JOURNAL OF
EMERGING INVESTIGATORS

Article

Comparative life cycle analysis: Solvent recycling and improved dewatering scenarios in PHB plastic production

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SUMMARY

Bioplastics have become a major focus of international research aiming to achieve sustainability. With the high technical similarity between bioethanol and polyhydroxybutyrate (PHB) bioplastic production, there lies an opportunity for Taiwan's bioethanol producers to expand into the growing bioplastics industry. Using SimaPro software and the ReCiPe 2016 Hierarchist assessment method to conduct life cycle analysis, this investigation aimed to provide insight into sustainable PHB production. Synthesizing data from literature, we determined the types and quantities of inputs and outputs in PHB production, then used SimaPro's built-in Ecoinvent datasets to tally the environmental impact scores of raw materials and pollutants. We compared the environmental impact of a standard PHB plastic production process to two proposed scenarios: one incorporating solvent recycling through distillation and another using an optimized dewatering process that replaced filter presses with centrifuges. According to existing studies, we hypothesized both solvent recycling and dewatering centrifugation scenarios would have lower overall environmental impact than the standard production process. However, our results revealed that only the dewatering centrifugation scenario achieved an overall decrease in environmental impact across 17 ReCiPe impact categories, ranging from carbon emissions and ecotoxicity to land use and water consumption. On the other hand, incorporating solvent recycling led to a 70% decrease in mineral resource consumption and a 42% decrease in water consumption but also a 45% increase in carbon emissions. Due to their energy-intensive nature, current distillation technologies for solvent recycling cannot yet significantly decrease the impact of PHB production; instead, it is more effective for manufacturers to focus on optimized dewatering to reduce environmental impact.

INTRODUCTION

Petroleum-based plastics have become ubiquitous in society due to their myriad uses; however, their prevalence threatens environmental health and sustainability. The U.S. Environmental Protection Agency estimates a global production of 400 million tons of plastic waste in 2022 alone (1). Due to its low biodegradability and tendency to fragment, plastic waste results in multiple impacts on human and environmental health. Microplastics, which are plastic particles less than five millimeters in size, have been detected in the human liver, kidney, and placenta, while chemical additives leached from landfilled plastic and fumes emitted from plastic combustion can result in health disorders and potentially cancer (1). Furthermore, more than 1500 animal species in marine and terrestrial environments are known to mistakenly ingest plastic, which may lead to indigestion, starvation, and internal injuries (1). Besides the detrimental effects of petrol-based plastics when disposed of in the environment, the manufacturing of plastics from fossil fuels also requires destructive mining practices and relies on non-renewable reserves (1). Considering the harmful effects of petrol-based plastics, biodegradable and bio-based plastics offer a potential solution.

Biodegradable and bio-based plastics can minimize harm to humans and wildlife. Bio-based plastics do not require fossil fuel excavation as they are manufactured through bacterial fermentation of organic raw materials such as corn or sugar (2). Degrading within much shorter time frames, biodegradable plastics reduce health risks and eliminate the need for incineration, leading to lower overall carbon footprints from production to disposal (2). To achieve a sustainable global future, there is an urgent need to develop biodegradable and bio-based plastic alternatives.

Polyhydrodalkanates (PHA) bioplastics, which can be synthesized by bacteria using organic waste, are a promising solution. Produced from bacterial fermentation with sugar, PHAs are both biodegradable and biocompatible, meaning they can be broken down into harmless organic products by bacteria and other organisms, while being non-toxic to living tissue (2, 3). Owing to its potential as an environmentally friendly alternative to petrol-based polyethylene terephthalate (PET) and polyethylene (PE) plastics, the production capacity for PHAs has grown by 41% between 2010 and 2017 (2). However, the commercial success of PHA still faces challenges due to the high cost of pure beet sugar and cane sugar substrates; thus, the use of low-cost organic waste substrates is of special interest in bioplastic research (2).

Within the category of PHAs, polyhydroxybutyrate (PHB) is one type of plastic that currently has the most developed manufacturing technology (4). PHB plastic is produced by cultivating particular species of bacteria under nutrient-limiting conditions, causing the bacteria to accumulate PHB within its cell cytoplasm; then, the PHB is extracted and dried to produce granular plastic (4). To extract PHB from the cell cytoplasm, propylene carbonate has been proposed as an environmentally friendly solvent. Propylene carbonate is non-toxic to wildlife, can be synthesized from carbon capture processes, and can also be recycled through distillation since it exhibits a higher boiling point than water, reducing production costs (4, 5). However, the use of solvent recycling in PHB production is not yet well-studied. Another challenge in PHB production efficiency is the large energy demand of dewatering and drying plastic granules; it is estimated that extraction, purification, and drying processes account for 60% of production costs in PHB manufacturing (4).

In Taiwan, the bioplastic industry faces numerous challenges. According to Taiwan's Ministry of Environment, the country consumes an estimated 7,000 tons of polylactic acid (PLA) bioplastic per year; yet Taiwan lacks the resources, infrastructure, and regulations for bioplastic production and recycling, relying mostly on imported plastic (6). Furthermore, although PLA bioplastic is compostable, it is not readily biodegradable when disposed of in the natural environment and thus requires professional methods for disposal (2). In Taiwan, only approximately 5% of PLA is recycled, while most is incinerated with other municipal waste (6). Consequently, there is a need for Taiwan to develop more efficient and advanced bioplastic production and disposal technology.

PHB has superior biodegradability to PLA, and PHB manufacturing also offers a unique economic opportunity for Taiwan (2, 7). Through decades of development, Taiwan has achieved mature technology for producing bioethanol by bacterial fermentation of cane molasses. According to an investigation conducted by the Industrial Technology Research Institute of Taiwan, there is potential for Taiwan's bioethanol producers to expand into the growing industry of bioplastic production, since bioethanol and PHB production involve similar equipment and technology (7).

PHB can be produced using a large variety of industrial methods. Before implementing production in Taiwan, it is necessary to examine which methods are most efficient and eco-friendly. To achieve this, we used SimaPro software to perform life cycle analysis (LCA) on three different PHB production methods, quantifying and comparing their environmental impact. This investigation used ReCiPe 2016 v1.1 impact assessment method, a recognized LCA method to analyze the quantities of inputs and outputs of a process, generating environmental impact scores in 17 midpoint categories and 3 endpoint categories, representing various impacts from carbon emissions to land use and water consumption (8, 9). First, we conducted LCA for a standard PHB production process using waste molasses substrate, *Cupriavidus necator* bacteria, and propylene carbonate solvent, referred to as Scenario A (**Figure 1**). Targeting the biggest contributors to environmental impact in Scenario A, we then proposed two improved scenarios B and C, which we hypothesized to have a lower environmental impact than the standard process. The goal of this investigation was to compare the LCA results of Scenarios A, B, and C to determine key factors that contribute to the efficiency and sustainability of PHB production.

