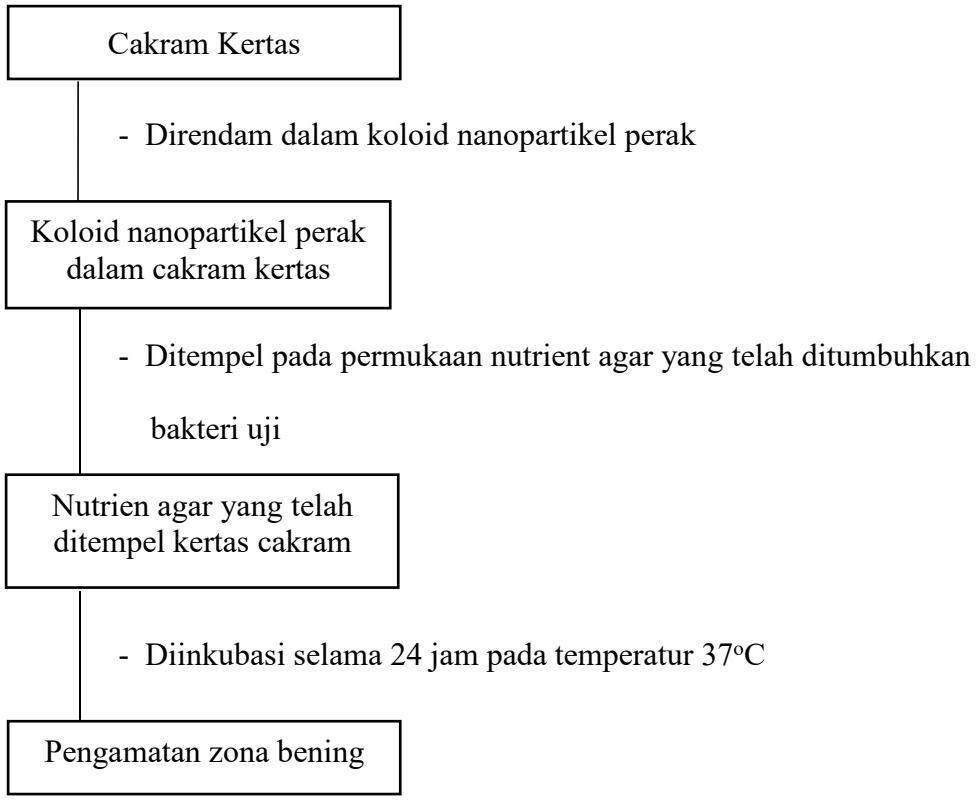
- Ditanamkan di atas permukaan agar miring yang memadat dan diinkbasikan selama 24 jam pada temperatur 37 ^{o}C

Satu Ose koloni sel bakteri

- Diambil dan diinokulasikan dalam 10 ml Nutrient broth
- Diinkubasi selama 24 jam pada temperatur 37 ^{o}C

Suspensi Bakteri

4) Pengujian Antibakteri



Lampiran 3. Persamaan Debye-Scherer

$$D = \frac{K.\lambda}{\beta \cos \theta}$$

Keterangan:

D : Ukuran Kristal (Ǻ)

λ : Panjang Gelombang X-Ray (0,154056 Ǻ)

β : FWMH (*Full Width at Half Maximum*)

θ : Sudut Difraksi (º)

K : Konstanta bentuk kristal (0,89)

a. Untuk 2θ = 37.8403º

$$2\theta = \frac{37,8403}{2}$$

$$\theta = 18,92015$$

$$D = \frac{0,98 \times 0,154178}{\left(\frac{3,14}{180} \times 0.17780\right) \times \cos(18,92015)}$$

$$= \frac{0,151094}{0,003101 \times 0,945971}$$

$$= 51,5071 \text{ nm}$$

b. Untuk 2θ = 44,0814º

$$2\theta = \frac{44,0814}{2}$$

$$\theta = 22,0407$$

$$D = \frac{0,98 \times 0,154178}{\left(\frac{3,14}{180} \times 0,17670\right) \times \cos(22,0407)}$$

$$= \frac{0,151094}{0,003082 \times 0,926917}$$

$$= 52,8900 \text{ nm}$$

c. Untuk 2θ = 64,4544º

$$2\theta = \frac{64,4544}{2}$$

$2\theta = 32,2272$

$$D = \frac{0,98 \times 0,154178}{\left(\frac{3,14}{180} \times 0,19890\right) \times \cos(32,2272)}$$

$$= \frac{0,151094}{0,003469 \times 0,999869}$$

$= 43,5611$ nm

d. Untuk 2θ = 77,5646º

$$2\theta = \frac{77,5646}{2}$$

$2\theta = 38,7823$

$$D = \frac{0,98 \times 0,154178}{\left(\frac{3,14}{180} \times 0,22690\right) \times \cos(38,823)}$$

$$= \frac{0,151094}{0,003958 \times 0,779531}$$

$= 48,9708$ nm

Lampiran 4. Data JCPDS Nanopartikel Perak

PCPDFWIN - [PDF # 030931, Wavelength = 1.5405 (A)]

PDFNumber Search Print View Data Conversion Window Clear Help

03-0931 [Deleted]

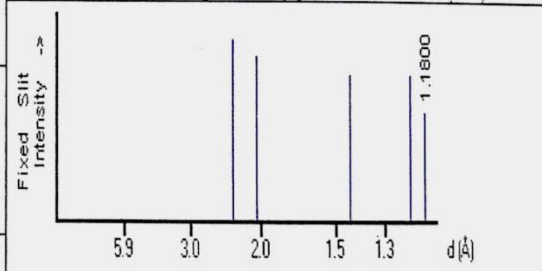
CAS Number:

Molecular Weight: 107.87
Volume[CD]: 67.27
Dx: 10.651 Dm: 10.500

Sys: Cubic
Lattice: Face-centered
S.G.: Fm3m (225)
Cell Parameters:
a 4.067 b c
α β γ

SS/FOM: F 4=3(0.360, 4)
I/Icor:
Rad: CuKa1
Lambda: 1.5405
Filter:
d-sp:

