



## Article

# Life Cycle Assessment of Exopolysaccharides and Phycocyanin Production with *Arthrospira platensis*

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**Abstract:** In the pursuit of sustainable solutions for contemporary environmental challenges arising from the increasing global demand for energy, this study delves into the potential of cyanobacteria, specifically *Arthrospira platensis* (commonly known as “spirulina”), as a versatile resource. Employing a life cycle assessment (LCA) in accordance with the ISO 14044:2006 standard and employing both midpoint and endpoint indicators, the study comprehensively evaluates environmental impacts. The research explored a range of scenarios, specifically investigating variations in light intensity and harvesting volume. These investigations were carried out using a pilot-scale photobioreactor, specifically an airlift reactor system featuring a horizontal tubular downcomer. The primary focus is on extracting valuable compounds, namely exopolysaccharides and phycocyanin. It emphasized the extraction of value-added products and strategic integration with a biogas plant for process heat, contributing to developing a sustainable supply network and offering insights into environmentally conscious algae cultivation practices with implications for renewable energy and the production of valuable products. The results emphasize the project’s potential economic feasibility with minimal energy impact from by-product extraction. The environmental assessment identifies marine ecotoxicity and fossil resource depletion as principal impacts, predominantly influenced by upstreaming and harvesting stages. After conducting comparisons across various scenarios, it was found that cultivations under higher light intensities have a lower environmental impact than cultivations with low light supply. However, regardless of light intensity, processes with shorter harvesting cycles tend to have a smaller environmental impact compared to processes with longer harvesting cycles. Overall, this research contributes a nuanced and realistic perspective, fostering informed decision-making in sustainable algae cultivation practices, with implications for renewable energy and valuable compound production.



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## 1. Introduction

In the quest for sustainable solutions to contemporary environmental challenges, the potential of algae as a multifaceted resource has garnered increasing attention. The escalating global demand for energy and chemicals, driven by population growth and development, underscores the need to explore alternative resources that can be accomplished by the potential of microalgae and cyanobacteria as a promising solution [1,2]. Notably, microalgae and cyanobacteria offer the potential to efficiently produce biomass for various applications, including food, feed, fuels, and chemicals, while addressing challenges in wastewater treatment and carbon capture [3]. The economic significance of algae in marine

biotechnology, coupled with their higher energy yields and faster photosynthesis, has attracted attention from economists [4,5]. Cyanobacterium, particularly *Arthrospira platensis*, known as spirulina, stands out for its diverse applications in health, cosmetics, nutrition, and bioremediation [6,7]. Despite the immense potential of microalgae and cyanobacteria, the commercialization of related technologies faces a significant hurdle: the high cost of microalgal biomass production, emphasizing the importance of careful species selection and cultivation practices [8].

Moreover, the utilization of algae in producing high-value compounds presents an avenue for climate change mitigation, reducing reliance on traditional energy-intensive production methods [9,10]. Algae-based biofuels hold promise in reducing greenhouse gas emissions and contributing to renewable energy alternatives [11–13]. Onyeaka et al. (2021) highlight the carbon sequestration potential of algae, emphasizing their role in carbon capture and storage initiatives [10].

In this respect, this study delves into the development of a comprehensive life cycle analysis (LCA) framework to produce antiviral exopolysaccharides (EPS) and phycocyanin (PC) with *A. platensis* (spirulina).

Phycocyanin, renowned as the most valuable component of *A. platensis*, stands out as a valuable blue pigment [14]. Regarding exopolysaccharides, studies research underscores the potential antiviral efficacy of EPS in preventing KHV infection in carp [15–17]. The cultivation growth occurred in a pilot-scale photobioreactor (PBR).

Prompted by the Antiviral Substances and Pigments project, supported by the Agency for Renewable Resources (Federal Ministry of Food and Agriculture of Germany), the primary objective is to contribute to the growing field of algae extraction and residue biomass valorization. The LCA will serve as a baseline structure applicable to diverse processes, technologies, and end products, aiming to improve the environmental performance of the project and facilitate widespread replication and scalability.

Hence, it holds significance to assess and gain a comprehensive understanding of each stage involved in a process. Implementing the LCA is a fitting method to analyze product impacts throughout their life cycles. The ISO 14044/14040 standard guides LCA analysis, emphasizing precise definition of process boundaries [18]. This involves delineating limits, which can range from cradle to grave, covering the entire production system cycle, or gate to gate, focusing solely on manufacturing. Adhering to these standards, the study evaluates cradle-to-gate boundaries, from cyanobacteria supply to value-added product production at the laboratory's gate.

