

DAFTAR PUSTAKA

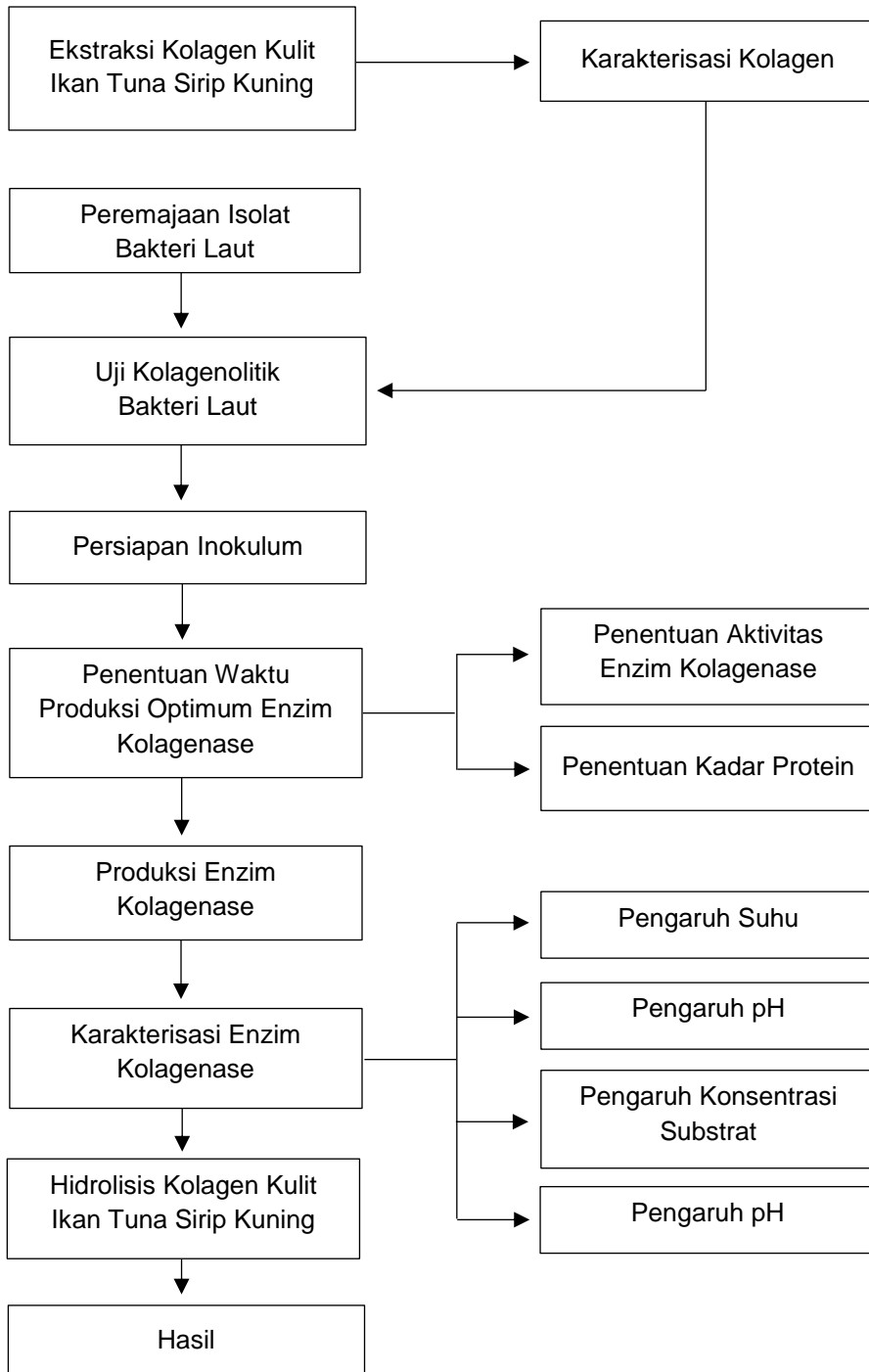
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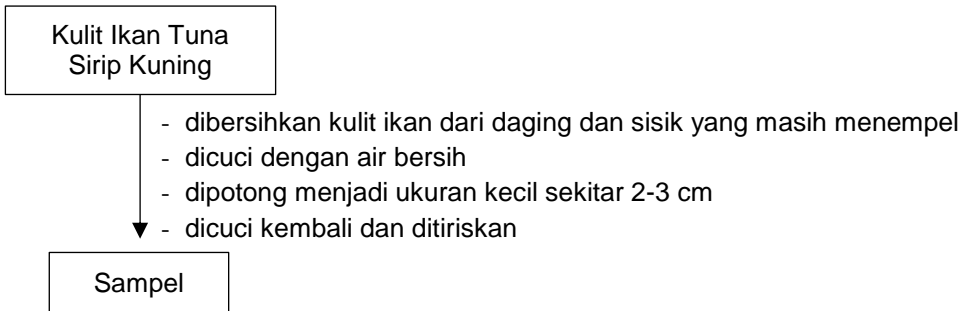
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Lampiran 1. Diagram Alir Penelitian