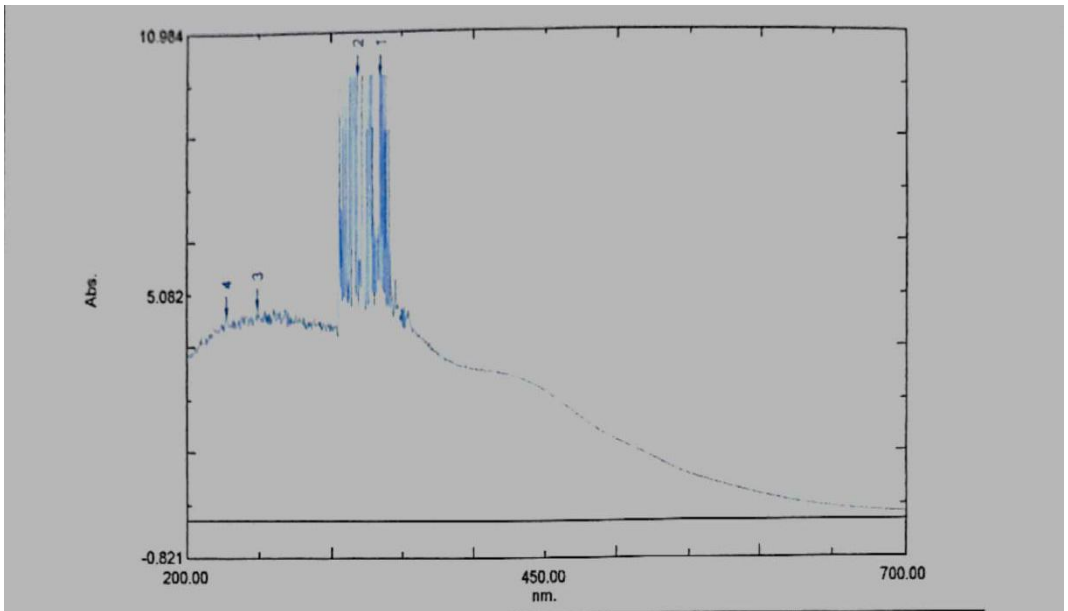
Ag
Silver
Ref: Jung, Z. Kristallogr., Kristallgeom., Kristallphys., Kristallchem., 64, 423 (1926)



d(Å)	Int-f	h	k	l	d(Å)	Int-f	h	k	l	d(Å)	Int-f	h	k	l
2.3200	100	1	1	1	1.4400	80	2	2	0	1.1800	60			
2.0500	90	2	0	0	1.2200	80	3	1	1					

Lampiran 5. Data Hasil Analisis Spektrofotometer UV-VIS

a. Ekstrak Biji Kluwak

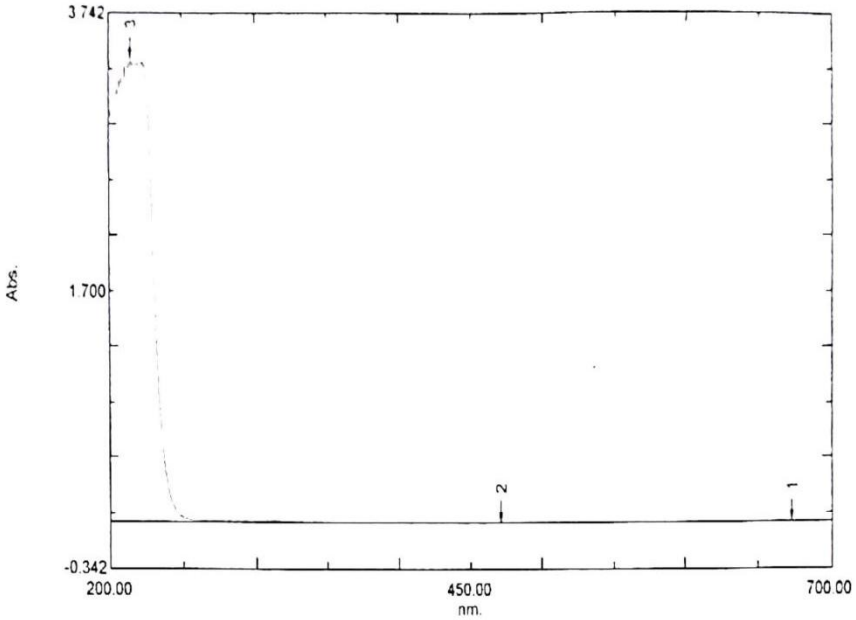


[Measurement Properties]

Wavelength Range (nm.):	200.00 to 700.00
Scan Speed:	Medium
Sampling Interval:	0.5
Auto Sampling Interval:	Enabled
Scan Mode:	Single

No.	P/V	Wavelength	Abs.	Description
1	↑	335.00	10.000	
2	↑	318.50	10.000	
3	↑	249.50	4.745	
4	↑	227.50	4.587	

b. $AgNO_3$ 2 mM

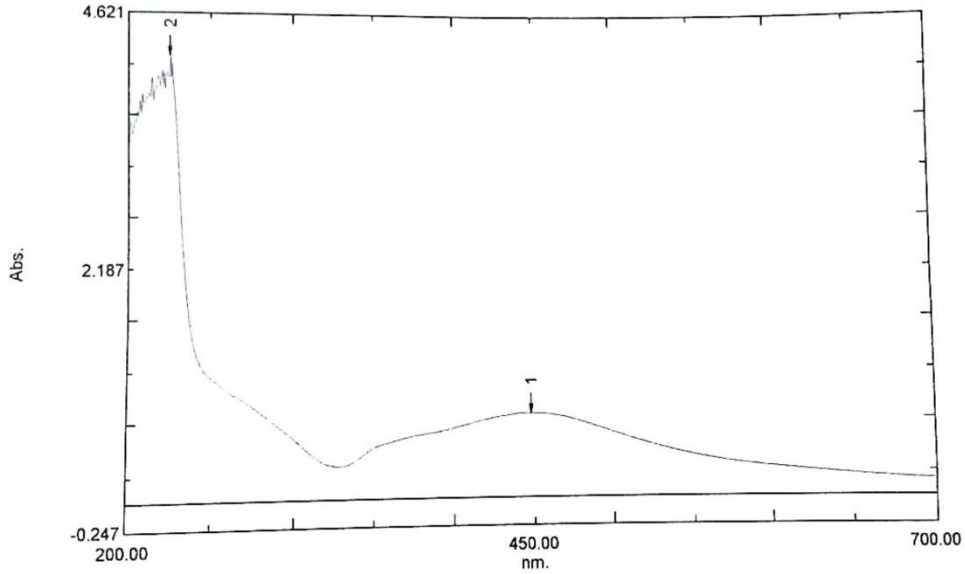


[Measurement Properties]

