

ABSTRACT

Lutfiah Munawaroh, 24020221140063. Isolation, Screening, and Identification of Amylolytic Bacteria from Soil Around Breadfruit Trees (Artocarpus altilis) at the KHDTK Wanadipa, Diponegoro University, Semarang. Under the guidance of Nurhayati and Susiana Purwantisari.

The global demand for amylase continues to increase along with the development of various industrial sectors that utilize amylase, particularly in food, feed, detergents, textiles, paper, and bioenergy. Soil bacteria in the roots of breadfruit trees at the KHDTK Wanadipa, Diponegoro University, have the potential to produce amylase because the soil in the roots of breadfruit trees is rich in starch, a substrate source for amylolytic bacteria. The study aimed to isolate amylolytic bacteria from the Special Purpose Forest Area (KHDTK) Wanadipa, Diponegoro University, and to molecularly analyze the screening results of the best amylolytic isolates. The method used included bacterial isolation using a multi-dilution method with Nutrient Broth (NB) media. Characterization was carried out macroscopically, microscopically, and by catalase test. Amylolytic screening was carried out using Starch Agar media and iodine solution. Measurement of amylase activity was carried out using a spectrophotometer method by measuring the glucose standard curve, growth curve, and amylase activity with DNS solution. Isolation of amylolytic bacterial DNA was carried out using the InstaGene Matrix kit. Molecular identification of the best amylolytic isolates was carried out through 16S rRNA gene analysis with primers 27F and 1492R. Sequence analysis was carried out using BLAST and MEGA to construct a phylogenetic tree. The results showed that there were 7 bacterial isolates from the screening of amylolytic bacteria. Isolate A2 was the best amylase producer with a high amylolytic index value of 3.35, indicating a very strong starch hydrolysis ability, so it can be categorized as a potential amylolytic isolate. Quantitative test of the amylase activity of isolate A2 using the DNS method showed the highest activity at 24 hours with an activity of 0.2057 U/mL, indicating the optimum point for enzyme production. Molecular identification showed that isolate A2 was highly similar to Bacillus albus.

Keywords: Amylase, Bacillus albus, amylolytic bacteria, KHDTK, 16S rRNA