In Scenario B, instead of disposing and using new propylene carbonate for each extraction batch, we modeled a solvent recycling process which uses distillation to recover propylene carbonate for repeated use. Pavan, et al. determined that recycling propylene carbonate solvent can reach a

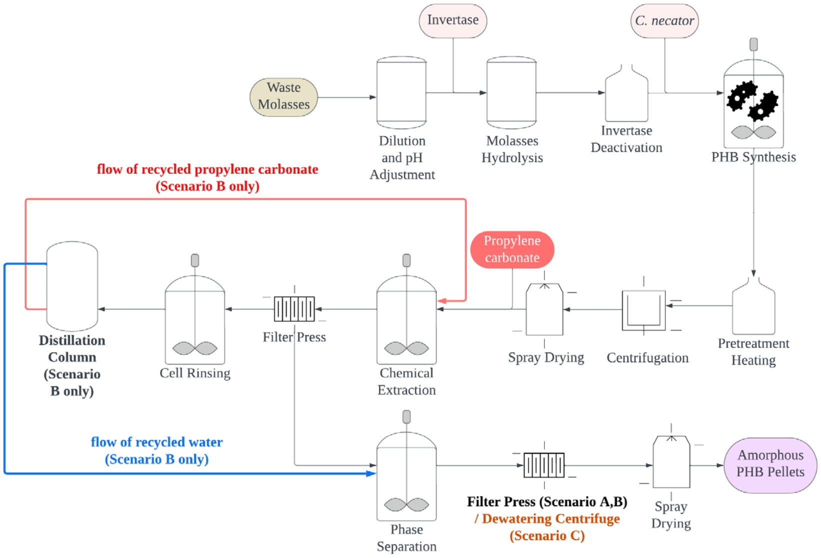

Figure 1. PHB production process from waste molasses, for Scenarios A, B, and C. The steps in industrial PHB manufacturing, from pretreatment of raw material and PHB synthesis to extraction and drying, indicating key differences in the three scenario analyzed in this study: Scenario A, the standard process, Scenario B, which incorporates solvent recycling, and Scenario C, which replaces the filter press with a dewatering centrifuge.

https://doi.org/10.59720/24-107

high efficiency of 99.82% (4). Thus, recycling and reusing propylene carbonate would eliminate the need to produce new solvent, suggesting Scenario B would effectively reduce the environmental impact of PHB production compared to Scenario A.

In Scenario C, we replaced the filter presses in Scenario A with dewatering centrifuges (**Figure 1**). In contrast with the standard use of a filter press that can reduce PHB moisture to 86% (4), previous work has found that dewatering centrifugation can reach moisture values as low as 11% (10). Furthermore, Uhrig et al. suggested that PHA-rich biomass is most effectively dewatered using centrifugation instead of filter presses due to its physical properties (11). If the PHB plastic has a lower moisture content when entering the spray dryer, the energy demand during the final spray drying step would also be lower. Thus, we hypothesized that Scenario C would also have a lower environmental impact than Scenario A.

After conducting LCA, we found that compared to the standard process, using distillation to recycle the propylene carbonate solvent (Scenario B) led to decreased impacts for terrestrial ecotoxicity, water consumption, and ionizing radiation; however, the total carbon footprint unexpectedly increased. On the other hand, replacing filter presses with more efficient dewatering centrifugation (Scenario C) effectively decreased impacts across all categories. These insights are valuable to PHB manufacturers, industrial engineers, and material engineers worldwide, contributing to the push for sustainable plastic production and a greener future.

RESULTS

PHB plastic production: Hotspots identification

To propose effective improvements to the PHB production process, we first identified key issues in the standard processes (Scenario A). We focused on the particular method of using cane sugar molasses feedstock, synthesis by *C. necator*, and extraction using propylene carbonate (**Figure 1**). Molasses and *C. necator* are already extensively used in PHB production; on the other hand, propylene carbonate is not yet widely used, but offers promising qualities - it can be synthesized through atmospheric carbon capture, while exhibiting low toxicity when disposed (4).

First, we performed hotspot analysis on Scenario A specifically to identify points for improvement within the standard PHB production process. Hotspot analysis involves comparing the proportional environmental impact of each step to determine the most significant sources of environmental impact, or impact "hotspots." To quantify environmental impact, we calculated the quantities of inputs and outputs in each step (**Tables S1 - S3**), then used SimaPro software to convert this data into impact scores based on existing Ecoinvent datasets, which contain known average environmental impact scores of common raw materials or pollutants. We divided Scenario A into three large steps – synthesis, extraction, and purification - , and compared their environmental impact scores across 17 categories, describing a wide range of influences such as global warming, ecotoxicity, land use, and water consumption. For all impact categories, PHB extraction, which involves mixing and heating PHB-rich cells with propylene carbonate, was the main contributor of environmental impact (**Figure 2**). In the category of global warming potential (GWP), defined as the contribution to global warming due to greenhouse gas emissions and measured in kilograms of carbon dioxide, solvent extraction accounted for 58 kg CO_2 per 1 kg PHB produced, which was 68% of the total carbon footprint of production.

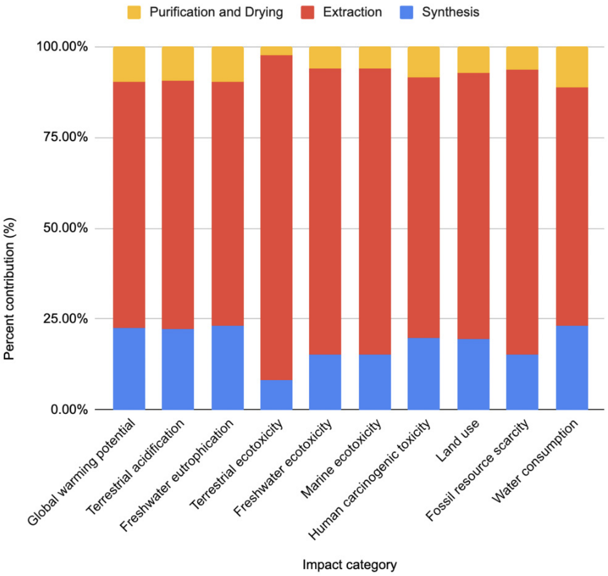

Figure 2. Contribution of each step in PHB production to ten key environmental impact categories in Scenario A. Percent contribution of synthesis, extraction, and drying steps in PHB production for ten environmental impact categories, including global warming, acidification, eutrophication, ecotoxicity, land use, fossil resource scarcity, and water consumption. Assessed with the ReCiPe 2016 Midpoint (H) V1.00 and SimaPro software.

Since PHB extraction accounted for most of the environmental impact in the full production process, we examined PHB extraction in detail by conducting another hotspot analysis on the extraction step only. To pinpoint specific inputs or processes key to high environmental impact, we subdivided PHB extraction into three even smaller steps and again compared their environmental impact scores. Based on the process design from Pavan et al., PHB extraction consists of three main steps (4). First, during pretreatment, the PHB-rich bacteria is heated to 95°C for 45 minutes to allow PHB to be released from the cells. Next, to eliminate extra moisture before extraction, the PHB-cell mixture is spray dried until only 0.6% moisture is present. Finally, during chemical extraction, propylene carbonate is added to dissolve and extract PHB, thus allowing the waste biomass to be separated. The waste emissions of extraction include cane molasses residue, bacterial residue, and used propylene carbonate, which are all organic, low-toxicity and highly biodegradable in nature; thus, their environmental impact is negligible.