Wavelength Range (nm.):	200.00 to 700.00
Scan Speed:	Medium
Sampling Interval:	0.5
Auto Sampling Interval:	Enabled
Scan Mode:	Single

No.	P/V	Wavelength	Abs.	Description
1	↑	673.50	0.001	
2	↑	471.00	0.004	
3	↑	214.50	3.402	

c. Nanopartikel Perak Hari 1

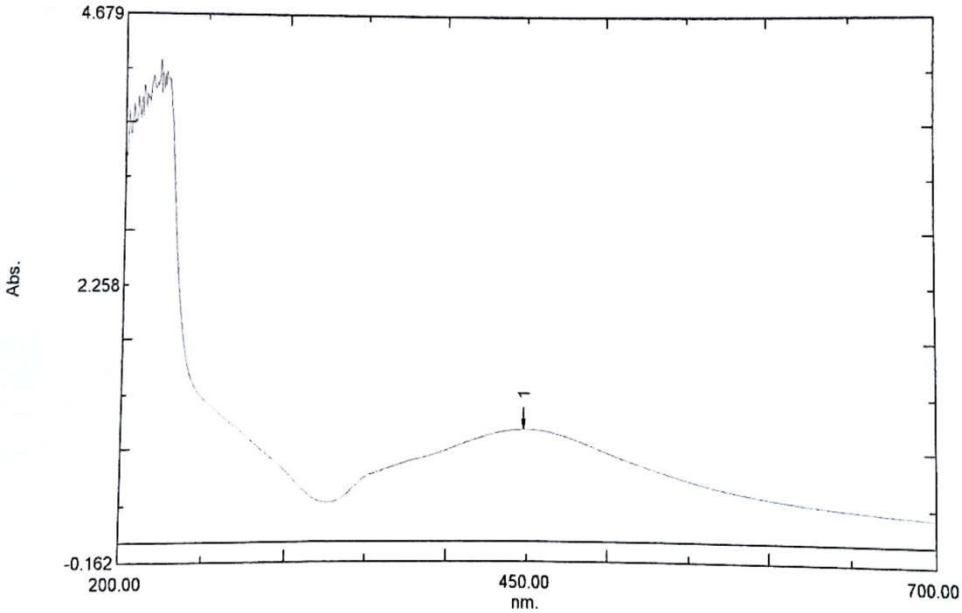


[Measurement Properties]

Wavelength Range (nm.):	200.00 to 700.00
Scan Speed:	Medium
Sampling Interval:	0.5
Auto Sampling Interval:	Enabled
Scan Mode:	Single

No.	P/V	Wavelength	Abs.	Description
1	↑	448.00	0.789	
2	↑	225.50	4.216	

d. Nanopartikel Perak Hari 3

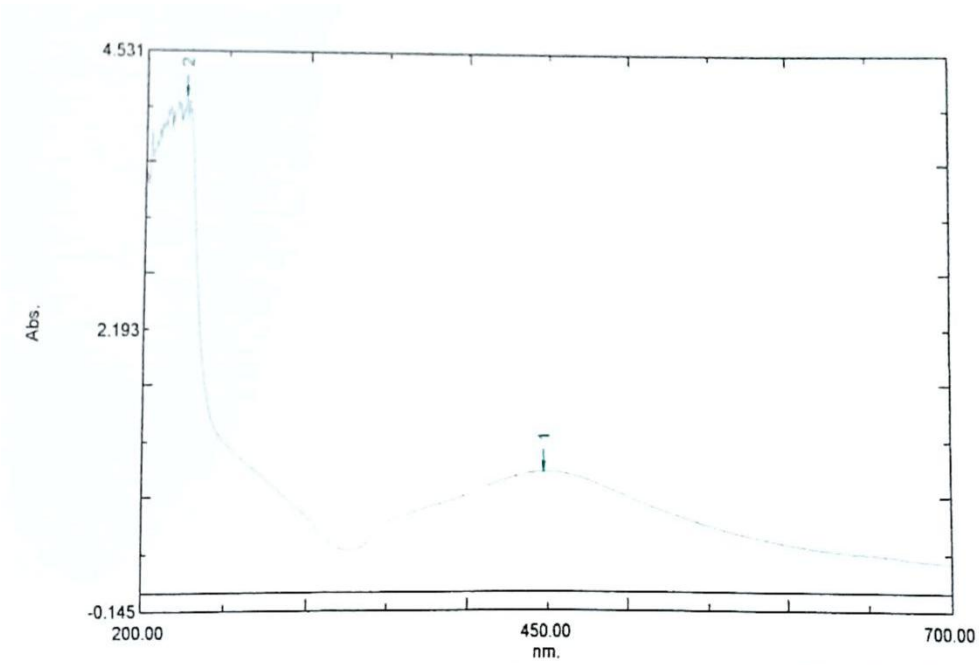


[Measurement Properties]

Wavelength Range (nm.):	200.00 to 700.00
Scan Speed:	Medium
Sampling Interval:	0.5
Auto Sampling Interval:	Enabled
Scan Mode:	Single

No.	P/V	Wavelength	Abs.	Description
1	↑	448.00	0.978	

e. Nanopartikel Perak Hari 5

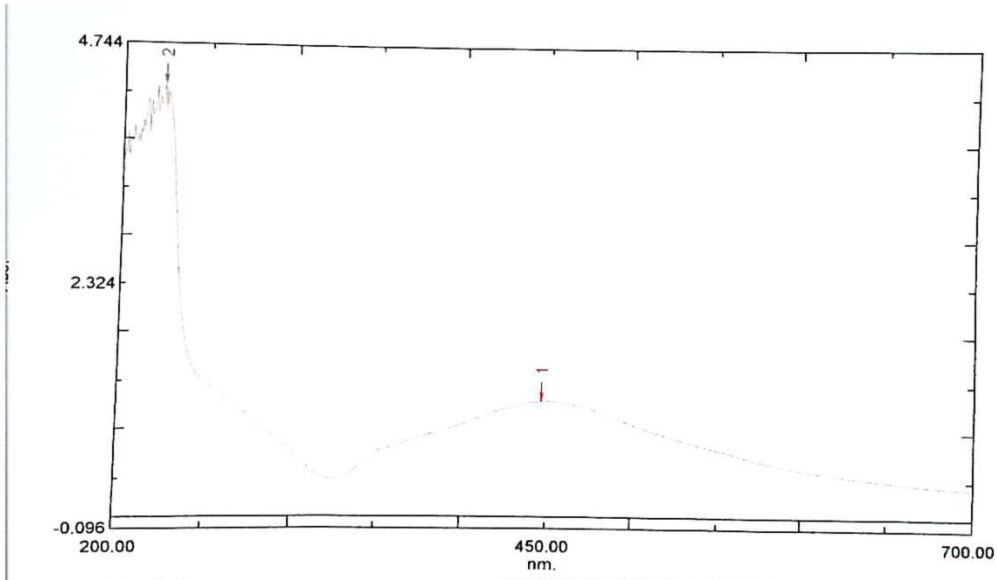


[Measurement Properties]

