Propylene Carbonate Extraction for Recovery of Poly-3-hydroxybutyrate

Abstract

Poly-3-hydroxybutyrate (PHB) is a biopolymer produced as a storage molecule for energy and carbon under poor growth conditions in certain organisms. PHB is a class of polyhydroxyalkanoates (PHAs), which have similar properties to plastics derived from petroleum. PHB provides an environmentally friendly alternative to petroleum-based plastics due to its biodegradability and method of production. It can be produced by recombinant Escherichia coli harvested from the cell, and recovered. One of the major expenses of producing PHB is the extraction of the PHB from the biomass. This study focused on evaluating different methods of PHB extraction for their ability to effectively extract PHB from E. coli biomass and the feasibility of scaling up those processes. These processes provide a way to produce PHB from growing E. coli on a waste product generated from algae processing.

Poly-3-hydroxybutyrate Extraction Techniques

There are several procedures published to extract PHB from biomass of different organisms. Some procedures use toxic solvents recovering high yields and purities, but use large quantities of hazardous solvents making it difficult to scale up to handle larger quantities of biomass. During the course of this project various extraction procedures were tested for their ability to extract PHB from the biomass, safety of the chemicals being used, the capability of the procedure to extract wet biomass, and finally the feasibility of scaling up the procedure being evaluated.

Evaluation of Different PHB Extraction Methods

<table>
<thead>
<tr>
<th>Method of Extraction</th>
<th>Are materials toxic?</th>
<th>Can use wet biomass</th>
<th>Is solvent required?</th>
<th>Solvent to Process 1 kg Biomass</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chloroform</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>10 L</td>
</tr>
<tr>
<td>Sodium Hydroxide</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>1 L</td>
</tr>
<tr>
<td>Acetone</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>10 L</td>
</tr>
<tr>
<td>Propylene Carbonate</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
<td>725 mL</td>
</tr>
</tbody>
</table>

Propylene Carbonate

Propylene Carbonate is an organic solvent produced by combining propylene oxide with carbon dioxide. It is considered a polar aprotic solvent (no proton to donate). Due to its characteristics (low vapor pressure, stability, and high polarity) it is used in many applications. These include production of fibers and textiles, dyeing, personal care agents and cosmetics, and a component of some adhesives. It is also considered non-toxic making it more suitable to use at larger scales.

Propylene Carbonate Extraction Procedure

Propylene Carbonate was first tested at the test tube scale, using 0.5 g to 1 g of dried E. coli biomass per sample. Confirmation that the Propylene Carbonate procedure was extracting PHB from the biomass and isolation of the PHB was possible was achieved by gas chromatography. Once confirmed, the Propylene Carbonate extraction procedure was scaled up to run 40 g of dry E. coli biomass per run. The procedure is described in the figure and pictures below:

Distillation

Distillation apparatus to recover Propylene Carbonate and Ethanol from waste stream

Results

The Propylene Carbonate extraction method was evaluated to test its effectiveness at recovering PHB from dried E. coli biomass. This was done using 0.5 g of E. coli biomass containing approximately 20% PHB (predetermined by GC). Yield was based on the mass of PHB recovered and the purity was based on results obtained from the GC (ratio of the mass of PHB determined by GC to mass of extract analyzed). Both fresh Propylene Carbonate and Distilled Propylene Carbonate were tested.

Future work

Optimize Propylene Carbonate Extraction parameters for E. coli biomass
  • Digestion time with Propylene Carbonate and temperature
  • Possible Pretreatments
    • Temperature
    • pH
    • Precipitation techniques

Energy and Mass Balance
  • Determine the overall effectiveness of this extraction procedure in relation to scale up.

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