

Penapisan dan karakterisasi actinobacteria termofilik potensial yang memiliki aktivitas amilolitik = Screening and characterization of potential thermophilic actinobacteria for amyloytic activity

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Abstrak

Tujuan penelitian adalah mengetahui aktivitas amilolitik 17 isolat 'Actinobacteria' termofilik pada suhu tinggi, dan memperoleh informasi spesies, dan posisi filogenetik isolat potensial berdasarkan data sekuen gen 16S rRNA, analisis filogenetik, karakterisasi morfologi, fisiologi, dan biokimia. Kemampuan tumbuh 17 isolat 'Actinobacteria' termofilik pada berbagai variasi suhu diuji menggunakan medium ISP 1 agar, dan diinkubasi pada suhu 45, 50, 55, 60 $^{\circ}\text{C}$ selama 7 hari. Berdasarkan hasil penelitian, 17 isolat memiliki pertumbuhan yang bervariasi pada suhu 45–60 $^{\circ}\text{C}$. Tujuh belas isolat tumbuh pada suhu inkubasi 45 $^{\circ}\text{C}$, 16 isolat pada suhu 50 $^{\circ}\text{C}$, enam isolat pada suhu 55 $^{\circ}\text{C}$, dan lima isolat pada suhu 60 $^{\circ}\text{C}$ terdiri atas SL1-2-R-2, SL1-2-R-3, SL1-2-R-4, SL2-2-R-15, dan SL3-1-R-16. Aktivitas amilolitik 17 isolat 'Actinobacteria' termofilik pada berbagai variasi suhu diuji dengan metode 'starch agar plate', menggunakan medium Minimal (Mm) agar dengan penambahan pati ('soluble starch') sebagai substrat sebanyak 1% (b/v), dan diinkubasi pada suhu 45, 50, 55, dan 60 $^{\circ}\text{C}$ selama 7 hari. Aktivitas amilolitik yang positif ditandai dengan terbentuknya zona bening di sekitar koloni bakteri setelah diteteskan larutan 'Lugol's iodine' sebanyak 1,5 ml. Hasil penelitian menunjukkan bahwa sebagian besar isolat yang diperoleh dari tanah di dekat geiser Cisolok memiliki aktivitas amilolitik yang bervariasi pada suhu 45–60 $^{\circ}\text{C}$. Lima belas isolat memiliki aktivitas amilolitik pada suhu 45 $^{\circ}\text{C}$, 13 isolat pada suhu 50 $^{\circ}\text{C}$, empat isolat pada suhu 55 $^{\circ}\text{C}$, dan hanya tiga isolat pada suhu 60 $^{\circ}\text{C}$. Namun demikian, dua isolat (SL2-2-R-15 dan SL3-1-R-16) tidak memiliki aktivitas amilolitik pada suhu 45, 50, dan 55 $^{\circ}\text{C}$ setelah diinkubasi selama 7 hari. Tiga isolat potensial yang memiliki aktivitas amilolitik pada suhu 60 $^{\circ}\text{C}$ (SL1-2-R-2, SL1-2-R-3, dan SL1-2-R-4), berdasarkan data sekuen gen 16S rRNA, analisis filogenetik, dan karakterisasi fenotipik tiga isolat potensial tersebut diidentifikasi sebagai 'Actinomadura keratinilytica'.

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The aims of this study were to screen for amyloytic activity of the 17 themophilic 'Actinobacteria' at high temperature, and to obtain species information and phylogenetic position based on 16S rRNA gene sequence, phylogenetic analysis, morphological, physiological, and biochemical characterizations. The ability to grow at various temperature was carried out on ISP 1 agar medium, incubated at 45, 50, 55, 60 $^{\circ}\text{C}$ for 7 days. The results showed that the 17 isolates 'Actinobacteria' have varying growth at a temperature of 45–60 $^{\circ}\text{C}$. Seventeen, 16, and six isolates grew at 45, 50, and 55 $^{\circ}\text{C}$, respectively, and only five isolates grew at 60 $^{\circ}\text{C}$, designated SL1-2-R-2, SL1-2-R-3, SL1-2-R-4, SL2-2-R-15, and SL3-1-R-16. Amyloytic activity of the 17 themophilic 'Actinobacteria' at various temperature was carried out using the starch agar plate method on Minimal (Mm) agar medium with the addition of 1% (w/v) soluble starch as substrate, and incubated at 45, 50, 55, and 60 $^{\circ}\text{C}$ for up to 7 days. Amyloytic activity was detected by flooding the plates with 1.5 ml of

Lugol's iodine solution. Clear zones around the colonies indicated positive results for amylolytic activity. The results showed that most of the isolates obtained from the soil near the Cisolok geyser have varying amylolytic activity at a temperature of 45–60° C. In this study, 15, 13, four, and three out of 17 isolates were positive for amylolytic activity at 45, 50, 55, and 60 °C, respectively. Meanwhile, two isolates, designated SL2-2-R-15 and SL3-1-R-16, showed negative results for amylolytic activity at 45, 50, and 55 °C, even after 7 days of incubation. Three potential isolates which have amylolytic activity at 60 °C (designated SL1-2-R-2, SL1-2-R-3, and SL1-2-R-4), based on 16S rRNA gene sequence, phylogenetic analysis, and phenotypic characterizations were identified as '*Actinomadura keratinilytica*'.