Direct PHA production from sewage sludge: a scalable strategy for wastewater resource recovery

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Abstract. Polyhydroxyalkanoates (PHA) are biodegradable polymers with wide-ranging industrial applications. Producing PHAs from sewage sludge offers a sustainable waste valorization and biopolymer synthesis strategy. Sewage sludge, a byproduct of wastewater treatment, is rich in organic carbon and thus serves as a suitable substrate for microbial PHA production. This process reduces the environmental burden of sludge disposal and enhances the economic feasibility of biopolymer production by using low-cost feedstock. Despite promising advances, large-scale implementation remains limited due to insufficient economic and ecological validation, as most studies focus on laboratory-scale optimization. This study presents encouraging preliminary results from batch experiments using three types of sludge: conventional activated sludge, integrated fixed-film activated sludge and mixed microalgae-activated sludge. The direct accumulation strategy was employed through controlled pulse-feeding and PHA was extracted using the green solvent 2-methyltetrahydrofuran. Preliminary results show consistent PHA production for activated sludge, achieving 23.1 ± 4.5 gPHA g^{-1} VSS. The extraction process yielded high purity (85 ± 6%) and recovery efficiency (87 ± 2%). Ongoing experimental activities will test the efficiency of different sludge samples, validating the integration of the direct accumulation approach into future water resource recovery facilities paving the way for environmentally sustainable and economically viable biopolymer production.

Keywords: Biosolids; Polyhydroxyalkanoate; Resource recovery.

1. Introduction

Polyhydroxyalkanoates (PHA), a family of bio-based polyesters, are produced by microorganisms from renewable sources like sugars and lipids. PHA have similar properties to conventional plastics and are biodegradable in various environments, helping reduce plastic pollution (Kourmentza et al., 2017; Mannina et al., 2020). Naturally accumulated by bacteria as energy reserves under nutrient-limited conditions, PHA has attracted research focused on lowering production costs and improving scalability. Strategies include using renewable feedstocks and mixed microbial (Liu et al., 2023). However, despite their potential, PHA is still far from large-scale deployment due to challenges like high costs, process variability and limited environmental assessments. In this context, direct accumulation has emerged as the most promising strategy for integrating PHA production into wastewater treatment plants, avoiding the preliminary enrichment step and directly exploiting the PHA producers within the withdrawn sludge. This study evaluates the feasibility of the direct accumulation approach using three different sludge matrices: conventional activated sludge (CAS), integrated fixed-film activated sludge (IFAS) and mixed microalgae-activated sludge.

2. Materials and methods

The experimental activities were conducted at the Water Resource Recovery Facility (WRRF) laboratory of the University of Palermo, Italy. The accumulation was carried out in batch-scale reactors adopting three different sludge matrices (Figure 1). The different sludge samples were withdrawn from the wastewater treatment line of the pilot plant hall of the WRRF. The carbon source used during the accumulation was composed of acetic acid (70%) and propionic acid (30%). The carbon source feeding was automatically controlled by tailor-made software adopting a feed-on-demand strategy. Tests were stopped when no substrate consumption was detected after about 2 h from the last injection. During the experiments, samples (ca. 20 mL) were collected and immediately mixed with formaldehyde (36% w/v in H_2O) to stop the biological activity. All the samples were centrifuged (40 min, 4046 RCF) and the obtained pellets were stored overnight at −80 °C and then lyophilised. Lyophilised samples were used to analyse the PHA concentration according to the protocol described by Werker et al., 2008. PHA extraction was carried out by mixing lyophilised samples (ca. 50 mg) with 1 mL of 2-methyl tetrahydrofuran (11 RCF, 75 °C, 3 h). The pellet obtained after the centrifugation was dried at 60 °C overnight. PHA purity and recovery yield were calculated in Equations 1 and 2, respectively.

$$PHA\ purity\ \% = \frac{Mass\ PHA}{Extracted\ polymer\ mass}$$ Equation 1

$$PHA\ rec.yield\ (\%) = \frac{PHA\ extraction\ yield * PHA\ purity}{PHA\ wt\ \%\ in\ lyophilized\ biomass}$$ Equation 2

3. Results

PHA concentration was calculated as the sum of polyhydroxybutyrate (PHB) and polyhydroxyvalerate (PHV) monomers. The preliminary results of the PHA

accumulation and extraction for sludge withdrawn from CAS are reported in Figure 2. Over a period of six hours, a progressive increase in total PHA content was observed, reaching approximately 23% gPHA g^{-1}VSS. The predominant monomer was PHB (18.4 ± 3.5 % gPHA g^{-1}VSS), with PHV slightly contributing to the overall PHA composition achieving 4.7 ± 1.0 % gPHA g^{-1}VSS. Notably, PHA productivity peaked during the initial phase of the accumulation (0.45 ± 0.04 gPHA $hour^{-1}$), gradually declining over time, suggesting optimal polymer synthesis rates in the early stages of substrate availability. Additionally, the extraction method using 2-methyltetrahydrofuran achieved a high purity of 85 ± 6% and recovery efficiency of 87 ± 2%, demonstrating the efficacy and environmental sustainability of the solvent-based approach.