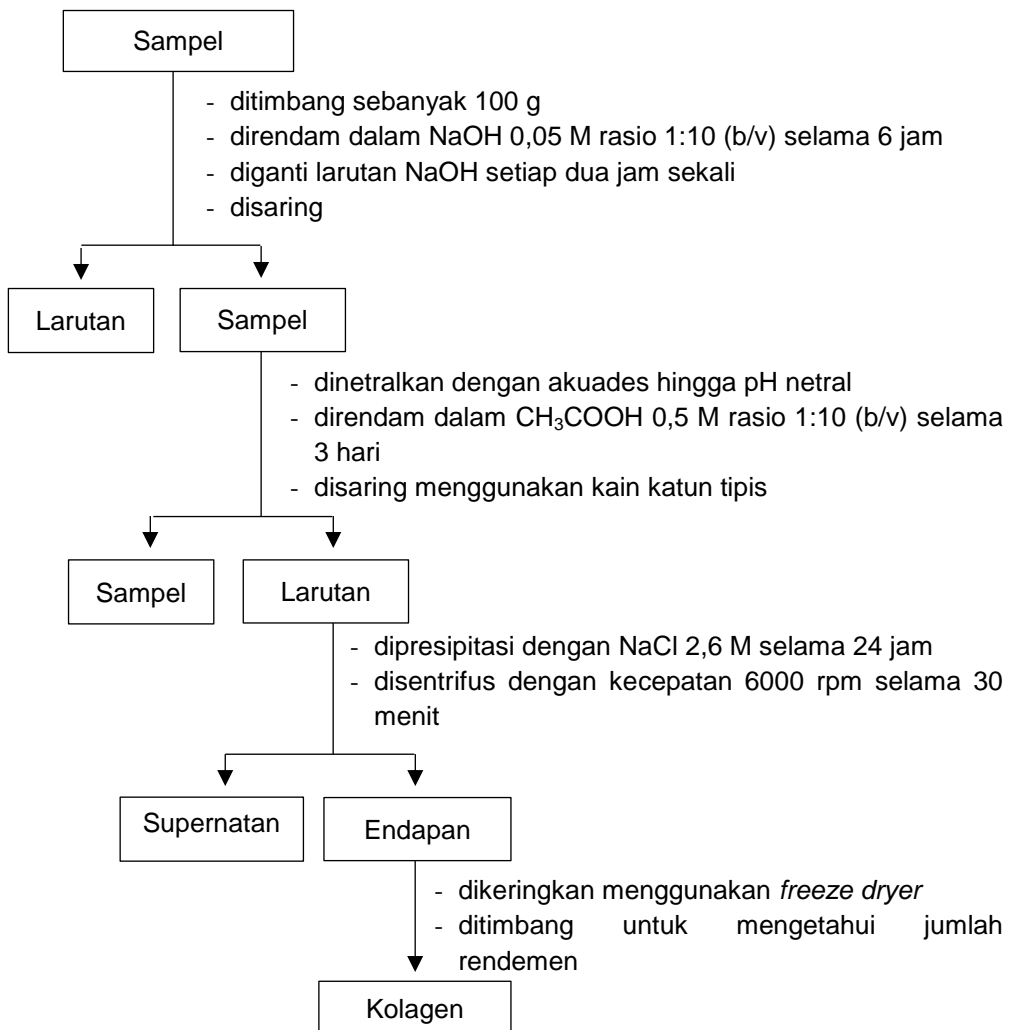
Lampiran 2. Prosedur Penelitian

1. Ekstraksi Kolagen Kulit Ikan Tuna Sirip Kuning

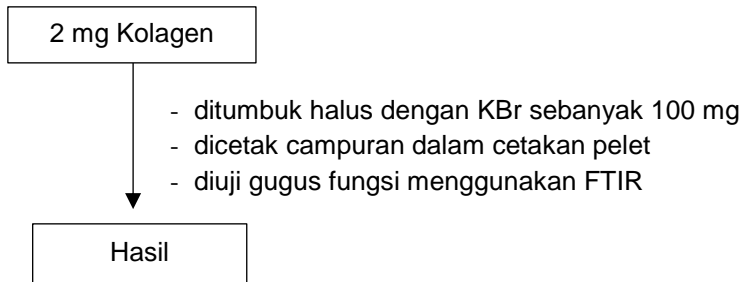
a. Preparasi Sampel



b. Ekstraksi Kolagen

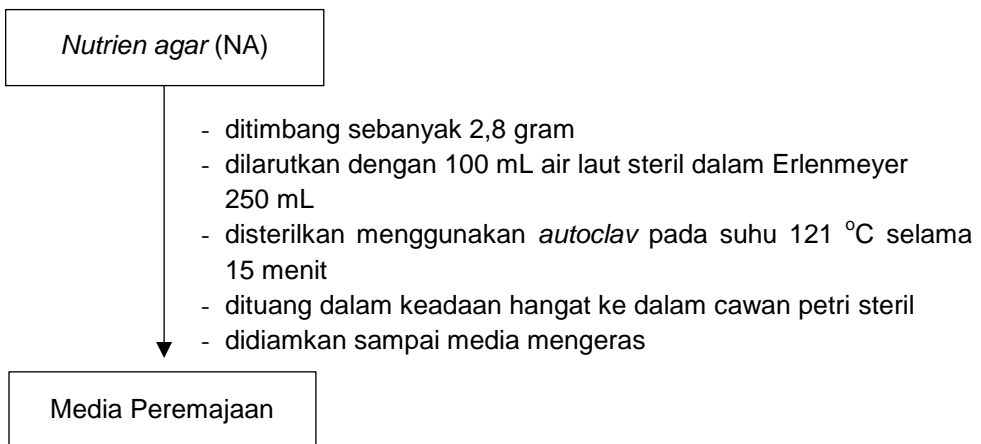


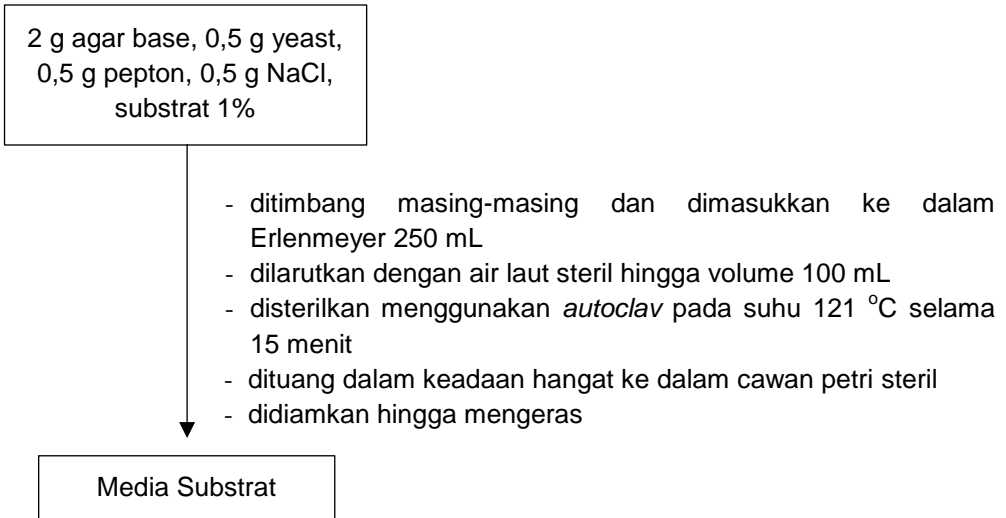
