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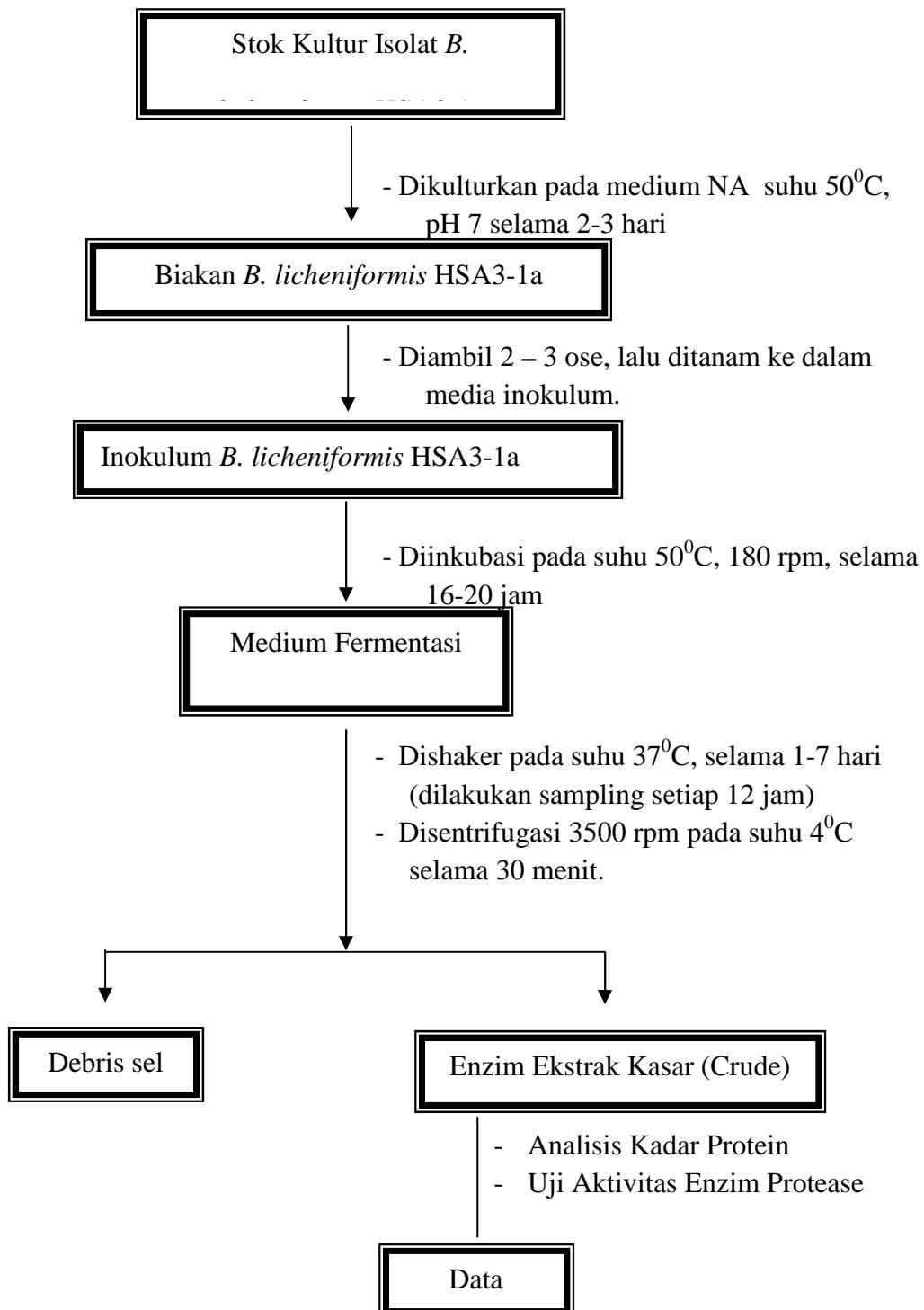
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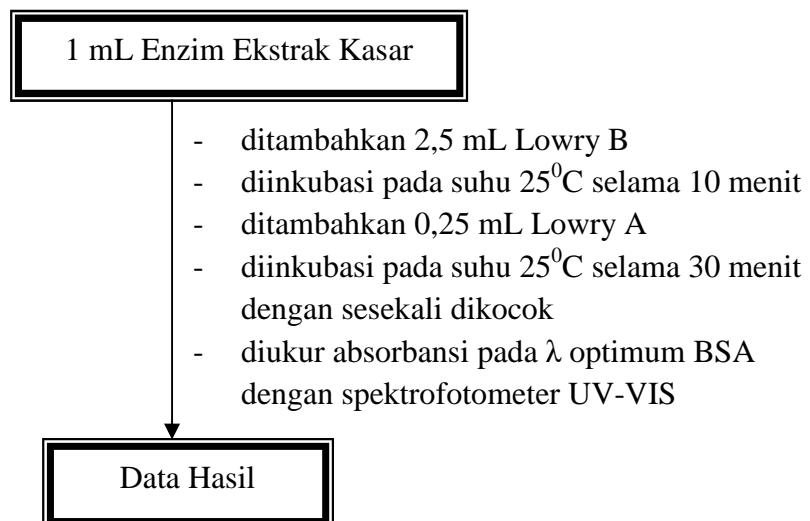
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Lampiran 1. Skema Isolasi Protease dari *B. licheniformis* HSA3-1a