4. Conclusion

Preliminary results demonstrate the promising potential of directly producing PHA from non-enriched sludge. High PHA accumulation coupled with effective eco-friendly solvent extraction underscores this approach's environmental and economic viability. These findings pave the way for future integration of direct PHA production into WRRF, contributing to sustainable waste management and circular economy objectives. Future experiments with diverse sludge matrices will further validate the scalability and robustness of this promising biopolymer production strategy.

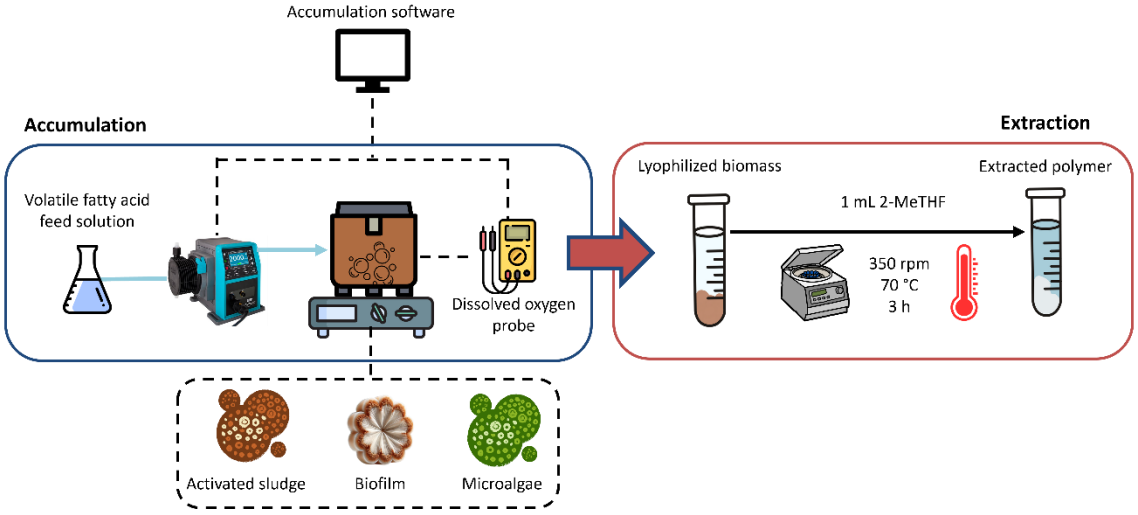

Figure 1. Schematic representation of the experimental setup.

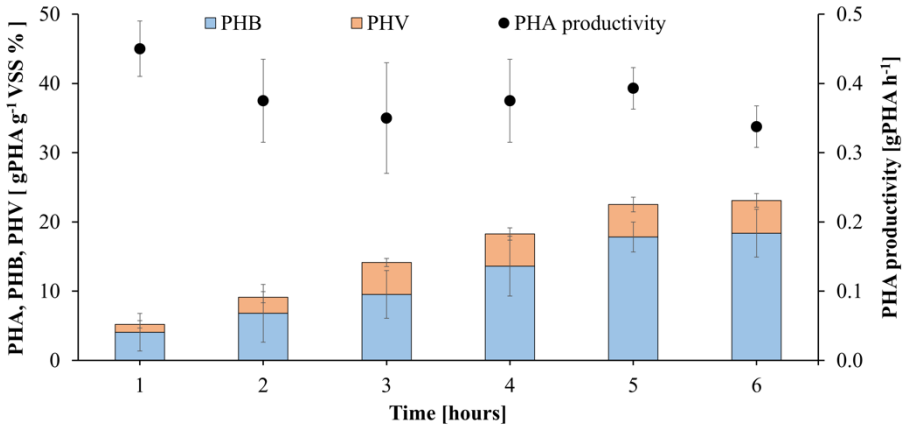

Figure 2. PHA concentration and productivity for sludge withdrawn from conventional activated sludge systems.

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