c. Karakterisasi Kolagen



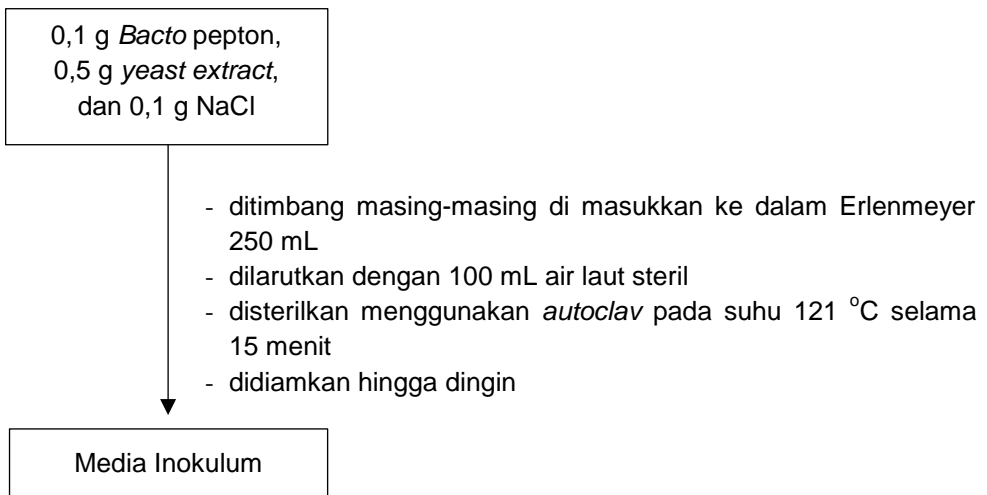
2. Pembuatan Media

a. Media Peremajaan

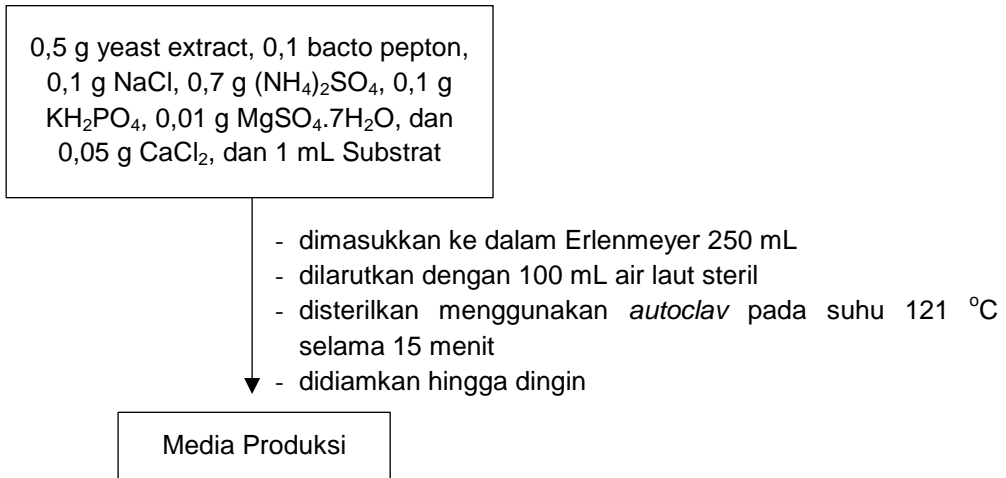


b. Media Substrat

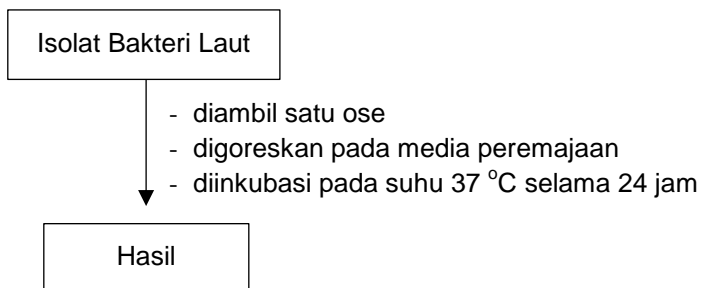
Catatan: prosedur yang sama dilakukan untuk pembuatan media substrat 2% dan 3%

