Isolation and Characterization of Polyhydroxyalkanoate Producing Bacteria from Putrefied Rice

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Polyhydroxyalkanoates (PHAs) are internal microbial polyesters that are produced when there is an abundance of carbon sources and a scarcity of nutrients like nitrogen to function as energy storage granules and give prokaryotes stress tolerance. These biopolymers are viable substitutes for carbon-based synthetic polymers and can be obtained from bacteria growing on putrefied rice. However, their commercialization is constrained by high production costs. The objective of this work was thus, to isolate and characterize PHA and PHA-producing bacteria using physical and biochemical processes in order to synthesize PHA-type biopolymer through a laboratory-scale procedure. A putrefied rice sample was used to collect the bacteria for this study, and it was then screened for PHA production. Microscopic staining was done to confirm the presence of PHA accumulation in the bacteria. When provided with a complex growth medium, bacteria growing in the medium exhibited diauxic growth. Molecular identification based-on 16srRNA marker gene and Sanger DNA sequencing confirmed the identity of the isolated PHA producing bacterium as *Bacillus tropicus* strain MCCC 1A01406. PHA produced from a cultured bacterial sample was characterized using Raman spectroscopy to confirm the chemical structure and functional groups present. Results from Raman analysis confirmed that the PHA produced was a copolymer between 3-hydroxybutyrate (3HB) and 3-hydroxyvalerate (3HV) units. The diauxic growth together with the production of a copolymer confirmed that the bacteria used in our study were feeding on more than one carbon substrate. This study clearly demonstrated that free-living environmental organisms can be isolated and conveniently used for the purpose of polyhydroxybutyrate (PHB) bioplastics production.

Keywords: Bioplastics, PHA, PHB, Polyhydroxyalkanoate, Raman Spectroscopy