



Regular article

Characterization of polyhydroxyalkanoates extracted from wastewater sludge under different environmental conditions



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ARTICLE INFO

Article history:

Received 2 April 2015

Received in revised form

14 December 2015

Accepted 30 December 2015

Available online 6 January 2016

Keywords:

Polyhydroxyalkanoates

Wastewater sludge

Aerobic processes

Anaerobic processes

Bioconversion

Batch processing

ABSTRACT

Polyhydroxyalkanoates (PHAs) are bioplastics that naturally accumulate in the microbial cells while performing organic substrate metabolism. PHAs bioconversion in microbial cells is affected by both growth environments (aerobic and anaerobic) and feeding systems (carbon and nutrient limitations). Sequential batch reactors (SBRs) were used in this research for producing PHAs; studies showed on an average 42% with a maximum of 63% PHAs yield under anaerobic–oxic conditions quantified by gas chromatography. Produced PHAs from phase 4 were thermally and physically characterized. Fourier transform infrared spectroscopy showed strong presence of carbonyl peaks at 1720 cm^{-1} in all PHAs. Gel permeation chromatography reported polydispersity index values in range of 2.4–3.6 showing non-uniformity of molecular weights in the slice of PHAs and differential scanning calorimetry reported melting point temperatures of $146\text{--}154\text{ }^{\circ}\text{C}$, confirming usefulness of produced PHAs in industrial applications. X-Ray Diffraction confirmed crystal structure in all PHAs with the most crystalline from SBR3. Thermogravimetric analysis further confirmed highest thermal degradation temperature of $283\text{ }^{\circ}\text{C}$ for PHAs from SBR3. Different blends of wastewater fed to mixed sodium acetate acclimatized biomass further showed the importance of substrate carbon source for PHAs production in various growth environments.

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1. Introduction

Strategically water polluting compounds can be turned into renewable value-added bioproducts. Many bacteria in their cell body have the capacity to produce these value-added bioproducts such as polyhydroxyalkanoates (PHAs), lipid oils and proteins, while consuming the pollutants from the wastewater [1–4].

PHAs are a group of natural bioplastics that accumulate inside the cell structure of microorganisms. It is a natural alternate to synthetic fossil fuel plastic that is made from processing of fossil fuel hydrocarbons. PHAs are easily biodegradable and have same synthetic plastic like physical and thermal properties [5,6].

PHAs types are defined by carbon chain length of repeated monomer (functional group, R) attached to 3-hydroxyalkanoate molecule, as short ($\text{C}_3\text{--}\text{C}_5$), medium ($\text{C}_6\text{--}\text{C}_{14}$) and long ($\text{C}_{15}\text{--more}$)

chain length bioplastics [7]. The functional group (R) attached to 3-hydroxyalkanoate molecule may be of Hydroxyalkanoate (HA), Hydroxybutyrate (HB), or Hydroxyvalerate (HV) unit.

Most of the studies performed for production of PHAs have been done with pure cultures of microorganisms. Some of the microorganisms being researched for efficient production are *Escherichia coli*, *Cupriavidus nector*, *Ralstonia eutropha* [8–10].

Use of microbial communities present within the ecosystem and utilizing in situ carbon and nutrients present in the ecosystem not only enrich microorganisms to effectively produce PHAs but can also reduce production cost [11,12]. This can also bring reduction in equipment cost as no sterile condition will be required for microorganism's growth. Feed carbon source along with available growth nutrients, system temperature, pH, and mixing are some of the controlling factors that can also improve the PHAs accumulation rate; product type and characteristics in the microbial cell.

Under this strategic vision, purpose of this study was to show the effectiveness of ecosystem engineering by synthesizing polyhydroxyalkanoates under aerobic and anaerobic–oxic dynamic