c. Media Inokulum

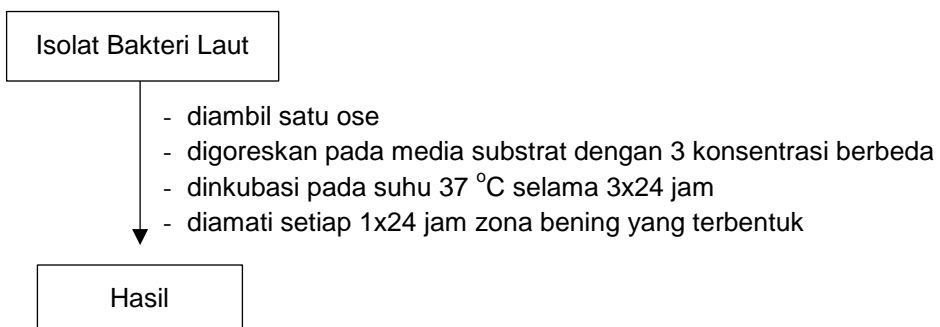
d. Media Produksi



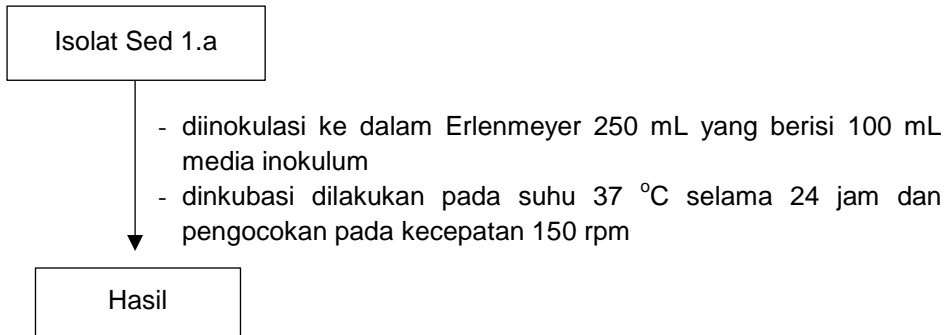
3. Peremajaan Isolat Bakteri Laut



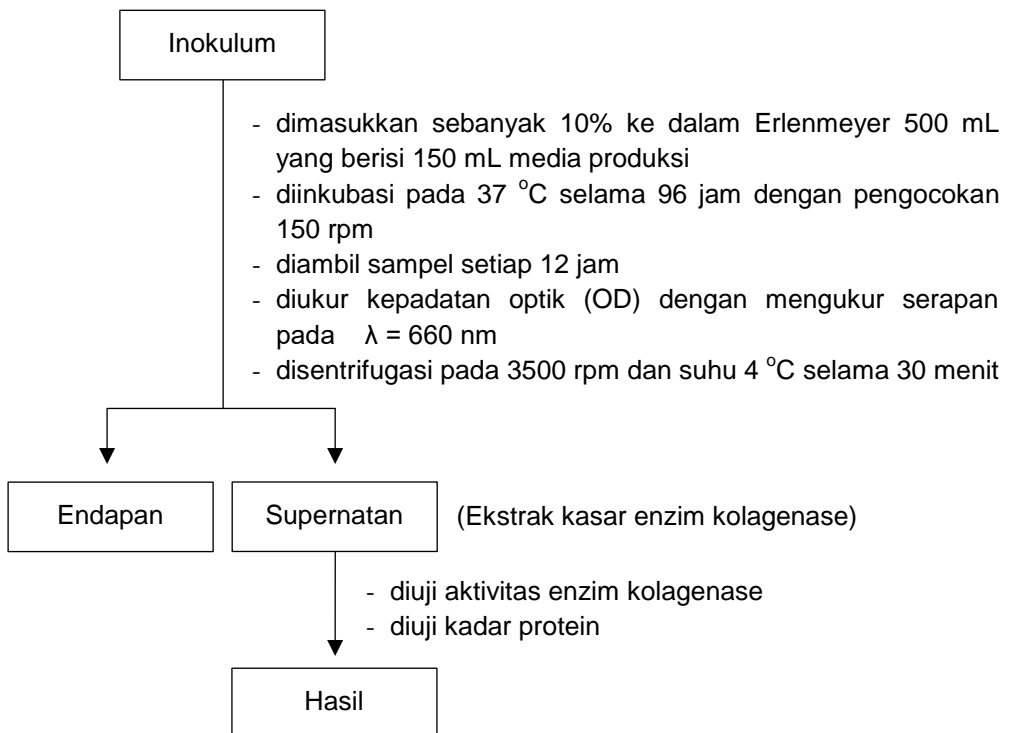
4. Uji Kolagenolitik Bakteri Laut



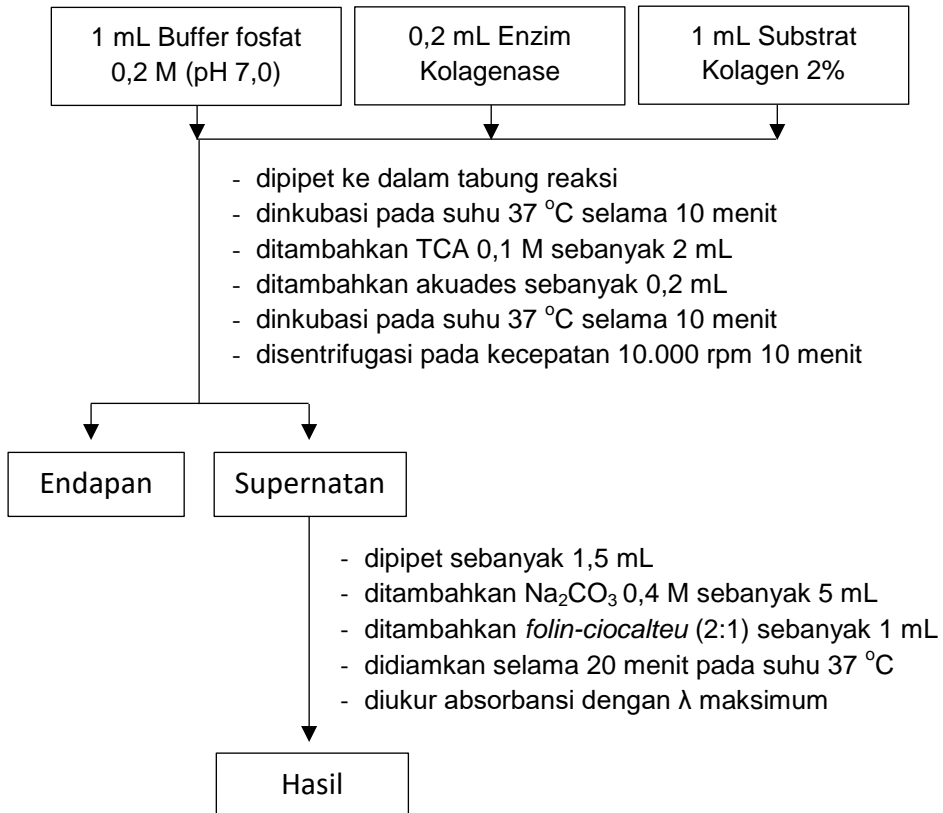
5. Persiapan Inokulum



6. Penentuan Waktu Produksi Optimum Enzim Kolagenase