Comparing the three sub-steps, we identified chemical extraction using propylene carbonate solvent as the key source of environmental impact (**Figure 3**). In the category of GWP, chemical extraction accounted for 31 kg-CO2 per kg PHB (53% of the total impact of extraction). Pretreatment heating accounted for 13 kg-CO_2 per kg PHB (23% total impact of extraction), while spray drying accounted for 14 kg-CO_2 per kg PHB (24% total impact of extraction).

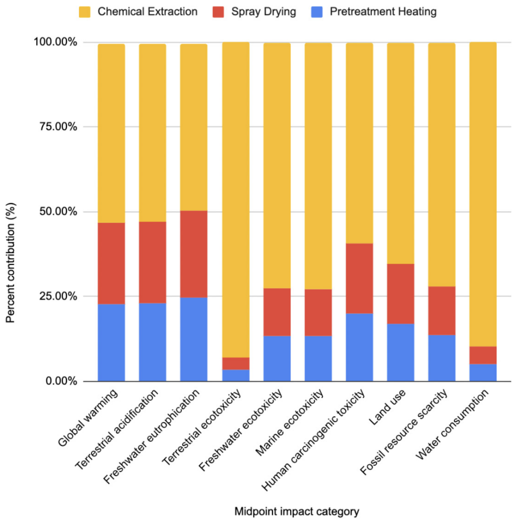

Figure 3. Contribution of each step in PHB extraction to ten key environmental impact categories in Scenario A. Percentage contribution to environmental impact of each detailed step in PHB extraction – pretreatment heating, chemical extraction with propylene carbonate, and spray drying. Assessed with ReCiPe 2016 Midpoint (H) V1.00 and SimaPro software.

Considering these insights, we designed two improved processes, Scenario B and C, to mitigate the impact of chemical extraction and spray drying, respectively. Scenario B incorporated solvent recycling to eliminate the impact of propylene carbonate production, while Scenario C used an improved dewatering process with centrifugation to reduce input moisture and improve the energy efficiency of spray drying. Although pretreatment heating also contributed a substantial impact in the global warming category, it is an essential step to deactivate *C. necator* and thus difficult to eliminate; therefore, this study focused on improving the other two main steps. We then compared scenarios B and C with Scenario A to evaluate the extent of success in reducing environmental impact.

Scenario A, B, and C: Comparison analysis

Assessing the three scenarios together, we found that Scenario B, solvent recycling, led to the lowest environmental impact of all three scenarios in the categories of terrestrial ecotoxicity, water consumption, mineral resource consumption, and ionizing radiation, while Scenario C, the use of centrifugation, led to the lowest environmental impact in all other categories (**Table 1**).

Next, we individually compared Scenarios B and C to the standard process, Scenario A. Compared to the standard process, the use of solvent recycling (Scenario B) achieved a 72.52% reduction in ionizing radiation, a 62% reduction in terrestrial ecotoxicity, a 70% reduction in mineral resource

Impact category	Unit	A (standard)	B (Recycling)	C (Improved dewatering)
Global warming	kg CO_2 eq	1.0E+02 (56%)	1.8E+02 (100%)	6.6E+01 (36%)
Stratospheric ozone depletion	kg CFC-11 eq	1.8E-05 (62%)	2.9E-05 (100%)	1.3E-05 (44%)
Ionizing radiation	kBq Co-60 eq	2.0E+00 (100%)	5.5E-01 (27%)	1.9E+00 (95%)
Ozone formation, Human health	kg NO_x eq	1.3E-01 (69%)	1.8E-01 (100%)	8.7E-02 (47%)
Fine particulate matter formation	kg PM2.5 eq	1.7E-01 (50%)	3.5E-01 (100%)	9.6E-02 (27%)
Ozone formation, Terrestrial ecosystems	kg NO_x eq	1.3E-01 (70%)	1.8E-01 (100%)	9.1E-02 (49%)
Terrestrial acidification	kg SO_2 eq	3.0E-01 (56%)	5.4E-01 (100%)	1.9E-01 (35%)
Freshwater eutrophication	kg P eq	3.3E-02 (53%)	6.3E-02 (100%)	1.9E-02 (29%)
Terrestrial ecotoxicity	kg 1,4-DCB eq	5.1E-02 (100%)	1.9E-02 (38%)	4.6E-02 (92%)
Freshwater ecotoxicity	kg 1,4-DCB eq	1.6E+00 (83%)	2.0E+00 (100%)	1.2E+00 (60%)
Marine ecotoxicity	kg 1,4-DBC eq	2.3E+00 (83%)	2.7E+00 (100%)	1.6E+00 (60%)
Human carcinogenic toxicity	kg 1,4-DBC eq	2.5E+00 (61%)	4.1E+00 (100%)	1.5E+00 (38%)
Human non-carcinogenic toxicity	kg 1,4-DBC eq	1.8E+03 (83%)	2.1E+03 (100%)	1.3E+03 (60%)
Land use	m^2 x yr crop eq	1.2E+00 (70%)	1.8E+00 (100%)	8.1E-01 (46%)
Mineral resource scarcity	kg Cu eq	1.2E-01 (100%)	3.6E-02 (30%)	1.1E-01 (95%)
Fossil resource scarcity	kg oil eq	2.9E+01 (81%)	3.6E+01 (100%)	2.3E+01 (65%)
Water consumption	m^3	7.4E-01 (100%)	4.3E-01 (58%)	6.8E-01 (93%)

Table 1. Impact scores for PHB production scenarios A, B, and C across 17 ReCiPe (H) midpoint categories. Impact scores computed with ReCiPe 2016 Midpoint (H) V1.00 and SimaPro software, displayed in both absolute value (category units) and relative size (%) – the relative percentage compares each scenario to the highest impact scenario, which is scaled to 100%. All units ending in "eq" are defined by the environmental impact when emitting one unit of the reference material into the environment; for example, kg 1,4 – DBC eq refers to kg 1,4-dichlorobenzene equivalents, defined as the increase in ecotoxicity when emitting 1 kilogram of 1,4-DBC to urban air, freshwater, seawater, or industrial soil (9). The unit for land use, m2 x yr, represents square-meter-years, defined by the environmental impact when one square meter of land is transformed or occupied for agricultural purposes over a certain time period (8).

scarcity, and a 42% reduction in water consumption. However, it also led to an increase in carbon emissions by 79%, nearly doubling the impact in the GWP category. On the other hand, the use of centrifugation to replace filter pressing (Scenario C) reduced all categories of environmental impact compared to the standard process, including a 35% reduction in GWP.

To obtain a more succinct interpretation that can assist decision-making in industry, we also conducted endpoint assessment, converting and generalizing the midpoint impacts into three ReCiPe endpoint categories: human health, measured in disability-adjusted life years (DALYs), ecosystem health, measured in species x year, and resource availability, measured with the value of the US dollar in 2013. DALYs represent the burden of disease and injury in a population, defined by the number of years of healthy life that are lost, either due to living with a disability or dying before reaching life expectancy (9). Damage to ecosystem health is represented by the local species loss integrated over time, while damage to resource availability is represented by the extra costs needed for future fossil or mineral extraction to produce the equivalent amount of resources that had been consumed (9).

