Scaling up of poly (3-hydroxybutyrate) production from oil palm frond juice by *Cupriavidus necator* (CCUG52238T)

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Abstract

It has been reported that oil palm frond juice (OPF) can be used as the sole renewable, alternative and cheap carbon source for the production of poly(3-hydroxybutyrate), P(3HB) at lab scale study. In order to further investigate the potential of OPF juice for P(3HB) production at larger scale, we scaled up the fermentation process from 500 ml shake flask to 20 L bioreactor based on constant volumetric mass transfer coefficient ($k_{la}$) of oxygen at both scale. It was interesting to note that the P(3HB) content obtained in this study was almost similar as reported in the previous study both in shake flask and 2 L bioreactor. The P(3HB) content for shake flask fermentation was 45 wt.%, while 44 wt.% and 42 wt.% in 2 L and 20 L bioreactor respectively.

Based on the current finding, it can be concluded that the scaling up method based on constant $k_{la}$ was successful due to the almost similar P(3HB) content obtained in shake flask and bioreactor scale. Overall, it can be postulated that OPF juice can be successfully used as a non-food fermentation feedstock for the production of P(3HB) both in small and large scale fermentation.

Keywords: Oil palm frond, renewable sugars, non-food fermentation feedstock, poly(3-hydroxybutyrate), volumetric mass transfer coefficient

Introduction

During recent years, a variety of biopolymers have become available for use in many applications that are not only compatible with human lifestyle but also are friendly to the environment. As our standing of the biosynthesis of biopolymers and fermentation process development has been advanced, it has become possible to produce an increasing number of biopolymers, in adequate quantities from renewable resources.¹ Today, some of these biopolymers are produced by bacterial fermentation and are used commercially in a wide range of applications such as foods, pharmaceuticals, plastics and agriculture. Not only refined carbohydrates but also agricultural and dairy byproducts can be used as substrates for the production of these biopolymers by fermentation process.¹

Some important biopolymer, for example polyhydroxyalkanoates (PHAs) have been drawing much attention because their physical properties are close to that of conventional plastics. In addition to that, PHA has been found to be superior compared to petrochemical-derived plastics in several aspects that include biocompatibility, biodegradability, and both environmental and human compatibility. However, industrial production of PHA is hindered due to their high production costs compared with chemically synthesized polymers that possess similar material characteristics. Consequently, much effort has been devoted to the development processes for biopolymer production by optimizing the upstream to downstream engineering strategies including the metabolic and cellular engineering of host cells, efficient fermentation and recovery process, and post-production modification of the PHA obtained.¹

One of the major bottlenecks facing many researchers to produce P(3HB) in large scale is the problem of scaling up the process for industrial scale production. For instance, most of the fermentation process performance could not sustain at larger scale compared to the lab scale. This was due to the difficulty in maintaining homogeneity in large systems, changes in surface to volume ratios, and changes in the cultures themselves due to the increased length of culture time.² Therefore, it is important to scale up the fermentation process from shake flasks to industrial scale in stages. For instance, the scaling up process should be done from shake flasks to 2 L bioreactor, then from 2 L to 20 L bioreactor and so on. This is due to fact that the main objective of scaling up process is to obtain a similar total production per unit volume at both scales in the same cultivation time.³

Oxygen transfer rate (OTR) is the most important parameter implied on the design and operation of aeration and agitation of bioreactors and in scale-up.⁴,⁵ Efficiency of aeration depends on oxygen solubilization, diffusion rate into broths, and bioreactor capacity to satisfy the oxygen demand of microbial population. However, the DO in the broths is limited by its consumption rate on cells or the oxygen uptake rate (OUR) as well as by its OTR.

The OTR could be affected by several factors such as geometry and characteristics of the vessels, liquid properties (viscosity, superficial tension etc.), the dissipated energy in the fluid, biocatalyst properties, concentration, and morphology of microorganisms. The OTR value depends on the air flow rate, the stirrer speed, mixing etc.