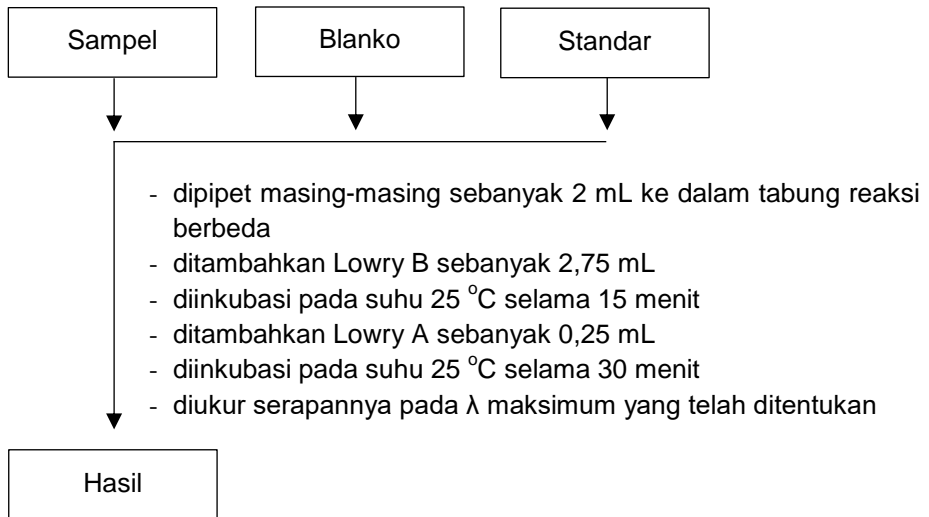


7. Penentuan Aktivitas Kolagenase

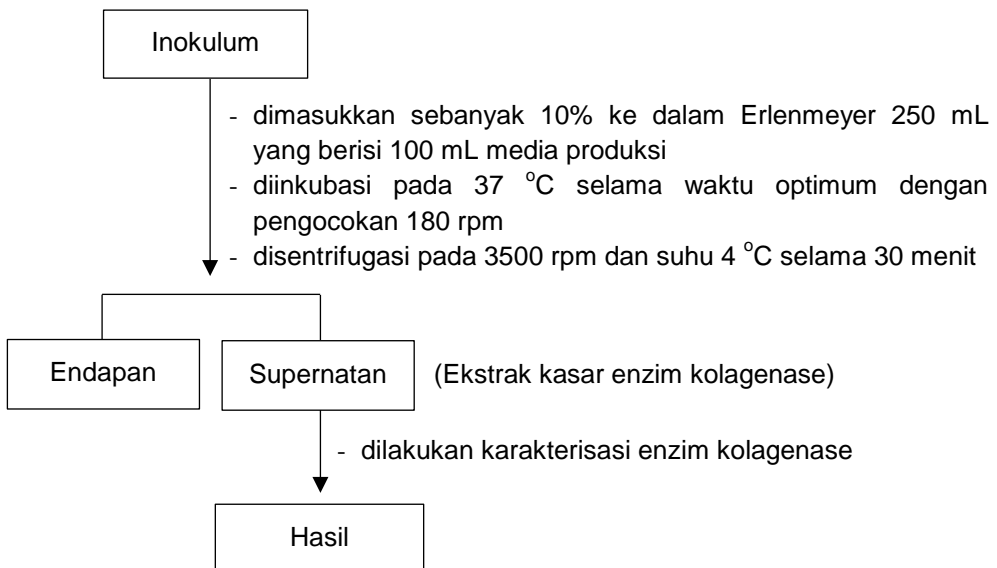


Catatan: Larutan tirosin digunakan sebagai larutan standar enzim kolagenase

8. Penentuan Kadar Protein

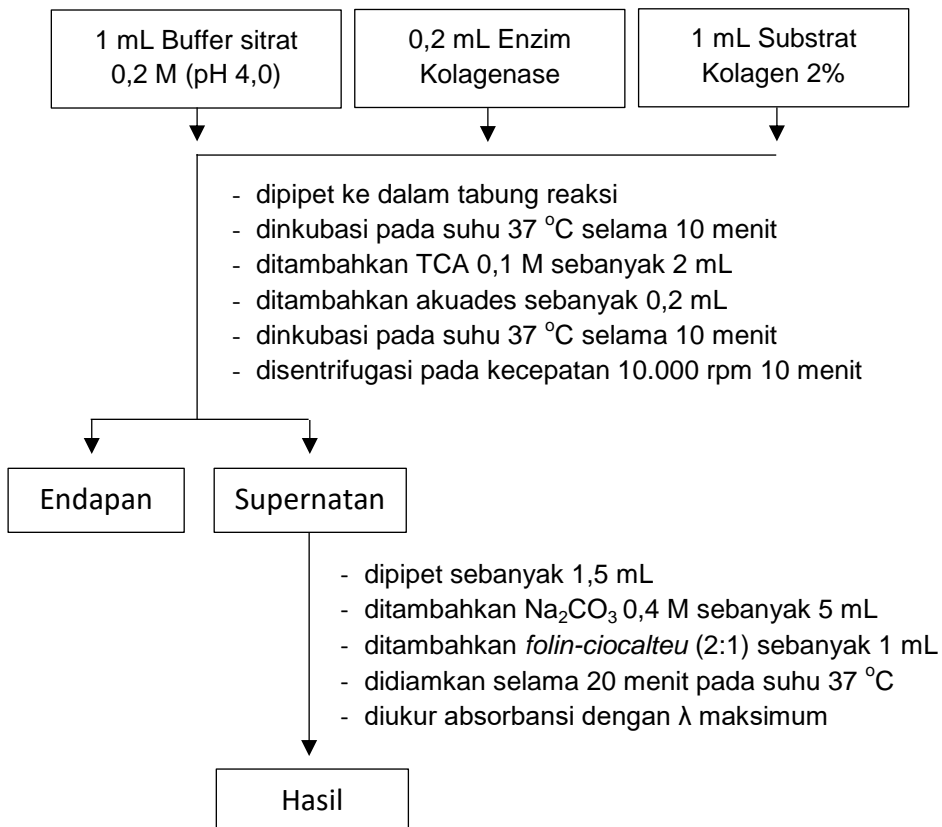


9. Produksi Enzim Kolagenase

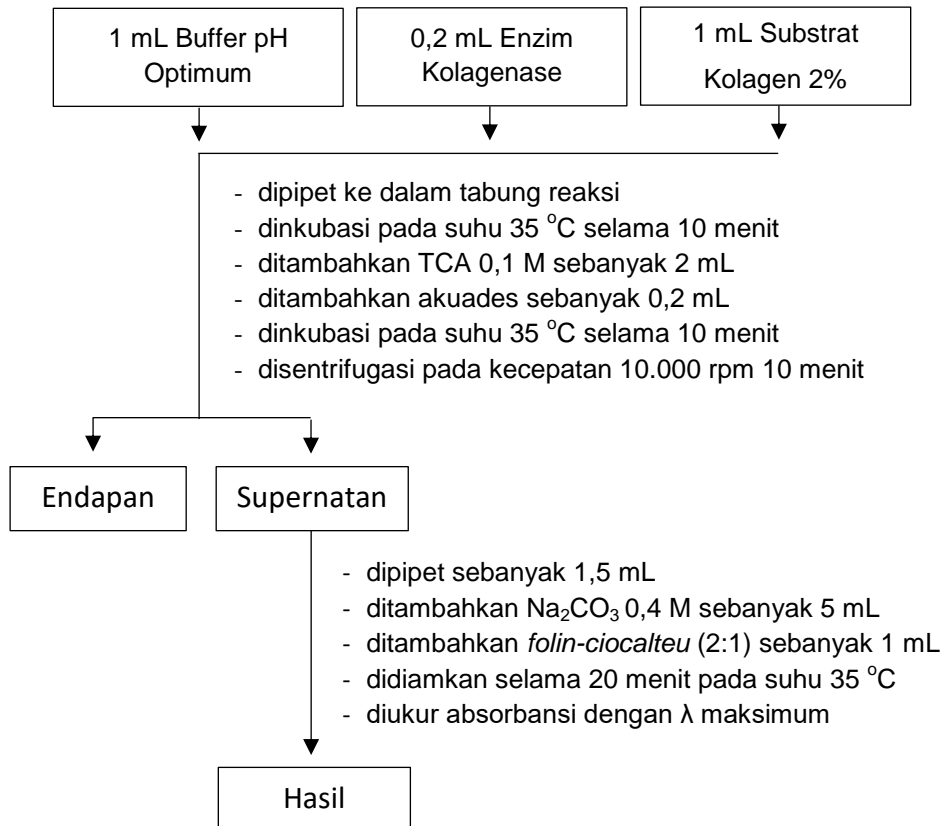


10. Karakterisasi Enzim Kolagenase