Scenario B, solvent recycling, caused approximately 50% increase in impacts on both human and ecosystem health compared to the standard scenario (**Figures 4 and 5**). However, it also achieved a 54% reduction in damage to resource availability (**Figure 6**). On the other hand, Scenario C, improved dewatering, achieved a 40% reduction in damage to human health and a 35% reduction in damage to ecosystem health (**Figures 4 and 5**), but led to a minor 2% increase in damage to resource availability (**Figure 6**).

DISCUSSION

Through LCA and hotspot analysis, we determined that solvent extraction and dewatering/drying are key sources of impact in a standard PHB production process using propylene carbonate. By designing and analyzing alternative scenarios, we saw that solvent recycling by distillation is effective in reducing only some of the environmental impacts. The impact categories that decreased, including terrestrial ecotoxicity, water consumption, mineral resource consumption, and ionizing radiation, are all related to the production of propylene carbonate. By introducing a recycling process, we mitigated these impacts by eliminating the need for the input of new propylene carbonate. However, the distillation process required to recycle propylene carbonate involves extra energy consumption, which led to heightened impacts in all other categories. In this study, we estimated the environmental impact of electricity generation using Ecoinvent datasets on coal power in Taiwan, which was the major energy source and made up 42.2% of Taiwan's energy grid in 2023 (12, 13). In this scenario, consuming more energy means burning more coal, releasing more sulfur oxides, nitrogen oxides, mercury, and other toxic pollutants and resulting in increased damage to ecosystems and human health (14). Although we mitigated the disposal of propylene carbonate in this scenario, diluted propylene carbonate is non-toxic, non-carcinogenic, and likely to biodegrade in natural environments; thus, its disposal does not contribute greatly to the overall environmental impact of the process (15). The environmental damage from increased consumption of coal power outweighs the benefits of mitigating propylene carbonate disposal.

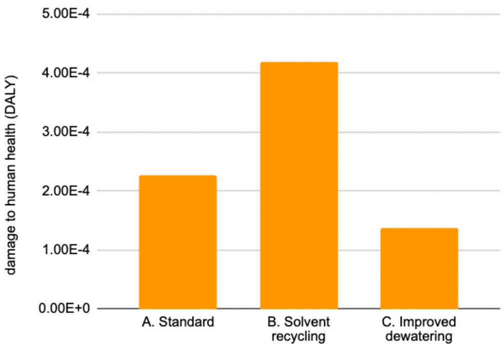

Figure 4. PHB production impact on human health under scenario A, B, and C. The damage to human health resulting from each scenario, measured in Disability-Adjusted Life Years (DALYs), defined by the number of healthy years of life lost in a population, either due to death before reaching life expectancy, or living with a disability. Assessed with the ReCiPe 2016 Endpoint (H) V1.00 and SimaPro software.

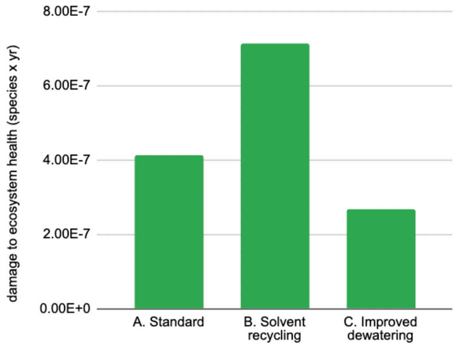

Figure 5. PHB production impact on ecosystem health under scenario A, B, and C. The damage to ecosystem health resulting from each scenario, represented in units of species x yr (species loss integrated over time). Assessed with the ReCiPe 2016 Endpoint (H) V1.00 and SimaPro software.

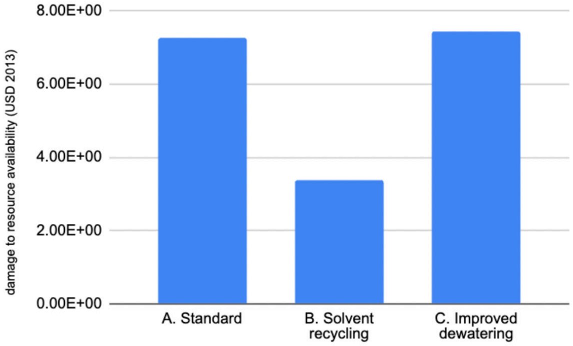

Figure 6. PHB production impact on resource availability under scenario A, B, and C. The damage to resource availability resulting from each scenario, represented by the extra costs associated with future resource extraction, measured in units of the US dollar in 2013. Assessed with the ReCiPe 2016 Endpoint (H) V1.00 and SimaPro software.

Although Scenario B is not beneficial when considering Taiwan's current electrical grid, its impacts may be drastically decreased by transitioning to renewable energy. Considering the energy-intensive distillation process needed to recycle propylene carbonate in Scenario B, transitioning to renewable energy would significantly reduce the impact score in the GWP category. In this case, solvent recycling could conserve resources while not causing overwhelming damage to human or ecosystem health. With renewable energy, an integrated approach combining Scenarios B and C - using both solvent recycling and centrifugation – may lead to the lowest environmental impact overall and is worth investigating in future LCA studies.

However, within the scope of this study, only Scenario C, the use of centrifugation during PHB production, successfully decreased all environmental impacts overall compared to the standard scenario by improving the energy efficiency of extraction and drying. Compared to filter pressing, centrifugation achieves much lower PHB moisture values, and thus, less electricity is needed to thoroughly dry the plastic in the subsequent spray drying process. This leads to less resource consumption and decreased pollution. According to these results, efficient drying is an important improvement and consideration for the PHB industry. Interestingly, the use of centrifugation resulted in a minor 2% increase in the endpoint score for resource availability, contradicting the results of the midpoint assessment. Since endpoint scores are highly aggregated, they naturally have a higher uncertainty, which may be the cause of this minor discrepancy.

Although these findings are promising, there are several limitations of our study that should be considered, such as the use of small-scale laboratory data, which may not accurately reflect industrial-scale production. This may partially explain discrepancies between the total carbon footprint computed by different researchers. The total carbon footprint of producing 1 kg of PHB plastic was determined to be 66 kg CO2-eq, which is higher than most LCA studies on similar production methods (16, 17). This investigation was based on data from Dalsasso, et al., which was a PHB production experiment conducted in a small lab setting, and thus likely used a higher substrate-to-bacteria ratio, using more materials and electricity than required to produce the same amount of PHB compared to industrial-scale production (18). Depending on lab conditions, the percentage accumulation of PHB in cells can also vary. Dalsasso, et al. obtained 56% PHB accumulation; however, there have been reports of higher accumulation percentages, such as 68.8 % and 72.08% (18 - 20). This may explain why the impact scores in this investigation are systematically skewed at the higher end of similar LCA studies. To increase the accuracy and validity of the assessment scores, real-world data from an operational PHB factory, such as Danimer Scientific, can be used in future analyses.