Wavelength Range (nm.):	200.00 to 700.00
Scan Speed:	Medium
Sampling Interval:	0.5
Auto Sampling Interval:	Enabled
Scan Mode:	Single

No.	P/V	Wavelength	Abs.	Description
1		447.00	0.993	
2		224.00	4.141	

f. Nanopartikel Perak Hari 8

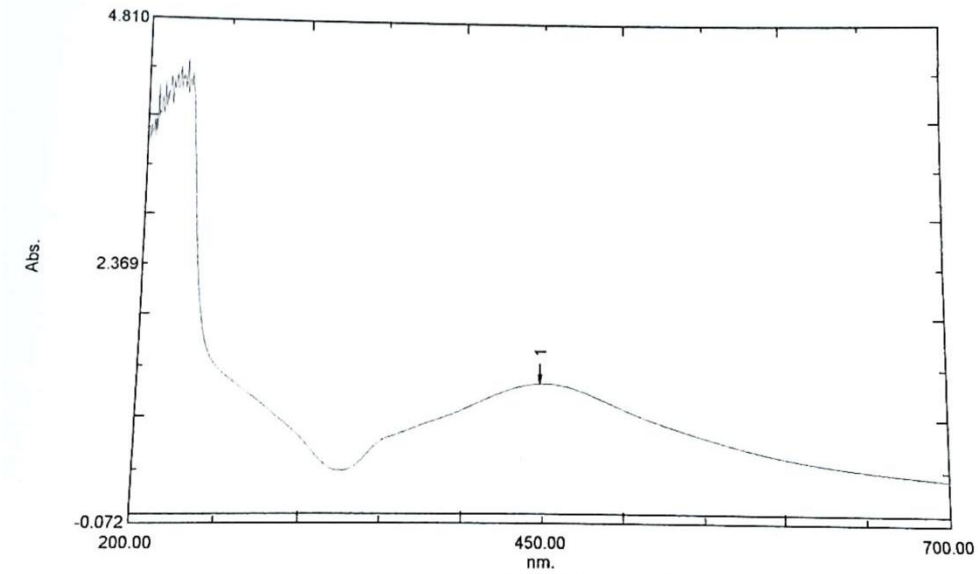


asurement Properties]

velength Range (nm.):	200.00 to 700.00
n Speed:	Medium
npling Interval:	0.5
o Sampling Interval:	Enabled
n Mode:	Single

No.	P/V	Wavelength	Abs.	Description
1		447.50	1.160	
2		223.00	4.341	

g. Nanopartikel Perak Hari 11

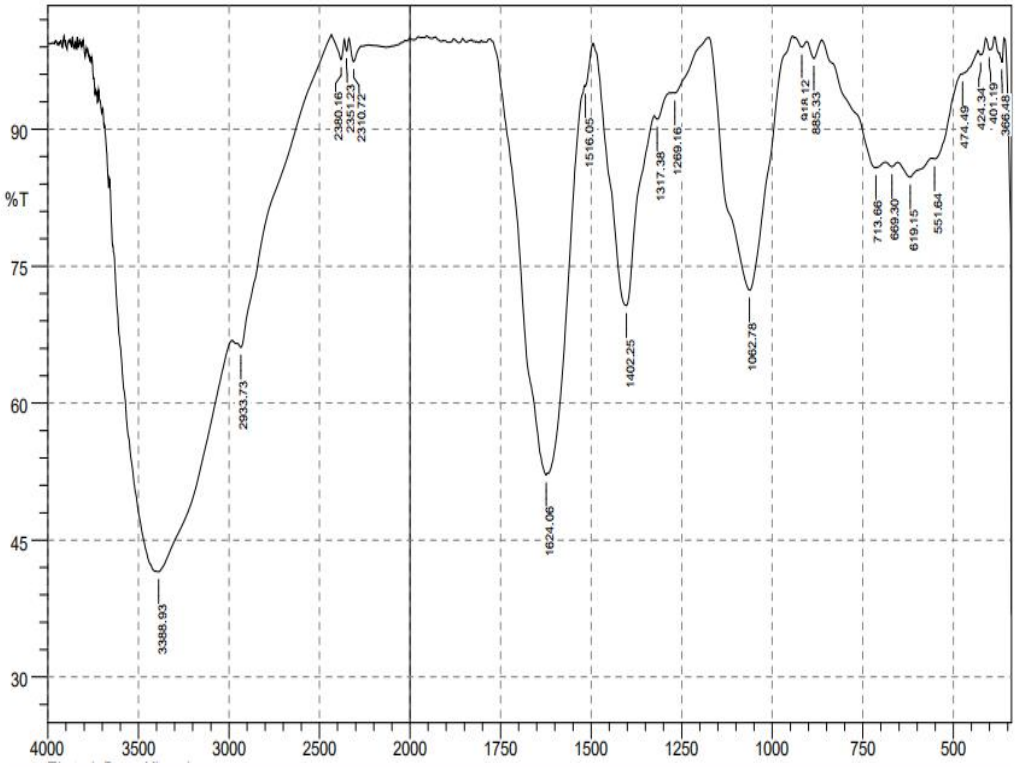


[Measurement Properties]

Wavelength Range (nm.):	200.00 to 700.00
Scan Speed:	Medium
Sampling Interval:	0.5
Auto Sampling Interval:	Enabled
Scan Mode:	Single