## Lampiran 2. Penentuan Kadar Protein dengan Metode Lowry



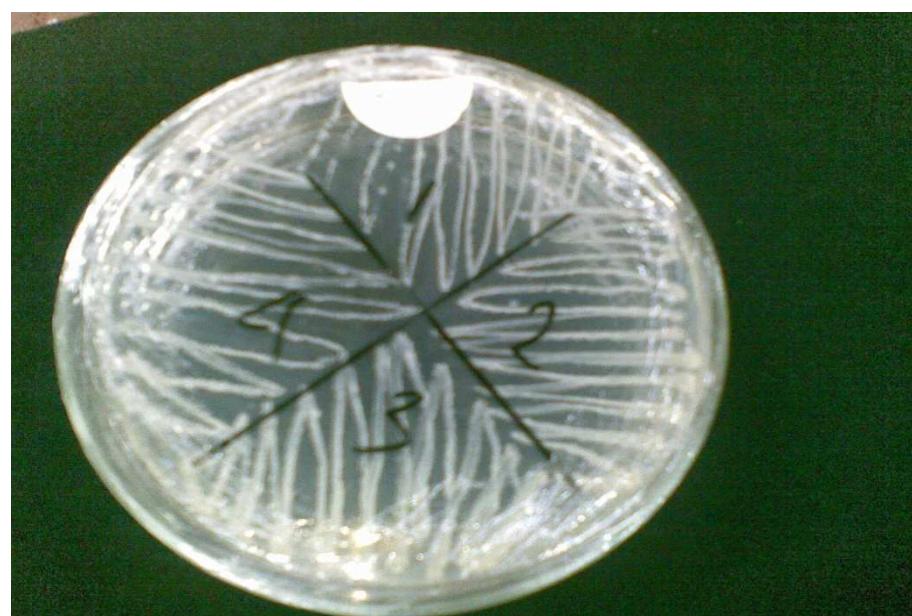
Lampiran 3. Skema Pengujian Aktivitas Enzim Protease

Pereaksi	Sampel (mL)	Blanko (mL)	Standard (mL)
Bufer (0,2 M), pH 7	0,5	0,5	0,5
Substrat kasein 1%, pH 7	0,5	0,5	0,5
Enzim	0,1	-	-
Tirosin standar	-	-	0,1
Akuades	-	0,1	-
Inkubasi pada suhu 50°C selama 10 menit			
TCA (0,1 M)	1	1	1
Akuades	0,1	-	-
Enzim	-	0,1	0,1
Didiamkan pada suhu 50°C selama 10 menit, dan disentrifuge 10.000 rpm selama 10 menit			
Filtrat	0,75	0,75	0,75
Na <sub>2</sub> CO <sub>3</sub>	2,5	2,5	2,5
Folin (2:1)	0,5	0,5	0,5
Didiamkan selama 20 menit pada suhu 37°C			
Diukur dengan spektrometer pada $\lambda=670$ nm			

