Bacterial production and extracellular degradation of polyesters

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Abstract

Poly(β-hydroxyalkanoates), PHAs, are thermoplastic polyesters, which have potential for use as biodegradable plastics. The material properties of PHAs depend upon their chemical structure. *Pseudomonas oleovorans* accumulates medium chain length (MCL) PHAs whose structure depends upon the carbon source used for PHA synthesis. In this study, *P. oleovorans* was used for the production of functionalized PHAs, using a range of carbon sources, including sodium octanoate, nonanoic acid, ω-bromoalkanoic acids, oleic acid, and 11-undecylenic acid, as well as alternative feed stocks such as tall oil, cuphea oil and vernonia oil. The PHAs produced from these substrates were characterized using gas chromatography, nuclear magnetic resonance, differential scanning calorimetry, and gel permeation chromatography. ^ The biodegradability of a wide range of PHAs, including both short chain length (SCL) and MCL PHAs, as well as that of a crosslinked PHA and several synthetic polyesters, was assessed using the bacterium *Comamonas* Strain P37C. This organism was isolated from residential compost for its ability to degrade MCL PHAs. A number of growth and enzyme assays were used to analyze the biodegradability of the aliphatic polyesters used in this study. These assays included overlay plates, clear zone tubes, respirometry, and colorimetric assays. With the exception of the crosslinked PHA, strain P37C was found to be capable of degrading all of the PHAs to which it was exposed. Enzyme assays using cell-free enzyme preparations indicated that strain P37C produces at least two enzymes for the hydrolysis of SCL and MCL PHA. ^ The SCL and MCL PHA depolymerases were concentrated and partially purified using ammonium sulfate fractionation, hydrophobic interaction chromatography, PHA affinity assays, gel electrophoresis, and activity gels. Ammonium sulfate fractionation led to a three-fold increase in specific activity of the MCL PHA depolymerase. The activity of this enzyme preparation was found to become increasingly unstable with degree of purification. Activity gels were prepared by overlaying isoelectric focusing gels with turbid PHA-containing polyacrylamide gels. The SCL PHA depolymerase activity was confined to two distinct bands which produced clear zones in a PHB overlay gel, suggesting that strain P37C produces two SCL PHA depolymerases. ^

Subject Area

Microbiology|Biochemistry|Polymer chemistry|Environmental science

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