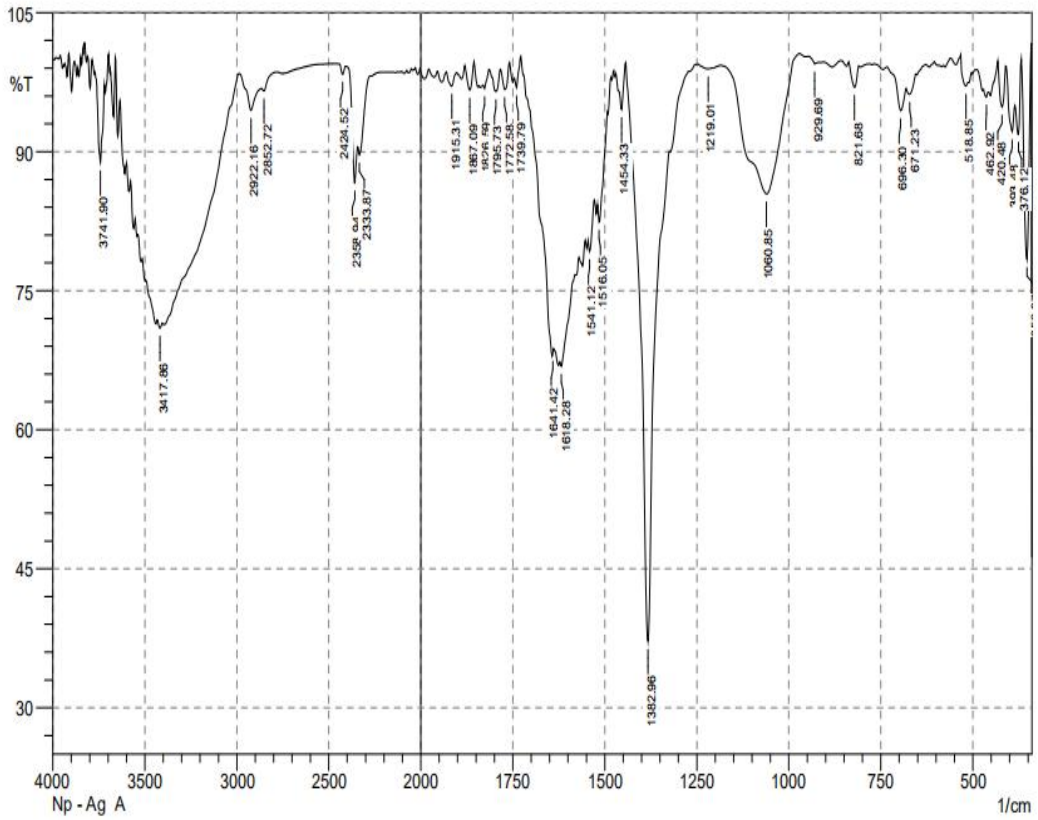
No.	P/V	Wavelength	Abs.	Description
1	1	448.50	1.228	

Lampiran 6. Spektrum FTIR Ekstrak Biji Kluwak



	Peak	Intensity	Corr. Intensity	Base (H)	Base (L)	Area	Corr. Area
1	366.48	97.352	1.781	372.26	360.69	0.099	0.055
2	401.19	98.671	1.334	410.84	385.76	0.08	0.082
3	424.34	98.133	0.965	432.05	410.84	0.134	0.067
4	474.49	96.038	0.128	476.42	432.05	0.581	0.069
5	551.64	86.753	0.936	559.36	478.35	3.445	0.326
6	619.15	84.745	1.784	653.87	561.29	6.224	0.435
7	669.3	85.895	0.458	684.73	655.8	1.88	0.037
8	713.66	85.784	2.648	862.18	686.66	6.71	0.967
9	885.33	97.754	1.927	904.61	864.11	0.233	0.178
10	918.12	98.989	0.8	937.4	904.61	0.08	0.055
11	1062.78	72.371	27.72	1174.65	945.12	16.405	16.497
12	1269.16	93.966	0.26	1273.02	1176.58	1.243	0.057
13	1317.38	91.089	0.845	1325.1	1282.66	1.435	0.068
14	1402.25	70.684	24.317	1492.9	1327.03	13.366	9.853
15	1516.05	94.686	0.508	1517.98	1496.76	0.319	0.041
16	1624.06	52.14	1.375	1778.37	1620.21	20.673	0.699
17	2310.72	97.401	2.239	2337.72	2264.43	0.489	0.338
18	2351.23	98.579	1.32	2362.8	2337.72	0.091	0.081
19	2380.16	97.632	2.342	2434.17	2362.8	0.327	0.359
20	2933.73	66.119	1.679	2953.02	2436.09	37.916	0.291
21	3388.93	41.542	0.37	3394.72	2983.88	118.008	4.251

Lampiran 7. Spektrum FTIR Nanopartikel Perak



	Peak	Intensity	Corr. Intensity	Base (H)	Base (L)	Area	Corr. Area
1	352.97	78.373	21.292	368.4	343.33	1.5	1.468
2	376.12	91.861	5.892	385.76	368.4	0.436	0.24
3	393.48	92.156	4.136	410.84	385.76	0.587	0.265
4	420.48	94.824	4.65	432.05	410.84	0.328	0.282
5	462.92	95.884	0.625	470.63	459.06	0.197	0.021
6	518.85	97.086	1.274	532.35	513.07	0.197	0.108
7	671.23	96.177	1.192	680.87	653.87	0.373	0.087
8	696.3	94.417	2.896	732.95	682.8	0.693	0.217
9	821.68	96.946	2.49	837.11	810.1	0.235	0.173
10	929.69	99.468	0.563	947.05	904.61	0.044	0.039
11	1060.85	85.441	14.595	1184.29	972.12	7.037	6.979
12	1219.01	98.934	0.157	1247.94	1211.3	0.144	0.019
13	1382.96	37.17	57.577	1442.75	1325.1	16.795	13.972
14	1454.33	94.557	4.548	1469.76	1442.75	0.382	0.266
15	1516.05	82.389	3.365	1519.91	1485.19	1.734	0.333
16	1541.12	79.21	2.293	1544.98	1529.55	1.391	0.126
17	1618.28	66.861	1.353	1622.13	1581.63	5.884	0.147
18	1641.42	68.259	1.172	1728.22	1639.49	6.332	0.136
19	1739.79	96.896	1.797	1745.58	1728.22	0.132	0.061
20	1772.58	96.719	2.506	1784.15	1759.08	0.239	0.157
21	1795.73	96.48	2.399	1815.02	1784.15	0.325	0.174
22	1826.59	96.824	0.767	1830.45	1815.02	0.163	0.029
23	1867.09	96.655	2.844	1880.6	1855.52	0.239	0.182
24	1915.31	97.07	1.477	1930.74	1903.74	0.27	0.099
25	2333.87	89.625	1.768	2341.58	2276	1.782	0.215
26	2358.94	86.631	5.992	2407.16	2343.51	1.878	0.53
27	2424.52	98.35	0.956	2457.31	2407.16	0.209	0.067
28	2852.72	96.551	0.56	2866.22	2787.14	0.784	0.043
29	2922.16	94.463	3.08	2985.81	2868.15	1.913	0.668
30	3417.86	70.97	0.675	3431.36	3406.29	3.695	0.063
31	3741.9	88.806	8.974	3770.84	3720.69	1.537	1.064