Furthermore, this research integrates environmental impact methodologies to aid in interpreting LCA studies. It achieves this by translating system emissions and resource extractions into concise environmental impact scores [19]. This is done using characterization factors (CFs), which measure the environmental impact per unit of stressor, such as per kilogram of resource used or emission released. These factors are crucial for impact assessment [20]. Two primary approaches, midpoint and endpoint, are employed in this process. Midpoint factors are strategically positioned along the impact pathway, establishing robust connections to environmental flows and minimizing uncertainty [21]. For instance, in the water consumption category, it represents the number of cubic meters of water consumed per cubic meter of water extracted, reflecting relative water loss through evaporation or incorporation in products [20]. Conversely, endpoint factors cover human health, ecosystem quality, and resource scarcity, offering valuable insights into environmental relevance with higher uncertainty. Using the water consumption example, the endpoint perspective quantifies damage to human health based on malnutrition potentially caused by water scarcity or vulnerability to water shortages.

Together, these two approaches complement each other in the comprehensive assessment of environmental impacts [20]. Given the diversity of methodologies, the models will vary in terms of the substances considered in the calculations and the characterization factors. These factors can significantly influence the choice of life cycle impact assessment (LCIA) methodology, making it an important decision point [22].

Based on prevailing literature, a predominant focus has been observed in LCA studies, with the majority concentrating solely on reporting global warming potential as part of the climate change impact category [23,24]. In contrast, our present study takes a different approach by selecting indicators relevant to the specific system under analysis. For the midpoint category, we consider marine eutrophication, climate change aspects, and cumulative energy demand associated with non-renewable resources. Some of these classifications have been used in other microalgae biomass studies [25,26]. Meanwhile, within the endpoint category, our focus extends to water consumption and its impact on aquatic ecosystems and human health. These chosen environmental impacts result from the applied methodology and normalization process, enabling the identification of the most significant impacts by converting diverse units into single scores [22,27].

In this context, specific impact categories such as abiotic depletion potential, acidification, freshwater and terrestrial ecotoxicity, and human toxicity may not be considered pertinent for the present study. The investigation of microalgae reveals that some of these categories are not representative [28]. This decision could be influenced by specific processes and materials flows of system that render these impact categories less significant or negligible. Despite being part of established methodologies, their exclusion may stem from a focus on more critical or pertinent environmental aspects within the context of the study, allowing for a streamlined and targeted LCA performance. The LCIA result will generate a profile offering valuable insights into environmental aspects, highlighting both favorable (potentials) and unfavorable (hot spots) performance within a product's life cycle.

In the pursuit of a holistic understanding, the research factors in realistic environments are subject to seasonal variations. These variations encompass diverse elements, including fluctuations in temperature and sunlight intensity. To elucidate the nature of real-life scenarios, the study introduces extreme scenarios, depicting high sunlight conditions akin to sunny days and low light scenarios indicative of overcast conditions or environments in different geographic locations. Incorporating such boundaries strives to provide a comprehensive understanding of the impact of varying environmental conditions on algae cultivation, extending to different harvesting schedules.

Therefore, this study aims to explore two fundamental factors. Firstly, the impact of different harvesting cycles on productivity and on the environment is examined. Secondly, an assessment is conducted on how variations in light supply quality, influenced by factors such as weather and location, impact productivity and environmental consequences. Furthermore, this line intends to encompass a broad spectrum of outcomes and gain insights into how diverse environmental conditions may shape results.

This nuanced approach contributes to a thorough evaluation of the environmental impact, and it helps establish clear boundaries and constraints for the analysis, empowering informed decision-making in sustainable algae cultivation practice.

Furthermore, the study seeks to evaluate the potential independence of LCA outcomes across different harvesting cycles in varied light scenarios. If confirmed, this observation could extend beyond countries with high sunlight intensity, such as Spain, to those in diverse conditions, including Denmark. This assumption implies that statistical annual light intensity is likely to align with the spectrum defined by the study's optimal and suboptimal scenarios. This discourse establishes the foundation for a nuanced exploration of the intricate interplay between environmental conditions, harvesting cycles, and LCA outcomes in the subsequent sections of the research.