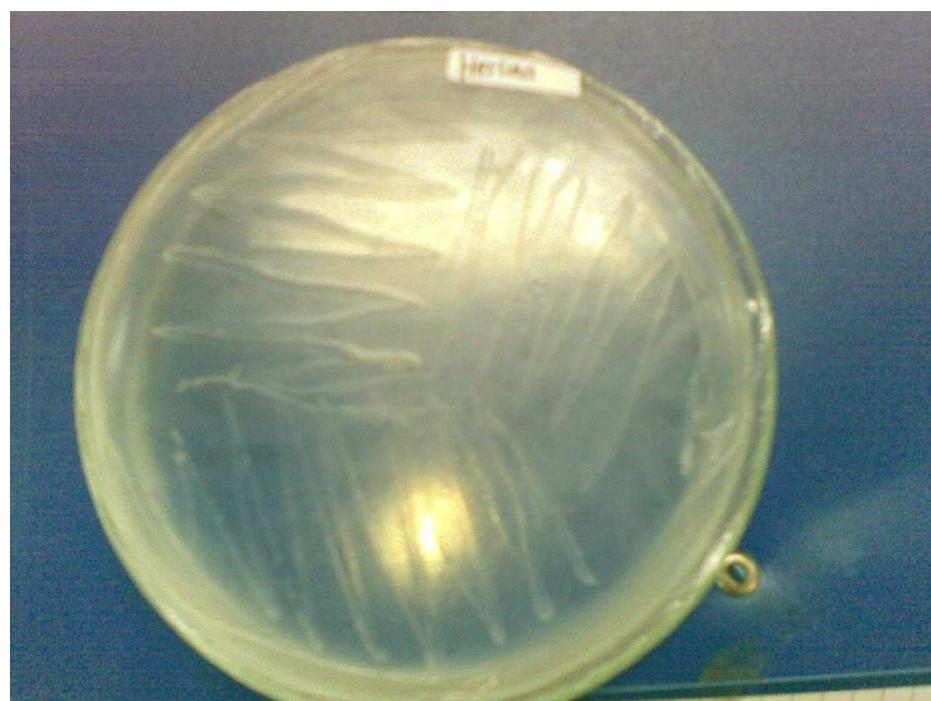
Keterangan:

- Penentuan aktivitas protease berdasarkan senyawa kofaktor digunakan variasi konsentrasi 0,01%; 0,015%; 0,02%; 0,025%; dan 0,03%.

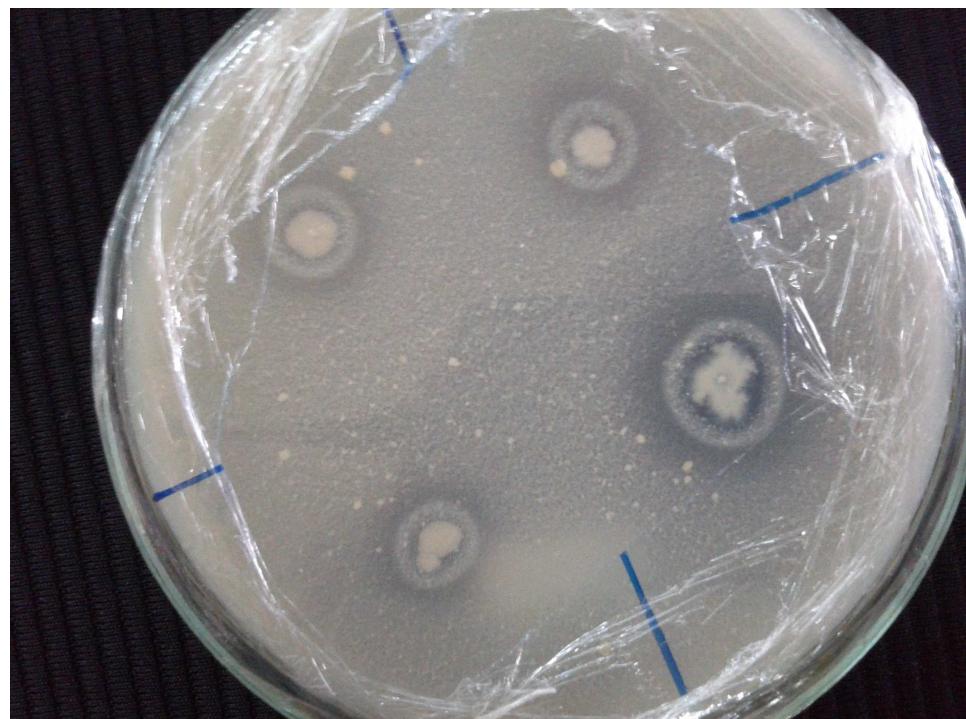
Lampiran 4. Peremajaan Bakteri *Bacillus licheniformis* HSA3-1a dengan penambahan CaCl<sub>2</sub>



Lampiran 5. Peremajaan Bakteri *Bacillus licheniformis* HAS3-1a tanpa penambahan CaCl<sub>2</sub>.