Lampiran 8. Hasil Analisis XRD

*** Basic Data Process ***

Group : Standard
Data : CHem2020#AgNan

Strongest 3 peaks

no.	peak no.	2Theta (deg)	d (A)	I/I1	FWHM (deg)	Intensity (Counts)	Integrated Int (Counts)
1	5	44.0814	2.05268	100	0.17670	1942	18985
2	13	77.5646	1.22980	78	0.22690	1520	18560
3	8	64.4544	1.44446	63	0.19890	1231	14044

Peak Data List

peak no.	2Theta (deg)	d (A)	I/I1	FWHM (deg)	Intensity (Counts)	Integrated Int (Counts)
1	33.9965	2.63492	15	0.15480	285	2826
2	37.8403	2.37564	63	0.17780	1215	12928
3	39.5603	2.27622	25	0.15160	486	4348
4	43.7600	2.06701	4	0.14220	78	1264
5	44.0814	2.05268	100	0.17670	1942	18985
6	57.5306	1.60071	18	0.16160	346	3106
7	57.8762	1.59197	3	0.10520	59	324
8	64.4544	1.44446	63	0.19890	1231	14044
9	64.7400	1.43878	3	0.09420	58	707
10	68.8525	1.36253	21	0.19040	404	4223
11	69.2791	1.35518	4	0.13240	77	558
12	77.1200	1.23577	3	0.28000	58	1764
13	77.5646	1.22980	78	0.22690	1520	18560
14	77.9800	1.22428	4	0.11140	71	900

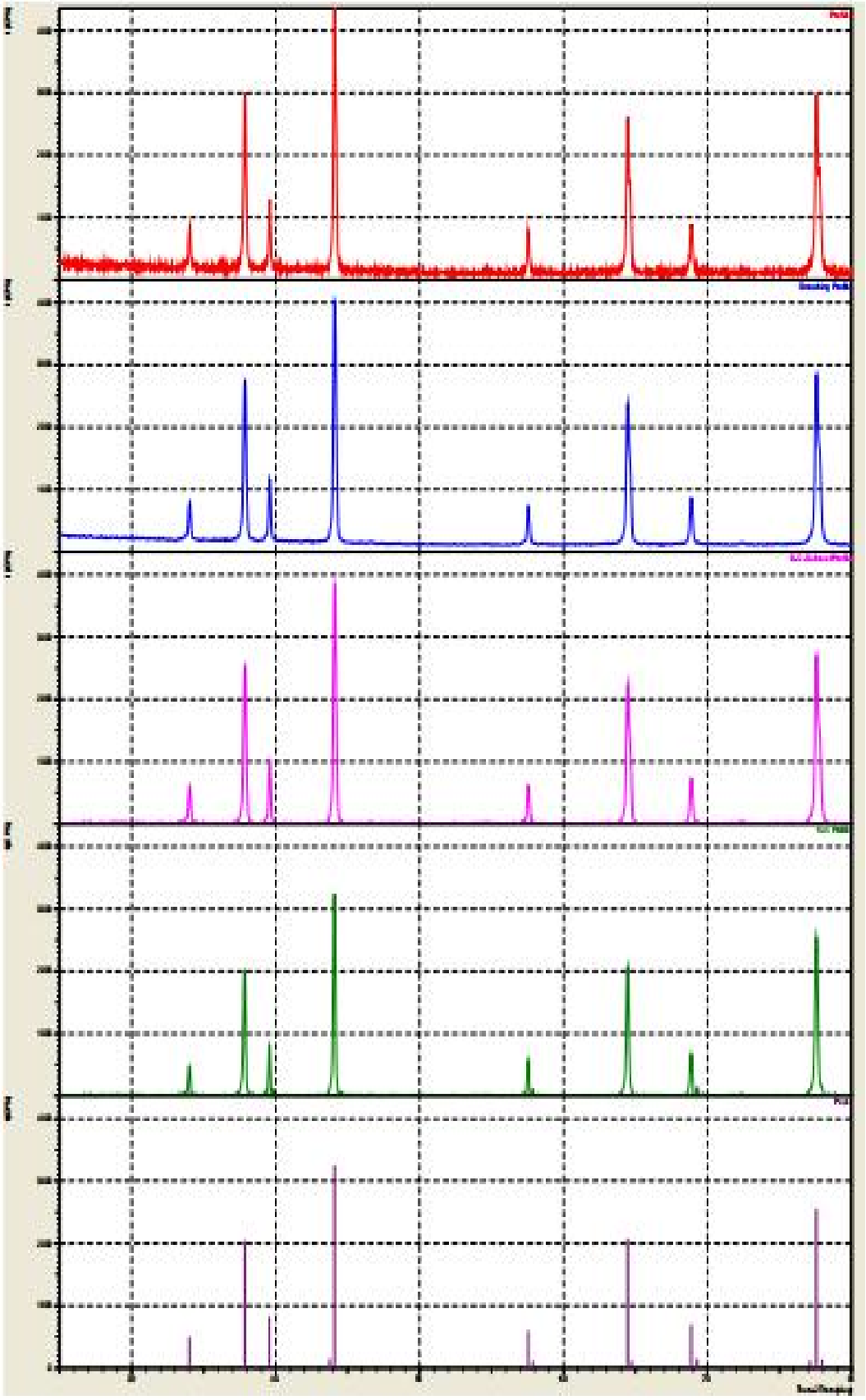
```
        ***  Basic Data Process  ***

# Data Infomation
        Group               : Standard
        Data                : CHem2020#AgNan
        Sample Nmae         : serbuk
        Comment             :
        Date & Time         : 10-13-20 07:16:10

# Measurement Condition
   X-ray tube
        target              : Cu
        voltage             : 40.0 (kV)
        current             : 30.0 (mA)
   Slits
        Auto Slit           : Used
        divergence slit     :   1.00000 (deg)
        scatter slit        :   1.00000 (deg)
        receiving slit      :   0.30000(mm)
   Scanning
        drive axis          : Theta-2Theta
        scan range          :   25.0200 - 80.0000 (deg)
        scan mode           : Continuous Scan
        scan speed          :    2.0000 (deg/min)
        sampling pitch      :    0.0200 (deg)
        preset time         :    0.60 (sec)

# Data Process Condition
   Smoothing                [ AUTO ]
        smoothing points    : 13
   B.G.Subtruction          [ AUTO ]
        sampling points     : 13
        repeat times        : 30
   Kal-a2 Separate          [ MANUAL ]
        Kal a2 ratio        : 50 (%)
   Peak Search              [ AUTO ]
        differential points : 11
        FWHM threhold       : 0.050 (deg)
        intensity threhold  : 30 (par mil)
        FWHM ratio  (n-1)/n : 2
   System error Correction  [ NO ]
   Precise peak Correction  [ NO ]
```