## 2. Materials and Methods

### 2.1. Life Cycle Assessment of Exopolysaccharide and Phycocyanin Extraction

For the LCA design, the present work relied on the International Standards Organization (ISO) 14000 series [18,29]. Therefore, the open-source software OpenLCA (Version 1.11, Mozilla Public Licence 2.0) was chosen. It specializes in this study area and was developed by Green Delta, an independent consulting and sustainability software company. In addition, the choice of OpenLCA took into account the fact that it is free (open-source

software) and allows modeling life cycle systems, with the ability to calculate environmental, social, and economic indicators, with plugins that provide different specific elements, and with an open system that facilitates the import and export of data and the integration of other data [30].

The LCA modeling adhered to the ISO 14044:2006 standard, which outlines the framework for conducting a comprehensive LCA. This standard underscores the importance of addressing the following key stages: 1. defining goal and scope; 2. compiling a life cycle inventory; 3. conducting a life cycle impact assessment; and 4. interpretation [18]. ISO 14044/2006 also encompasses the life cycle inventory (LCI), which addresses the process of gathering data. In the aforementioned step, data collection and input–output flow accounting are underpinned by pilot-scale laboratory data acquired within the Antiviral Substances and Pigments project, supported by the Agency for Renewable Resources (Federal Ministry of Food and Agriculture of Germany).

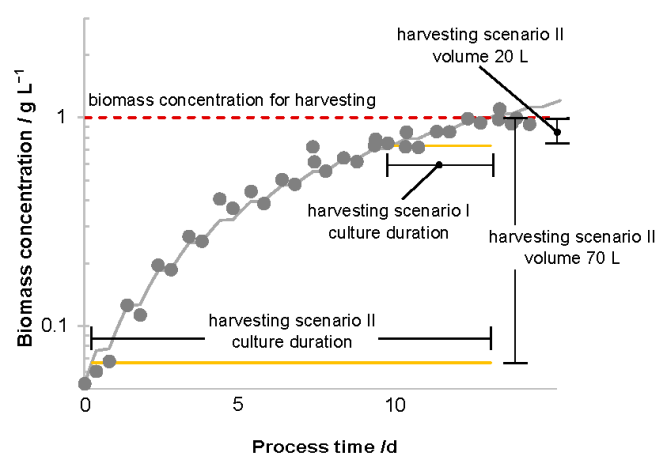
## 2.2. Goal and Scope

The purpose of this work is to develop a framework for an LCA of the extraction of EPS and PC from *A. platensis* (spirulina) that will further contribute to the mentioned research, as well as be further implemented in upcoming projects endeavors. Given this context, the focus of this research aims to evaluate the overall environmental impacts by comparing two different scenarios based on light intensity (low at  $100 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ , and high at  $450 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ ) and harvesting volume (20 L and 70 L). Owing to this, for the sake of meaningful comparisons, the study's key parameters will vary mainly in the number of harvesting days per week and the initial water volume introduced into the system, among other factors that will undergo specific modifications across nearly all steps. These scenarios are computed to consider two functional units: (1) 1 kg for the extraction of EPS and (2) 1 kg of PC.

Furthermore, to develop a sustainable supply network by integrating recycling and efficient technologies into production processes, a unique feature of the project is the linking of the value chain to a biogas plant. The strategic connection of the cascade with a biogas facility enables algae culture to be supplied with process heat.

## Scenarios and Parameters

Figure 1 depicts growth behavior within the photobioreactor under examination over time for a culture cultivated under low light intensities. Furthermore, Figure 1 shows the different harvesting scenarios, which differ in the amount of harvesting volume and therefore in the process duration for the repeated batches.



**Figure 1.** *A. platensis* growth behavior in a PBR reactor. Empirical data points (●) illustrate the growth of *A. platensis*. The dashed red line (—) indicates the harvesting biomass concentration. The yellow solid line (—) indicates starting biomass concentration and duration of scenario I and II. The solid gray line (—) represents the simulation results using the method of Jung et al. [31].