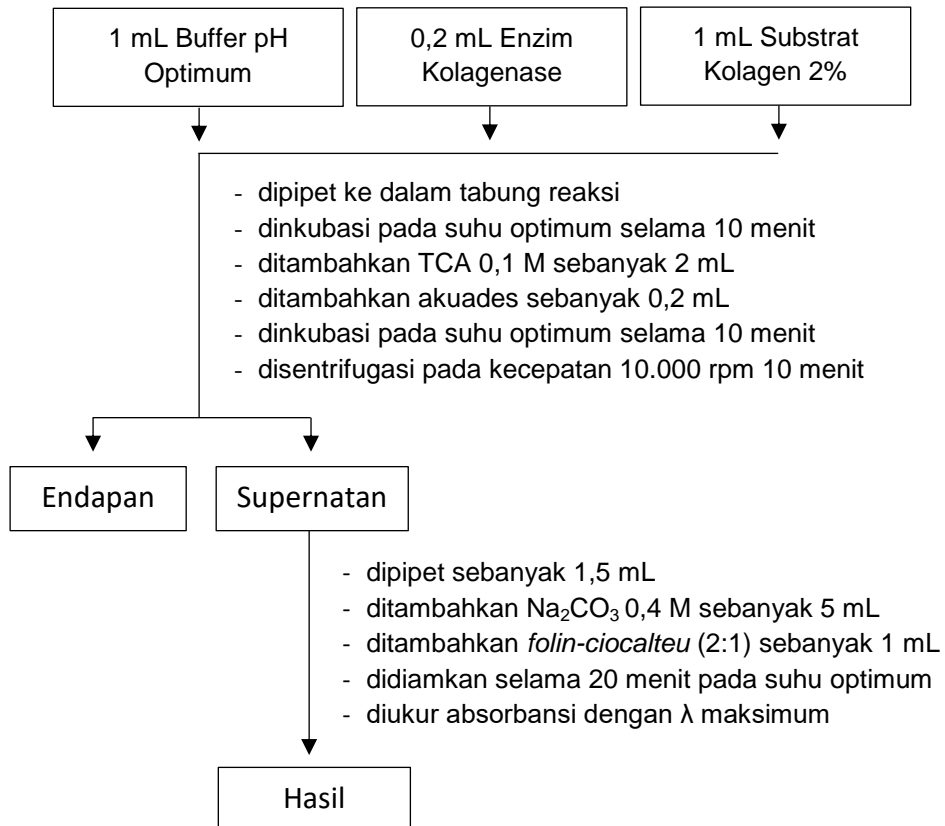
a. Pengaruh pH Terhadap Aktivitas Enzim Kolagenase



Catatan: Prosedur yang sama dilakukan untuk pH berbeda menggunakan larutan buffer sitrat 0,2 M (pH 4,0; 5,0; dan 6,0) dan buffer fosfat 0,2 M (pH 7,0; dan 8,0)

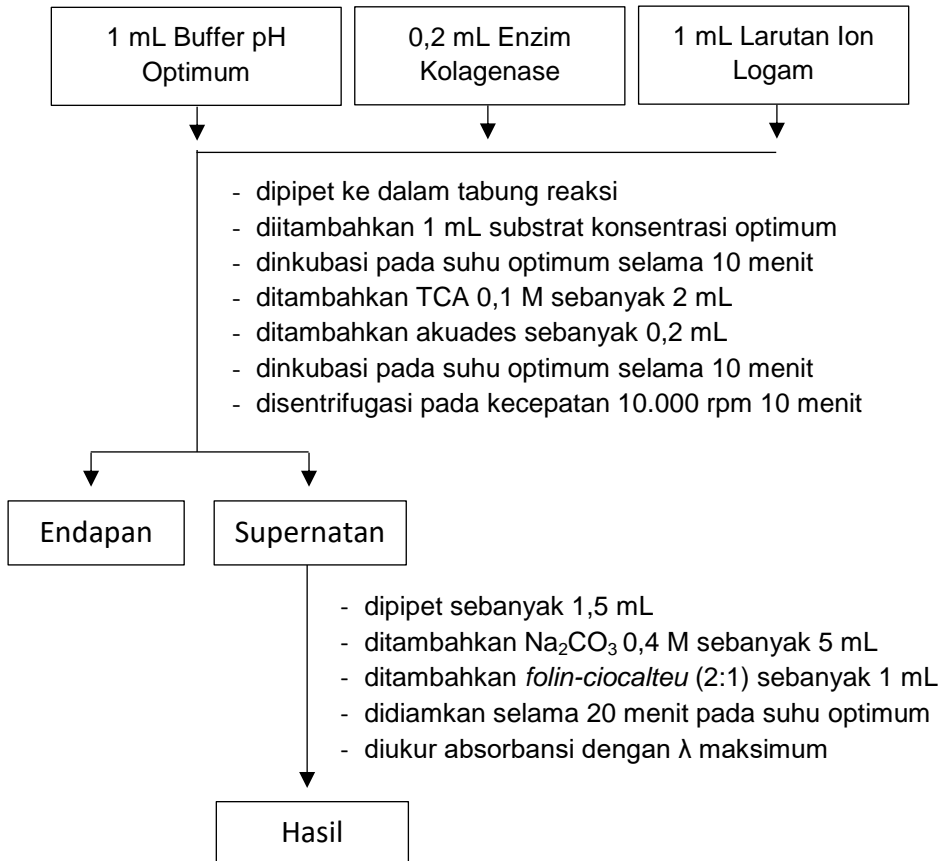
b. Pengaruh Suhu Terhadap Aktivitas Enzim Kolagenase

