

## Identification of a Best Thermostable Alkaline Protease Producing Bacterial Strain

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The objective of this study was to characterize one of the selected bacterial strains, among the best five locally isolated alkaline protease producers (strains DS<sub>1</sub>, DS<sub>2</sub>, DS<sub>3</sub>, DS<sub>4</sub> and DS<sub>5</sub>) from dog decaying soil. These five strains produced the alkaline proteases (from 90 to 1760 UmL<sup>-1</sup>) which were active at pH 9.5 and 70°C. The strain DS<sub>2</sub> produced highest alkaline protease activity (1760 UmL<sup>-1</sup>) and considered for identification. Strain DS<sub>2</sub> cells are rod shaped, gram positive, motile, facultative anaerobic, catalase positive and formed round or oval spores in swollen sporangia. No soluble pigment was produced by the strain DS<sub>2</sub> on nutrient agar. It had the ability to produce oxidase, urease and indole. It gave negative results to Vogues Proscateur test. Hydrogen sulfide is not produced by strain DS<sub>2</sub>. It did not reduce nitrite and hydrolyse tyrosine. It had the ability to hydrolyse starch and casein. Strain DS<sub>2</sub> grown at 40, 45 and 50°C, with 5, 7, 10 % of NaCl and showed tolerance to the pH values of 7, 8, 9, 10 and 11. Based on the above characters the strain DS<sub>2</sub> expected to belong to genus *Paenibacillus*. The colonies of strain DS<sub>2</sub> had tip-splitting, irregular margin, 1.4(±0.3) mm in diameter, pale in color, low convex elevation, moist and shiny surface, at 24h of growth. It possessed the ability to produce acid from glycerol, D-ribose, D-adonitol, D-galactose, D-glucose, D-mannose, methyl- $\alpha$ -D-mannopyranoside, methyl- $\alpha$ -D-glucopyranoside, N-acetylglucosamine, esculin, D-cellobiose, D-maltose, amygdalin, arbutin, salicin, D-lactose, D-melibiose, D- sucrose, D-trehalose, D-melezitose, D-raffinose, amidon, glycogen, gentiobiose, D-turanose, D-fucose, L-fucose and potassium gluconate and did not produce acid from erythritol, L-arabinose, D-arabinose, D-xylose, L-xylose, D-fructose, L-rhamnose, methyl- $\beta$ -D-xylopyranoside, L-sorbose, dulcitol, inositol, D-mannitol, D-sorbitol, inulin, xylitol, D-tagatose, D-lxyose, D-arabitol, L-arabitol, potassium-2-ketogluconate and potassium 5-ketogluconate. Based on the morphological characters, the strain DS<sub>2</sub> expected to be belonging to the genus *Paenibacillus* or *Clostridium*. Based on the biochemical test the strain DS<sub>2</sub> was expected to belong to *Paenibacillus dendritiformis*. By the 16S rDNA sequencing, the strain DS<sub>2</sub> was confirmed to be belong to the Kingdom: Procaryotae; Division: Bacteria; Order: Bacillales; Family: Paenibacillaceae; Genus: *Paenibacillus*; Species: *dendritiformis*.

**Key words:** Protease, biochemical tests, gene sequence, morphology, strain, genus.