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Isolation, biochemical characterization, and identification of thermostable alpha amylase producing bacteria from hot water springs in Sri Lanka.

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The bacteria isolated from hot water springs are good sources of thermostable α -amylases, which can be used in industries in Sri Lanka. In this study we have isolated and characterized biochemical properties of thermostable α -amylase producing bacteria from two hot water springs in Sri Lanka. Water samples were obtained from Maha Oya (Ampara District) and Nelumwewa (Polonnaruwa District) hot water springs. Soil-water mixtures were also collected from Nelumwewa. The highest temperature of 57°C was recorded at Maha Oya. Selected samples were inoculated into starch nutrient broth containing 1% (w/v) soluble starch as the carbon source (pH was 6.9). The enrichment of thermostable α -amylase producers was done by incubating the cultures for 72 hours at 50°C. The enriched samples were inoculated into starch nutrient agar plates by the streak plate and spread plate methods. The isolated strains were screened for α -amylase production using the starch hydrolysis test. Altogether ten different strains of bacteria were isolated. Four strains (S₁₂, S₁₈, S₂₁, and S₅₁) were selected based on starch hydrolysis test on starch nutrient agar plates. The strains were further characterized by morphological and biochemical tests. The organism S₅₁ was the most efficient α -amylase producer among the above 4 strains, which was confirmed by starch utilization analysis in starch nutrient broth by the dinitrosalicylic acid (DNS) method. The strain S₅₁ was isolated from the water sample collected from a well at Maha Oya hot water springs which recorded the highest temperature of 57°C. The selected species produced 1.85 mg/mL of reducing sugars (as maltose) in 12 hours. 16S rRNA gene sequencing revealed the organism S₅₁ was 99% similar to the organism *Caldimonas manganoxidans* (GenBank Accession No. NR 113844.1). The bacteria was named as *Caldimonas manganoxidans* MS1, which is a good alpha amylase producer, therefore the research was continued for the purification and functional characterization of the thermostable alpha amylase from *Caldimonas manganoxidans* MS1.

Key words: α -Amylase, *Caldimonas manganoxidans* MS1, Thermostable

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