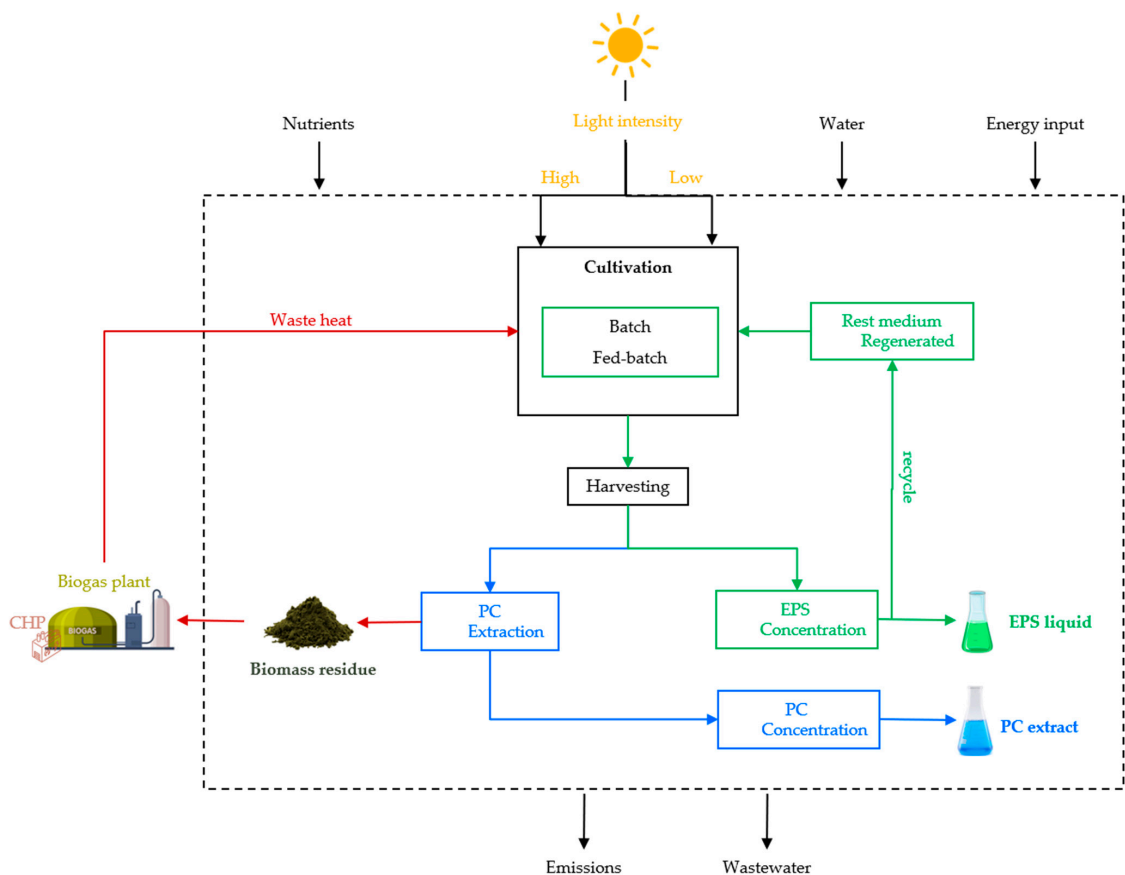
The harvesting point, denoted by the horizontal dotted red lines, is chosen at the early stationary phase when the biomass concentration reaches approximately  $1 \text{ g}\cdot\text{L}^{-1}$ . The processes are operated so that the harvesting point is always at a specific state with known quantities of the high-value products. This study examines four different scenarios (see Table 1). In Scenario II, the process time needed to reach the harvesting biomass concentration is calculated, based on experimental data: 16 days for low light intensities and 6 days for high light intensities. The harvesting volume (70 L) in Scenario II is significantly higher than in Scenario I (20 L), reflecting the need for more extensive harvesting due to the extended cultivation time.

**Table 1.** Critical parameters from harvesting process step employed to analyze scenarios I and II.

Light intensity <sup>1</sup> ( $I_0$ )	Scenarios: I/II	
	Process Time (d)	Harvesting Volume (L)
Low ( $I_0$ )	4/16	20/70
High ( $I_0$ )	2/6	20/70

<sup>1</sup> Low light intensity accounts for  $100 \mu\text{mol m}^{-2}\text{s}^{-1}$ , while high light intensity corresponds to  $450 \mu\text{mol m}^{-2}\text{s}^{-1}$ .

The process boundaries for this study encompass a cradle-to-gate approach (Figure 2), commencing with the cultivation process and ending with the extraction of the two high-value-added products. Logistics and transportation within the raw material supply chain are not considered.