< Group: Standard Data: Chem2020#Aglian >



Lampiran 9. Hasil Analisis PSA

BECKMAN COULTER

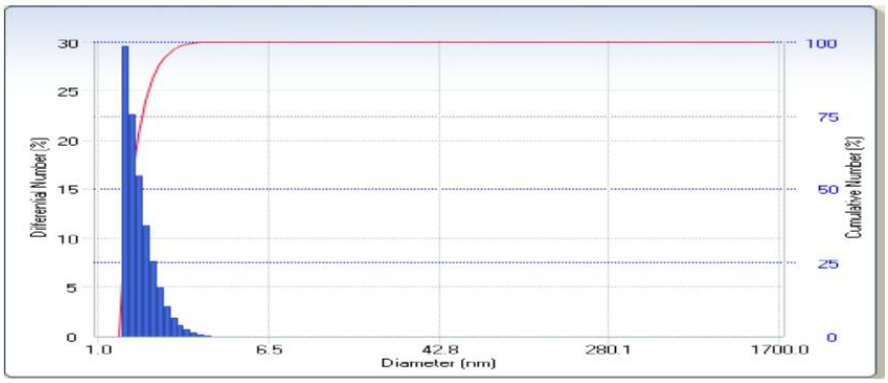
Delsa™ Nano

Number Distribution — Common

User	: Common	Group	:	Repetition	: 1/1
Date	: 10/2/2020	File Name	: AgNp2_20201002_14044		
Time	: 14:04:47	Sample Information	:		
SOP Name	: Nano				

Version 1.34 / 2.00

Number Distribution



Distribution Results (Contin)

Peak	Diameter (nm)	Std. Dev.
1	1.6	0.3
2	48.4	11.5
3	0.0	0.0
4	0.0	0.0
5	0.0	0.0
Average	1.6	0.3

Residual : 1.963e-002 (O.K)

Cumulants Results

Diameter	(d)	: 58.0	(nm)
Polydispersity Index	(P.I.)	: 0.599	
Diffusion Const.	(D)	: 8.486e-008	(cm^2/sec)

Measurement Condition

Temperature	: 25.0	(°C)
Diluent Name	: WATER	
Refractive Index	: 1.3328	
Viscosity	: 0.8878	(cP)
Scattering Intensity	: 6859	(cps)

Number Distribution Table

d (nm)	f(%)	f(cum.%)	d (nm)	f(%)	f(cum.%)	d (nm)	f(%)	f(cum.%)	d (nm)	f(%)	f(cum.%)
1.0	0.0	0.0	6.5	0.0	100.0	42.8	0.0	100.0	280.1	0.0	100.0
1.1	0.0	0.0	7.1	0.0	100.0	46.1	0.0	100.0	302.0	0.0	100.0
1.2	0.0	0.0	7.6	0.0	100.0	49.8	0.0	100.0	325.5	0.0	100.0
1.3	0.0	0.0	8.2	0.0	100.0	53.6	0.0	100.0	350.9	0.0	100.0
1.4	29.6	29.6	8.8	0.0	100.0	57.8	0.0	100.0	378.3	0.0	100.0
1.5	22.7	52.3	9.5	0.0	100.0	62.3	0.0	100.0	407.8	0.0	100.0
1.6	16.4	68.7	10.3	0.0	100.0	67.2	0.0	100.0	439.6	0.0	100.0
1.7	11.4	80.1	11.1	0.0	100.0	72.4	0.0	100.0	473.9	0.0	100.0
1.8	7.6	87.7	11.9	0.0	100.0	78.1	0.0	100.0	510.9	0.0	100.0
2.0	4.9	92.6	12.9	0.0	100.0	84.2	0.0	100.0	550.8	0.0	100.0
2.1	3.1	95.7	13.9	0.0	100.0	90.8	0.0	100.0	593.8	0.0	100.0
2.3	1.9	97.5	15.0	0.0	100.0	97.8	0.0	100.0	640.1	0.0	100.0
2.5	1.1	98.7	16.1	0.0	100.0	105.5	0.0	100.0	690.0	0.0	100.0
2.7	0.7	99.3	17.4	0.0	100.0	113.7	0.0	100.0	743.9	0.0	100.0
2.9	0.4	99.7	18.7	0.0	100.0	122.6	0.0	100.0	801.9	0.0	100.0
3.1	0.2	99.9	20.2	0.0	100.0	132.1	0.0	100.0	864.5	0.0	100.0
3.3	0.1	100.0	21.8	0.0	100.0	142.4	0.0	100.0	932.0	0.0	100.0
3.6	0.0	100.0	23.5	0.0	100.0	153.6	0.0	100.0	1004.7	0.0	100.0
3.9	0.0	100.0	25.3	0.0	100.0	165.5	0.0	100.0	1083.1	0.0	100.0
4.2	0.0	100.0	27.3	0.0	100.0	178.5	0.0	100.0	1167.6	0.0	100.0