Catatan: Prosedur yang sama dilakukan pada variasi suhu (40, 45, 50, dan 55 °C)

c. Pengaruh Konsentrasi Substrat Terhadap Aktivitas Enzim Kolagenase

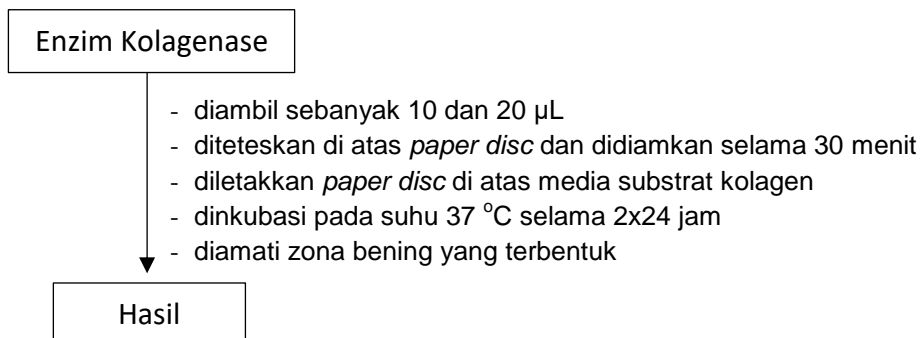
Catatan: Prosedur yang sama dilakukan pada variasi konsentrasi substrat 1%, 1,5%, 2,5% dan 3%.

d. Pengaruh Ion Logam Terhadap Aktivitas Enzim Kolagenase

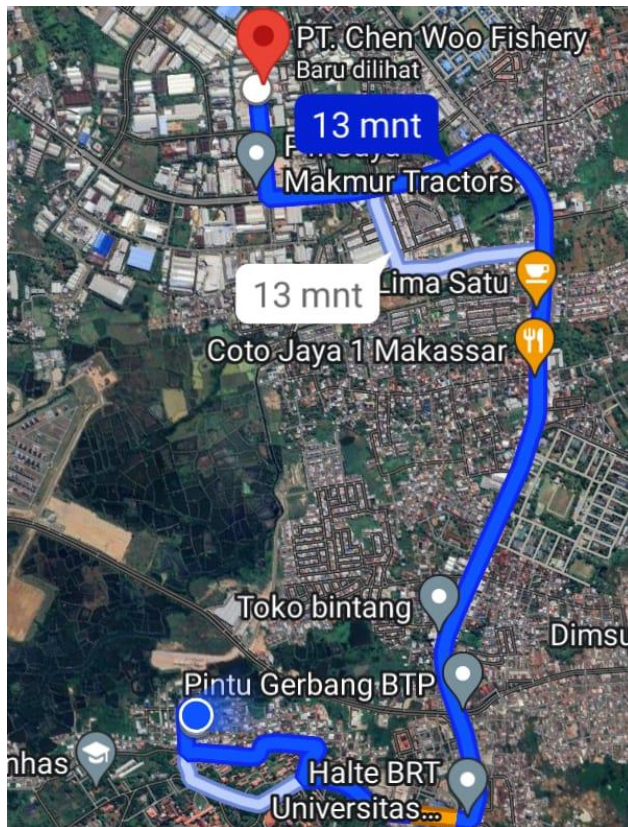


Catatan: Prosedur yang sama dilakukan pada variasi ion logam (K^+ , Na^+ , Ca^{2+} , Mg^{2+} , dan Hg^{2+}) dengan konsentrasi 5 dan 10 mM

11. Hidrolisis Kolagen Kulit Ikan Tuna Sirip Kuning



Lampiran 3. Peta Lokasi Pengambilan Sampel



Lampiran 4. Perhitungan Pembuatan Larutan

1. Pembuatan NaOH 0,05 M sebanyak 1000 mL

$$\begin{aligned} g &= V \times M \times Mr \\ &= 1 \text{ L} \times 0,05 \text{ M} \times 40 \text{ g/mol} \\ &= 2 \text{ g} \end{aligned}$$

2. Pembuatan CH₃COOH 0,5 M sebanyak 1000 mL

$$\begin{aligned} M &= \frac{\% \times bJ \times 1000}{Mr} \\ &= \frac{\% \times 1,05 \text{ g/cm}^3 \times 1000}{60 \text{ g/mol}} \\ &= 17,5 \text{ M} \end{aligned}$$

$$V_1 \times M_1 = V_2 \times M_2$$

$$V_1 \times 17,5 \text{ M} = 1000 \text{ mL} \times 0,5 \text{ M}$$

$$V_1 = \frac{1000 \text{ mL} \times 0,5 \text{ M}}{17,5 \text{ M}}$$

$$V_1 = 28,57 \text{ mL}$$

3. Pembuatan NaCl 2,6 M sebanyak 850 mL

$$\begin{aligned} g &= V \times M \times Mr \\ &= 0,8 \text{ L} \times 2,6 \text{ M} \times 58,5 \text{ g/mol} \\ &= 121,68 \text{ g} \end{aligned}$$

4. Pembuatan TCA 0,1 M sebanyak 200 mL

$$\begin{aligned} g &= V \times M \times Mr \\ &= 0,2 \text{ L} \times 0,1 \text{ M} \times 163,4 \text{ g/mol} \\ &= 3,268 \text{ g} \end{aligned}$$

5. Pembuatan Na₂CO₃ 0,4 M sebanyak 500 mL

$$\begin{aligned} g &= V \times M \times Mr \\ &= 0,5 \text{ L} \times 0,4 \text{ M} \times 106 \text{ g/mol} \\ &= 21,2 \text{ g} \end{aligned}$$

6. Pembuatan Lowry A dan Lowry B

a) Lowry A

5 mL folin ciocalteu : 5 mL akuades (1:1)

b) Lowry B

- **Pembuatan NaOH 0,1 N sebanyak 100 mL**

$$\begin{aligned} g &= V \times N \times Be \\ &= 0,1 \text{ L} \times 0,1 \text{ N} \times 40 \\ &= 0,4 \text{ g} \end{aligned}$$