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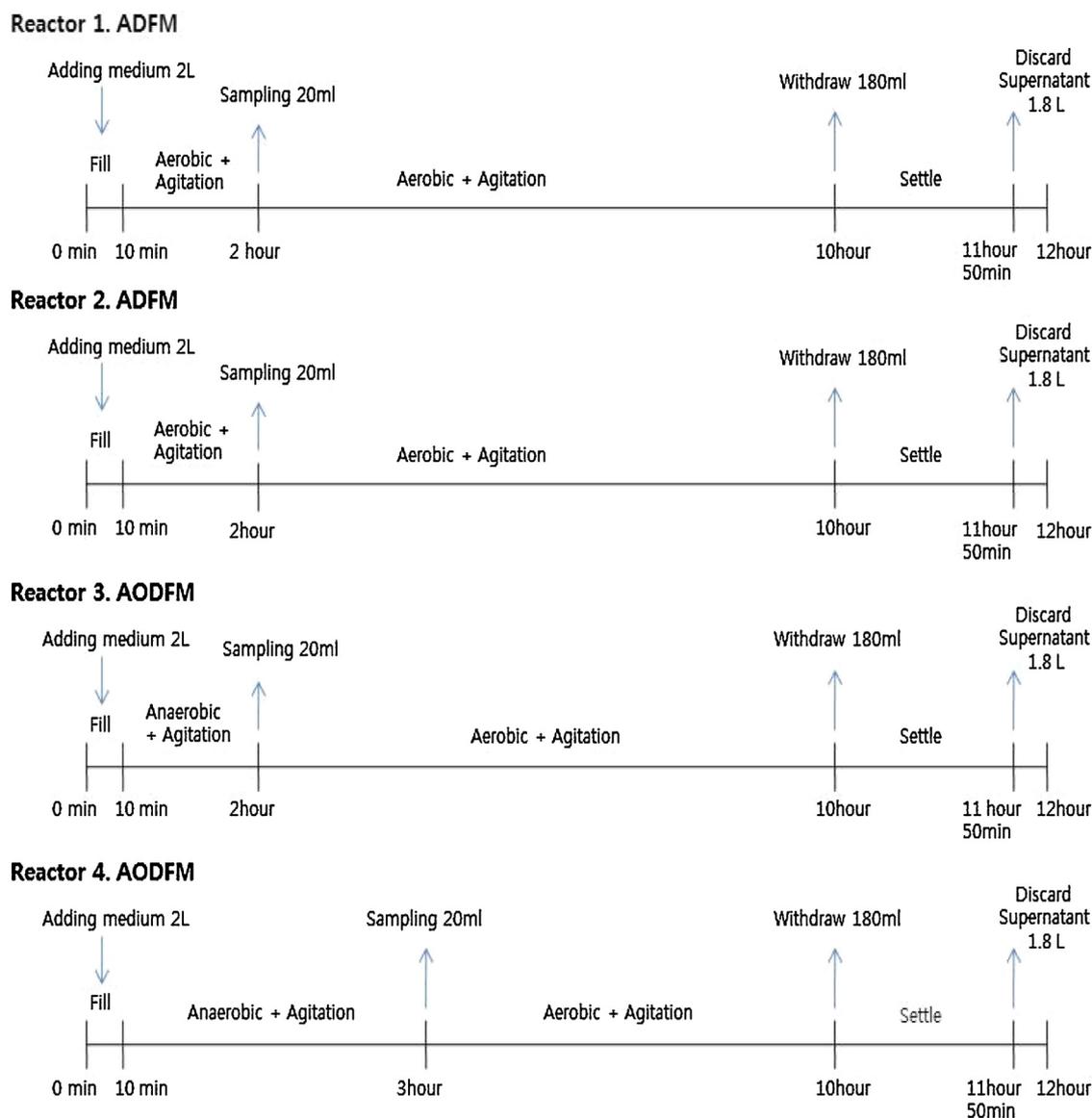


Fig. 1. Reaction scheme for four sequential batch reactors.

feeding mode in an enriched consortium of wastewater sludge biomass. Furthermore, it was to quantify and qualify the physical, chemical and thermal characteristics of extracted polyhydroxyalkanoates from microbial cell biomass of wastewater treatment using sequential batch reactors (SBR).

2. Experimental

2.1. Materials

Grab samples of activated wastewater sludge were collected from Gul-wa, wastewater treatment plant in Ulsan, South Korea. Substrate composition added in each SBR reactor for biomass PHAs growth was purchased as chemical grade. Table 1. shows detail of substrate composition used. Up to phase 4, only synthetic substrate was fed to the sludge biomass as carbon source.

2.2. Setting up of lab reactor

Four sequential batch reactor volumes were set up with substrate containing sodium acetate, nutrients and trace elements. All

four reactors were operated in total for 12 h with mixing for 10 h including 10 min of feeding. Sequential batch reactor 1 and 2 were set in aerobic dynamic feeding mode (ADFM) up to 10 h but at different sodium acetate substrate concentration. Reactor 2, 3 and 4, operated at same sodium acetate substrate concentration. During ADFM oxygen was supplied in reactor 1 and 2 for whole ten hours. Reactor 3 and 4 were set in anaerobic–oxic dynamic feeding mode (AODFM). Operation was performed without oxygen for initial 2 and 3 h, anaerobic, respectively before shifting to ADFM for 8 and 7 h, aerobic, respectively to complete 10 h of mixing (Fig. 1). After 30 days of acclimatization with sodium acetate, all reactors followed 5 major phases (Table 1) while nitrogen/phosphate concentration in all phases were kept fixed along with other nutrient and trace element composition as shown in Table 2.

2.3. Wastewater sludge analysis

On wastewater sludge sampling, mixed liquor suspended solids (MLSS) and PHAs concentration of all samples were measured using following standard methods.

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