Screening and selection of a cyanobacteria for production of poly-β-hydroxybutyrate in a closed photobioreactor

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Abstract

Polyhydroxybutyrate (PHB) is a melt-processable, semi-crystalline thermoplastics made by biological fermentation. A key feature of this plastic is its biodegradability.

PHB is currently produced by bacterial fermentation, and is constrained by its high production costs compared to the more conventional petroleum derived polymers with comparable properties. PHB is also produced within certain species of microalgae. These photosynthetic microooganisms use CO_2 as their sole carbon source and so offer a potentially cheaper method for producing PHB, as well as sequestering what would otherwise be a contributor to global warming. For this process to be successful it is necessary to find a species of cyanobacteria that has a high occurrence of PHB within the cells and is also suitable for commercial production. In this research, selection criteria were developed for the screening of microalgae for PHB accumulation and suitability for culture in a novel closed photobioreactor (CPBR), developed by CSIRO using a "top-down" approach. The selection criteria were developed, through a series of preliminary experiments, and economic and environmental considerations. Preliminary experiments were conducted in the CPBR using *Synechoccous* PCC7002, a species of cyanobacteria thought to produce PHB, to identify any system specific selection criteria. The experiments were conducted at several different light and temperature boundary conditions of the CPBR to determine the characteristics of the microalgae. From these experiments it was found that for the alga to be successfully cultivated in the CPBR it must be able to withstand bubbling aeration and not form microbial mats. From the economic considerations it was determined that high productivity, high final cell density, and high PHB content are desirable. Looking at the environmental considerations, it is also necessary that a native species of microalgae is used.

Following a literature review many species of microalgae that produce PHB were found however there was a paucity of literature concerning the potential of Australian species to produce PHB. The following six species of microalgae were identified as possible PHB producers, as they were similar to species already known to prduce PHB: *Anabaena flos-aquae*, *Anabaena solitaria*, *Nodularia spumigenia*, *Pseudanabaena*, *Microcystis aeruginosa*, and *Microcystis flos-aquae*. These species were tested against the selection criteria to choose a species for growth in the CPBR. Initial gross screening was conduced in aerated flasks to identify if the species were able to produce PHB and met the system specific selection criteria; rigorous screening was then conducted on the remaining species that passed the initial gross screening. Rigorous screening was conducted in flasks under different nutrient conditions to determine which species had the highest productivity, cell density, and PHB content. From these experiments a final candidate was selected, *Anabaena solitaria*. The methodology was then validated by cultivating the selected species in the CPBR; this was successful with *Anabaena* solitaria accumulating 8.5 mg L^{-1} PHB under the balanced conditions.

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