Lampiran 6. Penentuan Zona Hidrolisis *Bacillus licheniformis* HSA3-1a

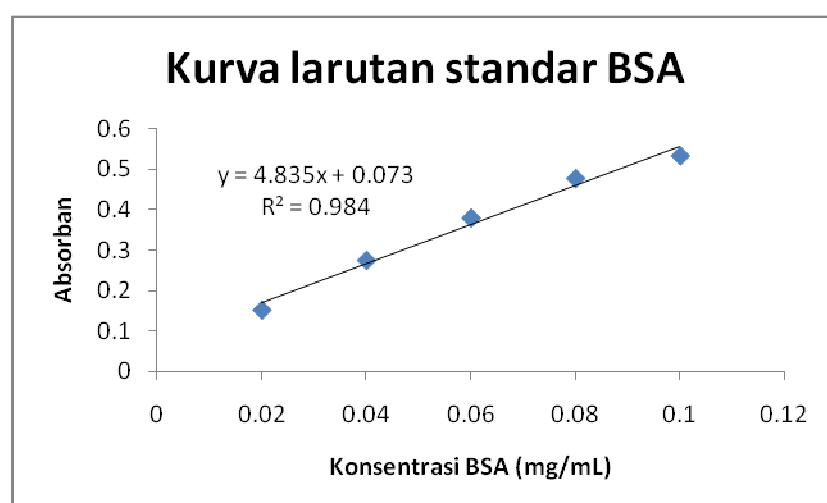


Lampiran 7. Penyiapan Medium Produksi Enzim Protease dari *Bacillus licheniformis* HSA3-1a



Lampiran 8. Kurva standar Bovin Serum Albumin pada  $\lambda$  670 nm

No	[BSA] (mg/mL)	Absorban
1	0.02	0.152
2	0.04	0.275
3	0.06	0.38
4	0.08	0.478
5	0.1	0.534



Lampiran 9. Data pengukuran Kadar Protein dari Enzim Protease

Kode	Absorban (nilai y)	fp	Konsentrasi protein (x)	Kadar protein ( x X fp) mg/mL
12 jam	0,158	50	0,0176	0,8790
24 jam	0,219	50	0,0337	1,5098
36 jam	0,230	50	0,0325	1,6236
<b>48 jam</b>	<b>0,268</b>	<b>50</b>	<b>0,0403</b>	<b>2,0165</b>
60 jam	0,215	50	0,0294	1,4685
72 jam	0,212	50	0,0341	1,4374
84 jam	0,210	50	0,0283	1,4168

Contoh perhitungan penentuan kadar protein:

Data absorban yang diperoleh disubstitusikan ke dalam persamaan standar

$y = 4,835x + 0,073$  dimana y adalah absorbansi protein untuk enzim protease.

Diketahui  $y = 0,268$

$$0,268 = 4,835x + 0,073$$

$$4,835x = 0,268 - 0,073$$

$$4,835x = 0,195$$

$$x = 0,0403 \text{ mg/mL}$$

Lampiran 10. Data Penentuan pH Optimum Enzim Protease dari *B. licheniformis HSA3-1a*

No	pH	Aktivitas Enzim (U/mL)
1	5	0
2	6	0,0099
<b>3</b>	<b>7</b>	<b>0,0104</b>
4	8	0,0064

Lampiran 11. Data pengukuran aktivitas protease dari *B. licheniformis HSA3-1a* terhadap pengaruh suhu

No	Suhu (°C)	Aktivitas Enzim (U/mL)
1	40	0,0083
2	45	0,0092
<b>3</b>	<b>50</b>	<b>0,012</b>
4	55	0,0054
5	60	0,0012

Lampiran 12. Data pengaruh kofaktor ( $\text{CaCl}_2$ ) terhadap aktivitas protease dari *B.licheniformis HSA3-1a*

No	[ $\text{CaCl}_2$ ] (M)	Aktivitas Enzim (U/mL)
1	Kontrol	0,001
2	0,01	0,0001
3	0,015	0,002
4	0,02	0,001
5	0,025	0,001
6	0,03	0,0001