

Repeated Batch Cultivation: A Novel Empty and Fill Strategy for the Enhanced Production of a Biodegradable Polymer, Polyhydroxy Alkanoate by *Alcaligenes latus*

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Abstract : In the present study, a simple drain and fill protocol strategy of repeated batch was adopted for enhancement in polyhydroxyalkanoates (PHAs) production using *alcaligenes latus* DSM 1124. Repeated batch strategy helped in increasing the longevity of otherwise decaying culture in the bioreactor by supplementing fresh substrates during each cycle of repeated-batch. The main advantages of repeated batch are its ease of operation, enhancement of culture stability towards contamination, minimization of pre-culture effects and maintenance of organism at high growth rates. The cultivation of *A. latus* was carried out in 7 L bioreactor containing 4 L optimized nutrient medium and a comparison with the batch mode fermentation was done to evaluate the performance of repeated batch in terms of PHAs accumulation and productivity. The statistically optimized medium recipe consisted of: 25 g/L Sucrose, 2.8 g/L (NH₄)₂SO₄, 3.25 g/L KH₂PO₄, 3.25 g/L Na₂HPO₄, 0.2 g/L MgSO₄, 1.5 mL/L trace element solution. In this strategy, 20% (v/v) of the culture broth was removed from the reactor and supplemented with an equal volume of fresh medium when sucrose concentration inside the reactor decreased below 8 g/L. The fermenter was operated for three repeated batch cycles and fresh nutrient feeding was done at 27 h, 48 h, and 60 h. Repeated batch operation resulted in a total biomass of 27.89 g/L and PHAs concentration 20.55 g/L at the end of 69 h which was a marked improvement as compared to batch cultivation (8.71 g/L biomass and 6.24 g/L PHAs). This strategy demonstrated 3.3 fold and 1.8 fold increase in PHAs concentration and volumetric productivity, respectively as compared to batch cultivation. Repeated batch cultivation strategy had also the benefit of avoiding non-productive time period required for cleaning, refilling and sterilization of bioreactor, thereby increasing the overall volumetric productivity and making the entire process cost-effective too.

Keywords : *alcaligenes*, biodegradation, polyhydroxyalkanoates, repeated batch

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