Anaerobic Autolysis of Polyhydroxyalkanoates Accumulated in Activated Sludge and the Feasibility of Their Conversion to Bioenergy

その他のタイトル	活性汚泥中に蓄積されたポリヒドロキシアルカン酸の嫌気条件下での自己分解とエネルギー資源への変換の可能性
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論文の内容の要旨
Dissertation Abstract

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（活性汚泥中に蓄積されたポリヒドロキシアルカン酸の嫌気条件下での自己分解とエネルギー資源への変換の可能性）

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The activated sludge processes that remove organic contaminants in wastewater are employed by a wide variety of microorganisms. In this process high energy requirement is one the key issues. Energy consumption may be improved significantly by utilizing microbial power. Some microorganisms can store organic matter in microbial cells as temporal carbon storage such as polyhydroxyalkanoates (PHA). The conversion of organic pollutants in wastewater into PHA requires less energy than usual oxidative wastewater treatment. Further, PHA can be regarded as a source of energy if it is easily converted to bioenergy. Yet, the nature of PHA accumulated in activated sludge is not well known. Usually, waste activated sludge is first thickened and then sent to anaerobic digester. The thickening process usually takes 1-2 days, and if PHA in activated sludge is not stable enough, PHA cannot effectively be converted to bioenergy. On the other hand, if PHA is too stable, it will take longer time to convert it into bioenergy. Thus, it is essential to observe the stability of PHA-accumulated in activated sludge.

The present study was conducted to elucidate the anaerobic autolysis of PHA-accumulated activated sludge and the feasibility to their conversion to bioenergy. In Chapter 1, the background as well as significance of the study and in Chapter 2, relevant research performed so far is concisely described. There have been studies on anaerobic degradation of polyhydroxybutyrate (PHB) but the PHB used in these studies were as biodegradable plastic materials, On the other hand, not so much work has been done on the degradation of PHA-accumulated inside the cells of activated sludge. Yet, mechanisms of anaerobic degradation of PHA in activated sludge are not well understood. Thus it is worth to study the anaerobic autolysis of PHA-accumulated in activated sludge to not only clarify the stability of PHA in activated sludge and but also to evaluate the feasibility of PHA degradation to bioenergy resource.

In order to achieve the objective, several experiments were conducted in this study. In Chapter 3, the materials and methods are explained. In short, waste activated sludge (WAS) was collected from a laboratory-scale

activated sludge reactor and aerobically incubated with organic carbon for 5 to 7 hours to increase the PHA content in WAS. The PHA-accumulated WAS was centrifugally thickened followed by resuspension, dispensed into glass vials, closed air-tight, flushed with nitrogen gas, and incubated at different temperatures without stirring. Samplings were done at the beginning of the incubation and after the incubation on days 0, 1, 2, 3, 4, and 7 or 8. The volatile solids (VS), pH, PHA content and volatile fatty acids (VFAs) were analyzed.

The evaluation of anaerobic autolysis of PHB-accumulated in activated sludge was explained in Chapter 4. The stability of PHB in activated sludge and the mechanisms of PHB degradation under anaerobic conditions were studied. The PHB-accumulated WAS was incubated under anaerobic conditions at 20°C, 30°C and 37°C, and the conversion of PHB to VFAs were observed. There has been an effect of temperatures on the stability of PHB. The degradation of PHB was faster at higher temperature. The degradation was slow at 20°C and only 20% was degraded during the 7 days incubation. At 30°C, 80% of the PHB was remained up to day 3 and on day 4, it rapidly went down to 30% and then the degradation was slowed down. At 37°C, 78% of PHB was degraded within initial 3 days and then degradation was almost stopped. Two novel fermentation models which include the formation of energy by the conversion of PHB into acetate and butyrate were postulated as the possible degradation mechanisms.

On the other hand, in Chapter 5, the anaerobic autolysis of P(3HB-co-3HV)-accumulated in activated sludge was investigated in the same condition. In this study, the stability of P(3HB-co-3HV) and the mechanisms PHV degradation were observed. The P(3HB-co-3HV) remained stable at 20°C and only 25% was degraded during 8-day incubation. At 30°C, the degradation was linear and by day 8 the degradation was 69% during the whole incubation time. At 37°C, the degradation was slow for initial 2 days and the last 5 days but it was rapid in between day 2 and 3. The P(3HB-co-3HV) degradation was 76% by day 8. The degraded P(3HB-co-3HV) was mostly converted to VFAs and the amount of VFAs generated corresponded to 85% to 103% of degraded PHA. Further, the novel anaerobic degradation model for PHV was postulated which yields ATP by the conversion of PHV to acetate, propionate and valerate.

These two experimental results showed slow degradation of PHB and P(3HB-co-3HV) at 20°C for the whole incubation time, initial 3 days at 30°C for PHB (Chapter 4) and initial 2 days at 37°C for PHV (Chapter 5) and then they were degraded rapidly by a day and again slowed down. Based on these results, the degradation of PHB, and P(3HB-co-3HV) is thought to be small and negligible during sludge thickening. As a result of the rapid degradation at 37°C, the PHB and P(3HB-co-3HV) can be a source of bioenergy.

In Chapter 6, the anaerobic autolysis of PHA-accumulated WAS was investigated and the dynamics of bacterial community by 16s rRNA pyrosequencing was studied. The experiment was conducted to study the behavior of PHA at 37°C with a view to observe their contribution to bioerergy resource. There were four sets of batch experiments depending on different feeding percentages of organic carbon to activated sludge biomass to increase the PHA content and the accumulated-PHA were anaerobically incubated at 37°C. The 3HB degradation was observed higher followed by 3HV, 3H2MV, and 3H2MB. The 3HB was degraded rapidly by day 2 and the 3HV and 3H2MV remained stable for a day and then the rapid degradation was observed in between day 1 and 2. Later the degradation was slowed down. In all sets, the degraded PHAs were converted to VFAs. Clostridia, one of the most interesting bacterial species was identified which gradually increased during the incubation period.

Finally, in Chapter 7, estimation of energy flow for the wastewater treatment process was described with a view to clarify the feasibility of the conversion of organic matters to bioenergy via PHA. By considering the PHA-accumulated sludge, the aeration energy will reduce and excess bioenergy will recover when PHA-accumulated WAS will be used as substrate for methanogens.

In Chapter 8, conclusions and some recommendations for future study were suggested. In the present study, the author point out a new way to recover energy in wastewater treatment via PHA. Organic pollutants in wastewater are first converted to PHA in activated sludge and then, the sludge will be thickened, and sent to anaerobic digestion. During thickening, degradation of PHA will be small, and the accumulated PHA will easily be converted to bioenergy via VFAs. The study showed the stability of PHA-accumulated activated sludge and their contribution to bioenergy. Still, there is much scope to study on the efficiency, applicability and finally, feasibility of the conversion of organic matters in wastewater to bioenergy via PHA.