Despite limited data accessibility, this investigation still provides valid insights for the comparison of three PHB production scenarios. We recommend that PHB manufacturers focus on optimizing the dewatering process to decrease their overall environmental footprint by drying the plastic as thoroughly as possible using a centrifuge before spray drying. On the other hand, solvent recycling, which has not yet been widely developed, may be unsuitable for implementation with current technology due to the high carbon footprint of distillation when powered by coal. However, if low-energy solvent recycling methods or renewable energy sources are used, solvent recycling will bring significant benefits for sustainable resource management. Low-energy alternatives to distillation, such as membrane filtration, are significant directions for further research. Due to varied production conditions, bioplastics do not always have competitive pricing or lower environmental impact compared to petrol-based plastics; however, utilizing renewable resources and mitigating plastic pollution is necessary for sustainable development, making bioplastics an invaluable research field for many years to come.

MATERIALS AND METHODS

This investigation used LCA, an analytical procedure, to estimate and quantify the environmental impact of three PHB production scenarios. The first step in LCA is to define the goal and scope of the study, including the functional unit of the product to be assessed. Next, to build the life cycle inventory, data on the inputs and outputs of the process is gathered from literature or experimentation. During life cycle impact assessment, the inventory data is computed through a set of standardized, existing mathematical methods into impact scores, which are numerical representations of how much the product influences different aspects of the environment. Finally, the results are analyzed and interpreted by the researcher, providing insight into sustainable industrial design or policy decisions.

Goal and scope definition

The functional unit considered for life cycle analysis was 1 kg of amorphous PHB plastic pellets, which has the function of serving as the raw material for commercial products such as plastic bags, plastic film, and plastic containers. A second order boundary for capital goods was used—all processes in the cradle-to-gate life cycle (from raw material to PHB plastic pellets) are included, but the production and disposal of capital goods, including bioreactors and fridges, are not considered. Data was gathered entirely from published literature.

Life cycle inventory

The standard cradle-to-gate process (Scenario A) of PHB production consists of the following main stages: molasses pretreatment, PHB synthesis, PHB extraction, and PHB purification and drying (**Figure 1**). In this study, fed-batch fermentation using the bacterial strain *C. necator* and PHB extraction by solvent were chosen for investigation, as they are some of the most well-known and established methods in PHB manufacturing (18). The next step, PHB purification and drying, is where Scenarios A, B, and C differ. In scenario A, after the solid PHB was separated from the solvent using a filter press, it was then spray-dried into amorphous PHB plastic granules. In Scenario B, the leftover water and propylene carbonate mixture from extraction was sent to a distillation column, producing pure propylene carbonate and pure water which was reused in the next round of extraction. In Scenario C, the filter press was replaced by a dewatering centrifuge in the step prior to spray drying.

After designing the steps of each scenario, the life cycle inventory was built – a collection of data on the quantities of inputs and outputs in PHB production, from physical materials to electricity (**Tables S1 - S3**). Data on the quantities of raw materials and byproducts were gathered from the literature, with the pretreatment and synthesis processes based on the work of Dalsasso et al. (**Table S4**) and the extraction and recovery processes based on the work of Pavan et al. (4, 18). The energy consumption of stirred-tank reactors, heating, distillation, and centrifugation were based on formulae proposed by Piccinno et al. (11).

For each reactor, total electricity consumption was calculated by summing the energy consumption of heating (Q_{react}) and stirring (E_{stir}). Qr_{eact} was calculated from the energy consumed for directly heating the material (Q_{heat}), the additional energy required to offset the loss of heat to the environment (Q_{loss}), and the overall efficiency of the system (η_{heat}) (**Eqn 1**) (22).

$$Q_{react} = \frac{Q_{heat} + Q_{loss}}{\eta_{heat}} \qquad \textbf{(Equation 1)}$$

Q_{heat} was calculated by multiplying the specific heat of the material being heated (c_p), the mass of material (m), and the difference between the temperature of material during the reaction (TR) and the ambient temperature of the environment (T_{out}) (**Eqn 2**) (22). For the molasses-water mixtures in this case study, the specific heat of the mixture was estimated by summing up the products of the fraction of each type of component and the specific heat of that component.

$$Q_{heat} = c_p * m * (T_R - T_{out}) \qquad \textbf{(Equation 2)}$$

Q_{loss} was calculated from the surface area of the reactor (A), the insulating material's thermal conductivity (λ), and the insulating material's thickness (s), using constant values suggested by Piccinno et al. for typical insulating materials, including concrete, steel, and polyurethane foam (**Eqn 3**) (22).

$$Q_{loss} = A * \frac{\lambda}{s} * (T_R - T_{out}) \qquad \textbf{(Equation 3)}$$

The energy consumption of stirring (E_{stir}) was estimated using N_p , N, and d, constants related to the stirring impeller design, based on values suggested by Piccinno et al. (**Eqn 4**) (22). It was assumed that the density of all mixtures in this study (ρ_{mix}) was approximately 1200 kg/m^3, close to the density of water, but slighter higher due to the addition of molasses. η_{stir}, the overall efficiency of the stirring system, is assumed to be 0.9.

$$E_{stir} = \frac{N_p * \rho_{mix} * N^3 * d^5}{\eta_{stir}} \qquad \textbf{(Equation 4)}$$

These general formulae were applied to all stirred-tank reactors in PHB production, using parameter values included in the appendix (**Tables S5, S6**). The energy consumption of distillation was also estimated using the same equations, with the addition of the energy needed to vaporize the water (**Table S7**). The latent heat of vaporization of water was taken to be 2,260 kJ/kg (23).

Filter presses and centrifuges, on the other hand, involve more complex mechanics that go beyond heating and stirring; thus, their energy consumption was estimated directly from existing data. To determine the energy consumption of a filter press, data from three sources was considered (24 - 26). Taking an approximate average, it was estimated that a typical filter press requires 30kWh of energy per ton of solid product, equivalent to 108 kJ/kg (**Table 2**). The electricity consumed during centrifugation was estimated to be 18 kJ/kg of material (22). For spray dryers, energy consumption was estimated as 4.87 GJ per ton of water evaporated (27).

Life cycle impact assessment and interpretation

After compiling the life cycle inventory, we then input the data and conducted life cycle impact assessment with SimaPro 8.4.0.0 PhD version, using the ReCiPe 2016 Hierarchist V1.00 midpoint and endpoint assessment methods (9). ReCiPe 2016 includes three different versions, each with certain assumptions and choices – individualist, which prioritizes short-term interests and assumes future technology can achieve sustainability, hierarchist, which is based on scientific consensus of the time frame and degree of environmental impacts, and egalitarian, which is the most precautionary, pessimistic perspective and accounts for all known possible environmental impacts (9). This study used the hierarchist perspective, the default version for life cycle assessment.

ACKNOWLEDGMENTS

We express our most sincere thanks to Pei-Chiun Li, postdoctoral researcher at the Graduate Institute of Environmental Engineering at NTU, who provided invaluable feedback and guidance throughout the study.