- **Pembuatan Na₂CO₃ 2% sebanyak 100 mL**

$$\% \text{ b/v} = \frac{\text{g zat terlarut}}{\text{V larutan}} \times 100\%$$

$$2\% = \frac{x}{100 \text{ mL}} \times 100\%$$

$$x = 2 \text{ g}$$

- **Pembuatan 50 mL Na-K-Tatrat 1%**

$$\% \text{ b/v} = \frac{\text{g zat terlarut}}{\text{V larutan}} \times 100\%$$

$$1\% = \frac{x}{50 \text{ mL}} \times 100\%$$

$$x = 0,5 \text{ g}$$

7. Pembuatan Deret Standar**a) Konsentrasi 0,01 mg/mL**

$$\begin{aligned} V_1 \times M_1 &= V_2 \times M_2 \\ V_1 \times 0,1 \text{ mg/mL} &= 2 \text{ mL} \times 0,01 \text{ mg/mL} \\ V_1 &= \frac{2 \text{ mL} \times 0,01 \text{ mg/mL}}{0,1 \text{ mg/mL}} \end{aligned}$$

$$V_1 = 0,2 \text{ mL}$$

$$\text{Volume akuades} = 2 \text{ mL} - 0,2 \text{ mL} = 1,8 \text{ mL}$$

b) Konsentrasi 0,02 mg/mL

$$\begin{aligned} V_1 \times M_1 &= V_2 \times M_2 \\ V_1 \times 1 \text{ mg/mL} &= 2 \text{ mL} \times 0,02 \text{ mg/mL} \\ V_1 &= \frac{2 \text{ mL} \times 0,02 \text{ mg/mL}}{1 \text{ mg/mL}} \end{aligned}$$

$$V_1 = 0,04 \text{ mL}$$

$$\text{Volume akuades} = 2 \text{ mL} - 0,04 \text{ mL} = 1,96 \text{ mL}$$

c) Konsentrasi 0,04 mg/mL

$$\begin{aligned} V_1 \times M_1 &= V_2 \times M_2 \\ V_1 \times 1 \text{ mg/mL} &= 2 \text{ mL} \times 0,04 \text{ mg/mL} \\ V_1 &= \frac{2 \text{ mL} \times 0,04 \text{ mg/mL}}{1 \text{ mg/mL}} \end{aligned}$$

$$V_1 = 0,08 \text{ mL}$$

$$\text{Volume akuades} = 2 \text{ mL} - 0,08 \text{ mL} = 1,92 \text{ mL}$$

d) Konsentrasi 0,08 mg/mL

$$V_1 \times M_1 = V_2 \times M_2$$

$$V_1 \times 1 \text{ mg/mL} = 2 \text{ mL} \times 0,08 \text{ mg/mL}$$

$$V_1 = \frac{2 \text{ mL} \times 0,08 \text{ mg/mL}}{1 \text{ mg/mL}}$$

$$V_1 = 0,16 \text{ mL}$$

$$\text{Volume akuades} = 2 \text{ mL} - 0,16 \text{ mL} = 1,84 \text{ mL}$$

e) Konsentrasi 0,16 mg/mL

$$V_1 \times M_1 = V_2 \times M_2$$

$$V_1 \times 1 \text{ mg/mL} = 2 \text{ mL} \times 0,16 \text{ mg/mL}$$

$$V_1 = \frac{2 \text{ mL} \times 0,16 \text{ mg/mL}}{1 \text{ mg/mL}}$$

$$V_1 = 0,32 \text{ mL}$$

$$\text{Volume akuades} = 2 \text{ mL} - 0,32 \text{ mL} = 1,68 \text{ mL}$$

Lampiran 5. Dokumentasi Penelitian

Pengambilan sampel



Preparasi sampel



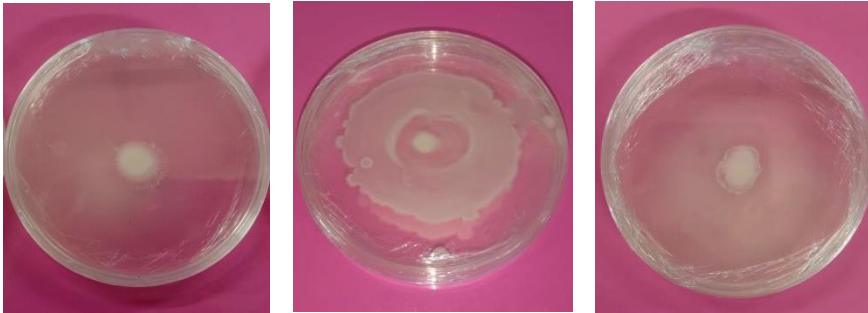
Perendaman dengan NaOH

Perendaman dengan CH₃COOH

Pembuatan media



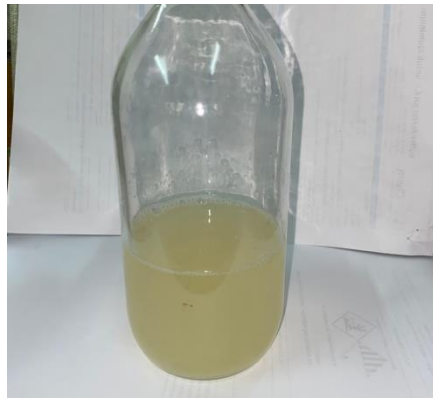
Peremajaan bakteri laut



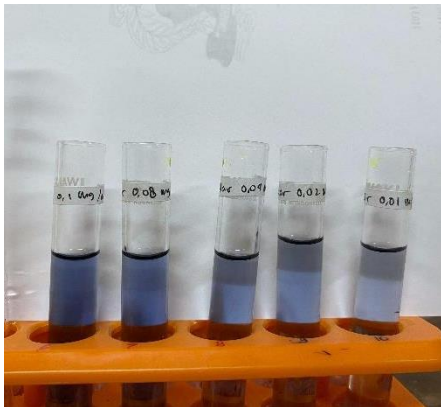
Hasil uji kolagenolitik substrat kolagen ikan tuna sirip kuning 1%, 2%, dan 3%



Produksi enzim di *shaker waterbath*



Ekstrak kasar kolagenase



Penentuan kadar protein



Karakterisasi ekstrak kasar enzim