Optimasi produksi dan karakterisasi lipase bacillus halodurans CM1 = Production optimization and characterization of lipase bacillus halodurans CM1 / Arina Aisyah

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Abstrak

ABSTRAK
Penelitian bertujuan meningkatkan aktivitas lipase dengan mengoptimasi produksi dan mengetahui karakteristik lipase Bacillus halodurans CM1. Bagian pertama penelitian, meningkatkan aktivitas lipase dengan optimasi komposisi media juga mutasi bakteri dengan radiasi gamma dan N-methyl-N rsquo;-nitro-N-nitrosoguanidine NTG . Tujuh media yang berbeda diseleksi untuk mendapatkan media produksi. Delapan variabel komposisi media dioptimasi dengan rancangan Plackett-Burman. Bakteri dimutasi dengan radiasi gamma dosis 0,1 mdash;0,4 kGy dan NTG 0,05 mdash;0,15 mg/mL dengan waktu inkubasi 1 mdash;3 jam. Hasil penelitian menunjukkan bahwa media produksi yang digunakan berdasarkan optimasi media dan komposisi media Plackett-Burman adalah media berdasarkan Bora Bora modifikasi yang mengandung 0,5 palm oil PO dan 0,09 CaCl2. Aktivitas lipase optimal diproduksi oleh bakteri hasil mutasi dengan NTG 0,1 mg/mL yang diinkubasi selama 3 jam. Bagian kedua, enzim dipekatkan dengan metode stirred-cell ultrafiltration UF -ammonium sulfat dan UF-polyethylene glycol PEG . Karakterisasi enzim dilakukan terhadap pengaruh pH, suhu, ion logam, dan deterjen. Rentang pH yang diujikan adalah pH 6 mdash;12, sedangkan variasi suhu 30 mdash;70o C. Ion logam yang diuji, yaitu Mg2 , Ca2 , Zn2 , Mn2 , Fe2 , dan K 1 mM dan 10 mM. Perkiraan berat molekul dilakukan dengan metode SDS-PAGE, kinetika enzim dihitung berdasarkan persamaan Lineweaver-Burk. Hasil penelitian menunjukkan bahwa pemekatan enzim UF-PEG berpengaruh dalam meningkatkan aktivitas enzim lipase sebesar 18,44 . Berat molekul lipase, sekitar 35,7 mdash;37,4 kDa. Aktivitas enzim lipase optimum pada pH 7, suhu 50o C dan relatif stabil pada pH 7 mdash;8, suhu 30 mdash;70o C. Seluruh ion logam yang diujikan mampu meningkatkan aktivitas enzim, namun ion Ca2 menghasilkan aktivitas relatif tertingi di antara ion lainnya. Nilai Km 0,23 mg/mL dan Vmaks 4,07 U/mL. Lipase relatif stabil dengan penambahan deterjen konsentrasi 1 mdash;2 dan mampu menghilangkan minyak sebanyak 8,40 .
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ABSTRACT
The research aimed to improve the activity of lipase by optimizing the production and know the characteristics of lipase Bacillus halodurans CM1. The first part of research aimed to improve the lipase activity by optimization of media composition and mutation of bacteria by gamma radiation and N methyl N 39 nitro N nitrosoguanidine NTG . Seven different media were selected to obtain production medium. Eight compositions of production medium optimized by Plackett Burman design. The bacteria mutated by gamma radiation doses 0.1 mdash 0.4 kGy and NTG 0.05 mdash 0.15 mg mL with incubation time 1 mdash 3 hours. The results showed that the production medium used was based on the Bora and Bora modified medium containing 0.5 palm oil PO and 0.09 CaCl2. High lipase activity produced by the bacterium mutated with NTG 0.1 mg mL were incubated for 3 hours. The second part aimed to examine the characteristics of lipase. The enzyme was concentrated by stirred cell ultrafiltration UF ammonium sulfate and UF polyethylene

glycol PEG methods. Enzyme characterization focused on the effect of pH, temperature, metal ions, and detergents to the lipase activity. Variations in pH tested were pH 6 mdash 12, while the temperature variations 30 mdash 70o C. Metal ions tested were Mg2 , Ca2 , Zn2 , Mn2 , Fe2 , and K 1 mM and 10 mM. Estimated molecular weight was carried out by using SDS PAGE and the enzyme kinetics calculated by Lineweaver Burk equation. The results showed that the concentration of enzymes by PEG UF affected to increase lipase activity 18,44 . The molecular weight of lipase, which was about 35.7 to 37.4 kDa. The optimum condition of lipase reached at pH 7, 50o C and relative stable at pH 7 mdash 8 and temperature 30 mdash 70o C. The whole metal ions tested were able to increase the activity of the enzyme. Ca2 showed the highest relative activity among others. Km value was 0.23 mg mL and Vmax 4.07 U mL. Lipase was relatively stable with the addition of 1 mdash 2 detergent concentration and was able to remove 8.40 the oil.