Received: April 20, 2024
Accepted: July 25, 2024
Published: June 13, 2025

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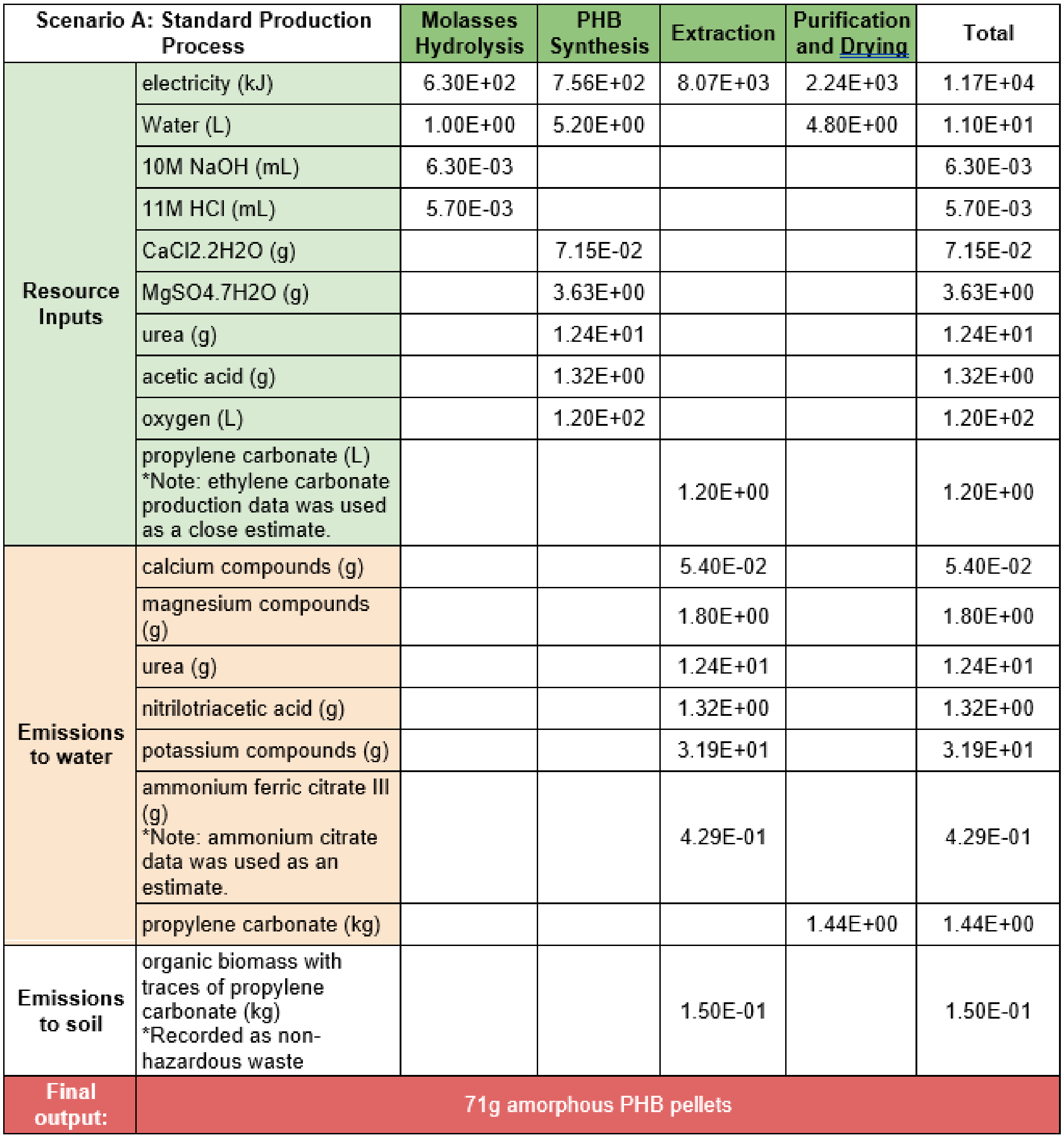
https://doi.org/10.59720/24-107

APPENDIX



Scenario A: Standard Production Process		Molasses Hydrolysis	PHB Synthesis	Extraction	Purification and Drying	Total
Resource Inputs	electricity (kJ)	6.30E+02	7.56E+02	8.07E+03	2.24E+03	1.17E+04
	Water (L)	1.00E+00	5.20E+00		4.80E+00	1.10E+01
	10M NaOH (mL)	6.30E-03				6.30E-03
	11M HCl (mL)	5.70E-03				5.70E-03
	CaCl2.2H2O (g)		7.15E-02			7.15E-02
	MgSO4.7H2O (g)		3.63E+00			3.63E+00
	urea (g)		1.24E+01			1.24E+01
	acetic acid (g)		1.32E+00			1.32E+00
	oxygen (L)		1.20E+02			1.20E+02
	propylene carbonate (L) *Note: ethylene carbonate production data was used as a close estimate.			1.20E+00		1.20E+00
Emissions to water	calcium compounds (g)			5.40E-02		5.40E-02
	magnesium compounds (g)			1.80E+00		1.80E+00
	urea (g)			1.24E+01		1.24E+01
	nitrilotriacetic acid (g)			1.32E+00		1.32E+00
	potassium compounds (g)			3.19E+01		3.19E+01
	ammonium ferric citrate III (g) *Note: ammonium citrate data was used as an estimate.			4.29E-01		4.29E-01
	propylene carbonate (kg)				1.44E+00	1.44E+00
Emissions to soil	organic biomass with traces of propylene carbonate (kg) *Recorded as non-hazardous waste			1.50E-01		1.50E-01
Final output:	71g amorphous PHB pellets					

Table S1. Life cycle inventory of PHB bioplastic production, Scenario A. The material inputs and outputs of producing 71 grams of amorphous PHB pellets through production scenario A, including quantities and units.