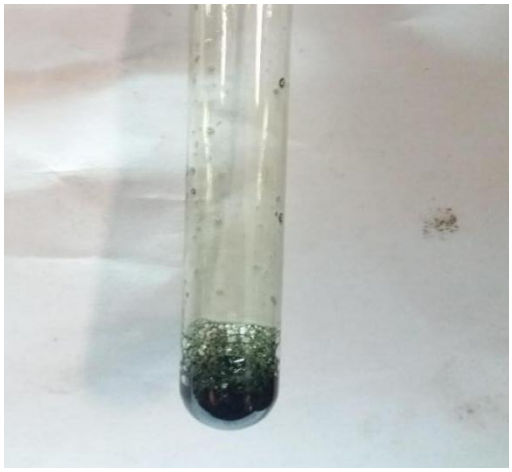
D (10%) : 1.3 (nm) | D (50%) : 1.4 (nm) | D (90%) : 1.9 (nm)

Number Distribution Table

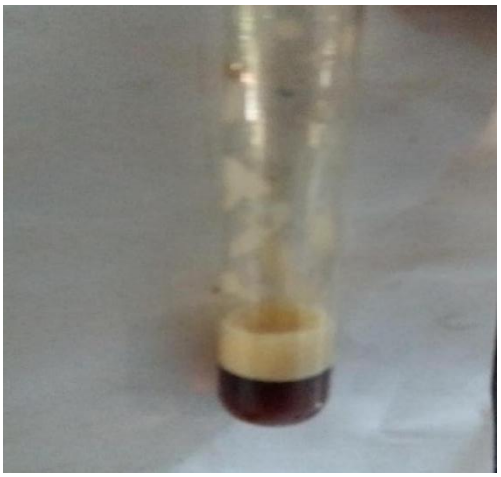
d (nm)	f(%)	f(cum.%)	d (nm)	f(%)	f(cum.%)	d (nm)	f(%)	f(cum.%)	d (nm)	f(%)	f(cum.%)
4.5	0.0	100.0	29.4	0.0	100.0	192.4	0.0	100.0	1258.7	0.0	100.0
4.8	0.0	100.0	31.7	0.0	100.0	207.4	0.0	100.0	1356.9	0.0	100.0
5.2	0.0	100.0	34.2	0.0	100.0	223.6	0.0	100.0	1462.8	0.0	100.0
5.6	0.0	100.0	36.8	0.0	100.0	241.0	0.0	100.0	1577.0	0.0	100.0
6.1	0.0	100.0	39.7	0.0	100.0	259.8	0.0	100.0	1700.0	0.0	100.0

D (10%) : 1.3 (nm)	D (50%) : 1.4 (nm)	D (90%) : 1.9 (nm)

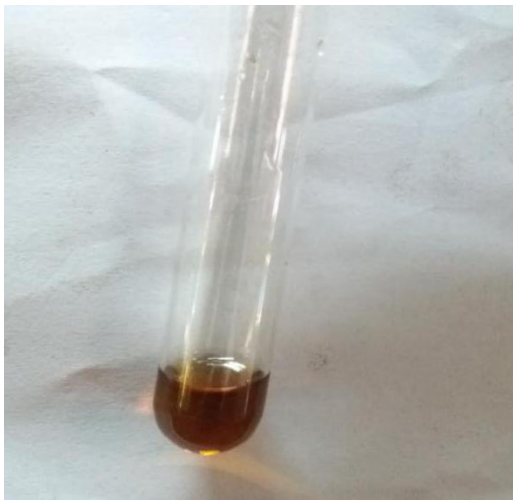
Lampiran 10. Hasil Pengujian Fitokimia



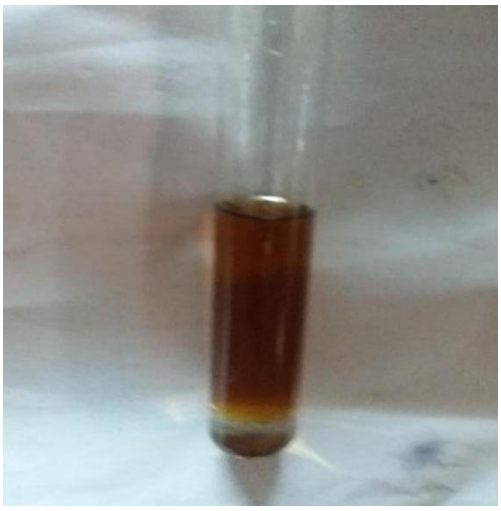
Gambar 7**.** Tanin



Gambar 8**.** Saponin



Gambar 9. Flavonoid

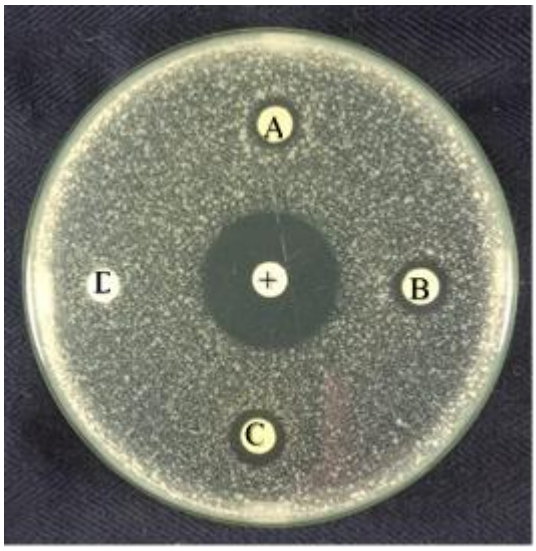


Gambar 10. Terpenoid

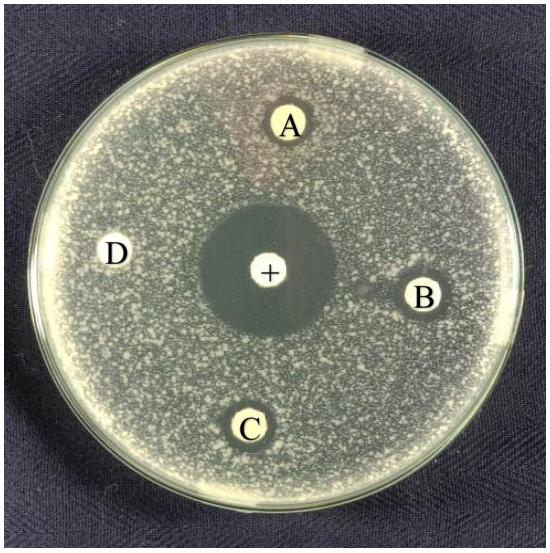
Gambar 11**.** Steroid

Gambar 12. Alkaloid

Lampiran 11. Hasil Pengujian Antibakteri



Gambar 13. *Pseudomonas aeruginosa*



Gambar 14. *Bacillus subtilis*