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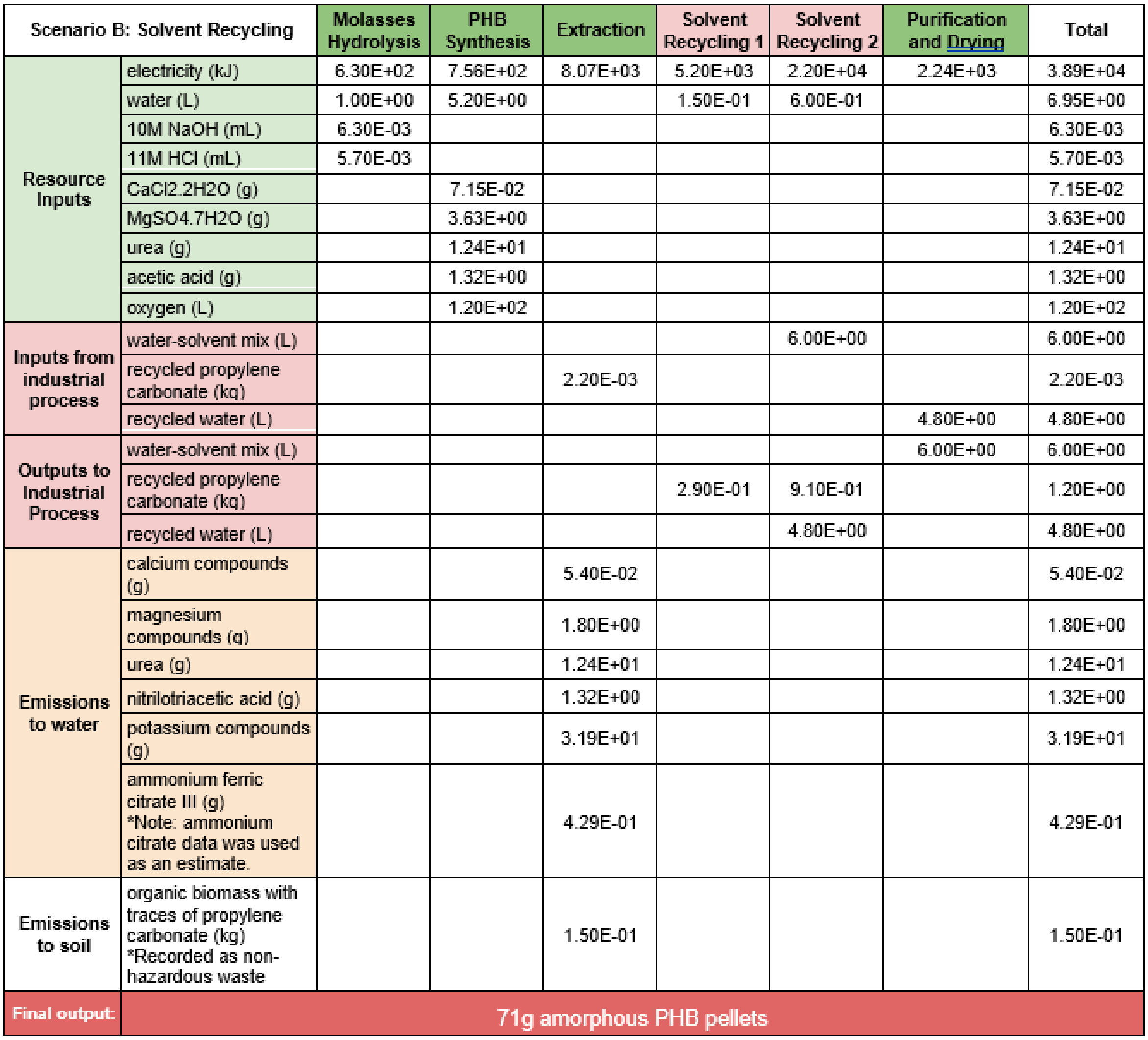
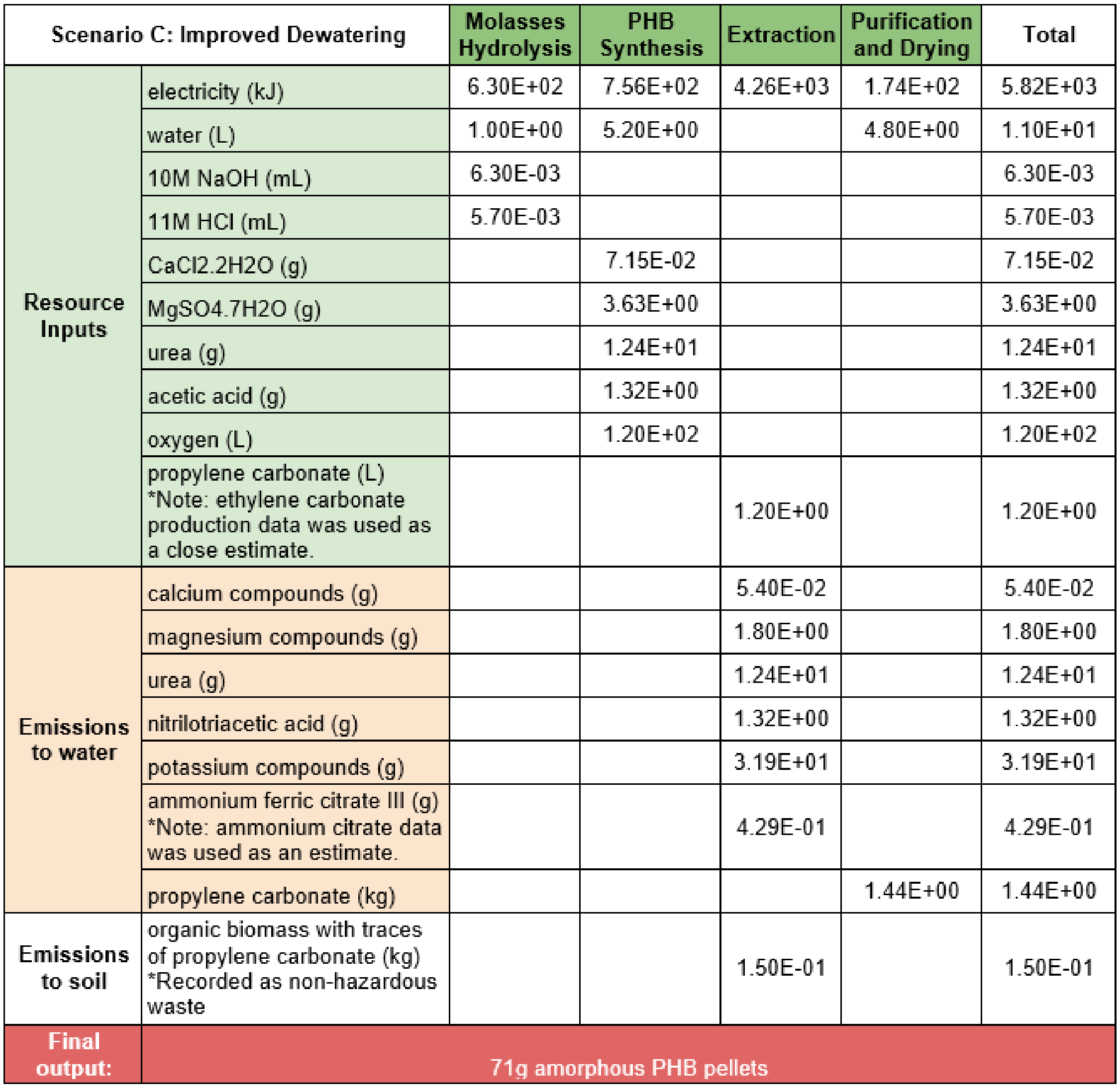


Scenario B: Solvent Recycling		Molasses Hydrolysis	PHB Synthesis	Extraction	Solvent Recycling 1	Solvent Recycling 2	Purification and Drying	Total
Resource Inputs	electricity (kJ)	6.30E+02	7.56E+02	8.07E+03	5.20E+03	2.20E+04	2.24E+03	3.89E+04
	water (L)	1.00E+00	5.20E+00		1.50E-01	6.00E-01		6.95E+00
	10M NaOH (mL)	6.30E-03						6.30E-03
	11M HCl (mL)	5.70E-03						5.70E-03
	CaCl2.2H2O (g)		7.15E-02					7.15E-02
	MgSO4.7H2O (g)		3.63E+00					3.63E+00
	urea (g)		1.24E+01					1.24E+01
	acetic acid (g)		1.32E+00					1.32E+00
	oxygen (L)		1.20E+02					1.20E+02
Inputs from industrial process	water-solvent mix (L)					6.00E+00		6.00E+00
	recycled propylene carbonate (kg)			2.20E-03				2.20E-03
	recycled water (L)						4.80E+00	4.80E+00
Outputs to Industrial Process	water-solvent mix (L)						6.00E+00	6.00E+00
	recycled propylene carbonate (kg)				2.90E-01	9.10E-01		1.20E+00
	recycled water (L)					4.80E+00		4.80E+00
Emissions to water	calcium compounds (g)			5.40E-02				5.40E-02
	magnesium compounds (g)			1.80E+00				1.80E+00
	urea (g)			1.24E+01				1.24E+01
	nitrilotriacetic acid (g)			1.32E+00				1.32E+00
	potassium compounds (g)			3.19E+01				3.19E+01
	ammonium ferric citrate III (g) *Note: ammonium citrate data was used as an estimate.			4.29E-01				4.29E-01
Emissions to soil	organic biomass with traces of propylene carbonate (kg) *Recorded as non-hazardous waste			1.50E-01				1.50E-01
Final output:	71g amorphous PHB pellets							

Table S2. Life cycle inventory of PHB bioplastic production, Scenario B. The material inputs and outputs of producing 71 grams of amorphous PHB pellets through production scenario B (propylene carbonate solvent recycling), including quantities and units. In the column headers, solvent recycling 1 involves the separation of solvent and waste biomass through stirred-tank cell rinsing, filtration, and distillation, while solvent recycling 2 involves the separation of solvent and water through distillation.

https://doi.org/10.59720/24-107



Scenario C: Improved Dewatering		Molasses Hydrolysis	PHB Synthesis	Extraction	Purification and Drying	Total
Resource Inputs	electricity (kJ)	6.30E+02	7.56E+02	4.26E+03	1.74E+02	5.82E+03
	water (L)	1.00E+00	5.20E+00		4.80E+00	1.10E+01
	10M NaOH (mL)	6.30E-03				6.30E-03
	11M HCl (mL)	5.70E-03				5.70E-03
	CaCl2.2H2O (g)		7.15E-02			7.15E-02
	MgSO4.7H2O (g)		3.63E+00			3.63E+00
	urea (g)		1.24E+01			1.24E+01
	acetic acid (g)		1.32E+00			1.32E+00
	oxygen (L)		1.20E+02			1.20E+02
	propylene carbonate (L) *Note: ethylene carbonate production data was used as a close estimate.			1.20E+00		1.20E+00
Emissions to water	calcium compounds (g)			5.40E-02		5.40E-02
	magnesium compounds (g)			1.80E+00		1.80E+00
	urea (g)			1.24E+01		1.24E+01
	nitrilotriacetic acid (g)			1.32E+00		1.32E+00
	potassium compounds (g)			3.19E+01		3.19E+01
	ammonium ferric citrate III (g) *Note: ammonium citrate data was used as an estimate.			4.29E-01		4.29E-01
	propylene carbonate (kg)				1.44E+00	1.44E+00
Emissions to soil	organic biomass with traces of propylene carbonate (kg) *Recorded as non-hazardous waste			1.50E-01		1.50E-01
Final output:	71g amorphous PHB pellets					

Table S3. Life cycle inventory of PHB bioplastic production, Scenario C. The material inputs and outputs of producing 71 grams of amorphous PHB pellets through production scenario C (replacing filter presses with dewatering centrifuges), including quantities and units.

Parameter	Unit	Value
Type	-	Fed-batch with molasses pulses
Initial cell concentration	g/L	0.4
Initial TRS concentration	g/L	20
Temperature	°C	35
pH	-	7.00
Cultivation time	h	24
Final cell concentration	g/L	20.9
Final PHB cell content	%	56
Final PHB yield	g/L	11.8
Average agitation	rpm	680
Estimated aeration	vvm	0.125
Residence Time	hours	24
T_R	°C	35

Table S4. PHB accumulation setup conditions. Temperature, time, and concentration conditions during the cultivation of *C. necator* bacteria to accumulate PHB, according to methods used by Dalsasso et al. (18).

Parameter	Unit	Value	Reference
Type	-	Fed-batch	(Dalsasso et al., 2019)
Material	-	Reinforced concrete (1% of steel) coupled with polyurethane foam	(Piccinno et al., 2016)
Reactor Volume	L	100	(Piccino et al., 2016)
c_p (mixture at T_R)	J/(kg °C)	3080	(Olbrich, 1963)
s	m	0.20 concrete + 0.05 polyurethane	(Piccinno et al., 2016)
η_{heat}	-	0.72	(Piccinno et al., 2016)
A	m^2	1.271 (for 100L cylindrical reactor)	(Piccinno et al., 2016)
N_p	-	0.79	(Piccinno et al., 2016)
N	1/s	3.052	(Piccinno et al., 2016)
d	m	0.173	(Piccinno et al., 2016)

Table S5. PHB accumulation reactor parameters. Physical properties and numerical constants of the PHB accumulation bioreactor, according to methods and data from Dalsasso et al. Piccinno et al., and Olbrich (18, 22, 28).

Parameter	Unit	Value	Reference
Type	-	CSTR	
Material	-	Stainless steel coupled with polyurethane foam	(Piccinno et al., 2016)
V	L	100	(Dalsasso et al., 2019)
Resident time	min	45 (pretreatment), 5 (extraction)	(Dalsasso et al., 2019)
T_R	°C	95 (pretreatment), 150 (extraction)	(Dalsasso et al., 2019)
c_p (mixture at T_R)	J/(kg °C)	3080 (pretreatment), 1640 (extraction)	(Olbrich, 1963)
s	m	0.05 steel + 0.01 polyurethane	(Piccinno et al., 2016)
η_{heat}	-	0.72	(Piccinno et al., 2016)
d	m	0.67	(Piccinno et al., 2016)
N	1/s	0.79	(Piccinno et al., 2016)

Table S6. PHA extraction reactor parameters. Physical properties and numerical constants of the PHB extraction reactor, used in pretreatment heating and solvent extraction steps, according to methods and data from Dalsasso et al. Piccinno et al., and Olbrich (18, 22, 28).

Parameter	Unit	Value	Reference
V	L	100	(Dalsasso et al., 2019)
c_p (mixture at T_R)	J/(kg °C)	3700	(Olbrich, 1963)
s	m	0.05 steel + 0.01 polyurethane	(Piccinno et al., 2016)
η_{heat}	-	0.72	(Piccinno et al., 2016)
d	m	0.67	(Piccinno et al., 2016)
N	1/s	0.79	(Piccinno et al., 2016)

Table S7. Distillation column parameters. Physical properties and numerical constants of the distillation column used for solvent recycling in Scenario B, according to methods and data from Dalsasso et al. Piccinno et al., and Olbrich (18, 22, 28).