**Figure 2.** System boundary of *A. platensis* cultivation for EPS and PC production and energy generation. The solid green line (—) delineates the procedural pathways integral to the liquid EPS production process. Concurrently, the solid blue line (—) elucidates the extraction routes for the vibrant blue pigment, phycocyanin. Lastly, the red line (—) illustrates the pathway for harnessing energy from the biomass residue.



Upstream processing consists of preparing the culture media, sterilization of the reactor and media, and the cultivation in a laboratory horizontal tubular airlift photobioreactor (PBR). The procedure was meticulously upheld and controlled to adhere to predetermined growth conditions studied previously [32]. The initial stage of downstream processing involves the separation of biomass from the supernatant. Subsequently, the culture is harvested via centrifugation, resulting in the generation of two distinct outputs: a liquid supernatant and biomass. From this step, we will embark on two distinct routes with the dual objectives of enhancing the value of the products and effectively treating the valuable waste, which will be recycled and reintegrated into the system.

The first route focuses on concentrating the supernatant through dynamic cross-flow filtration (DCF), resulting in concentrated EPS. Subsequently, the residual liquid undergoes permselective dialysis to regenerate the medium's carbon source through pH reduction, thereby shifting the equilibrium from the hydrogen carbonate formed during the cultivation process to the consumable bicarbonate. Through this process, the remaining media compounds and water can be reused after cultivation by adding the lacking nutrients. This approach promotes sustainability by efficiently recycling the treated media within the system, further enhancing resource efficiency.

In parallel, the second route extracts valuable compounds from the wet biomass using a pulsed electric field (PEF) to release diluted PC, which is further concentrated through DCF. The residual biomass, acquired postextraction, is used within a biogas CHP plant. The waste heat of this process can be further used to maintain the culture at optimum temperature, thus efficiently closing the process cycle and ensuring a sustainable value chain.

The process system and LCA study will adhere to specific system parameters and boundaries, as follows.

- The balancing comprises material and energy balances of the cultivation and downstream processing.
- The "cradle" stage starts with the cultivation in batch and fed-batch culture, which will be running on a pilot-scale photobioreactor.
- Both high-value products are concentrated using DCF methodology.
- The supply processes are beyond this work's scope (cutoff).
- Main energy inputs by the processes will be included throughout the life cycle.
- Waste heat from the combined heat and power (CHP) unit is used for heating the cultivation process.
- Heating during the cultivation process is not counted toward the LCA.

### 2.3. Process Description

#### 2.3.1. *A. platensis* Cultivation and Photobioreactor Design

To facilitate growth of phototrophic microorganisms, it is essential to provide them with a suitable environment, which includes various factors that enable their growth and reproduction, such as light energy, temperature, pH levels, and nutrient availability. These considerations led to the customization of both the cultivation process and photobioreactor design, focusing on establishing process conditions for optimal growth. The central emphasis in these endeavors is twofold: enhancing the economic efficiency of *A. platensis* cultivation while concurrently maintaining the superior quality of the extracted products and advancing sustainability efforts.

The cultivation of *A. platensis* was conducted in Zarrouk's nutrient-rich media (SOT) [33] in a 75 L horizontal tubular airlift photobioreactor.

The closed reactor system consists of horizontally arranged glass tubing ( $\varnothing = 5$  cm,  $V = 29.62$  L) connected by U-shaped bends and a vertical section comprised of two glass cylinders ( $\varnothing_1 = 15$  cm,  $V_1 = 21.2$  L;  $\varnothing_2 = 30$  cm,  $V_2 = 24.18$  L). The aerated ( $\dot{V} = 50$  L $\cdot$ min $^{-1}$ ) vertical cylinders function as the riser of the airlift reactor, while the horizontal tubing serves as the downcomer. According to Torzillo and Zittelli, the selection of smaller diameters

for the vertical tubes is predicated upon the objective of optimizing light penetration and supply to the cultured organisms [34].

This inceptive setting serves as the initial data collection point for quantifying the material and energy consumption essential to operate the bioreactor. The experiments were carried out primarily in batches, followed by repeated-batch mode, with liquid volume of 75 L culture broth and an annual operating time of 365 days/year. While acknowledging that this perspective may deviate from real-world scenarios, the exclusion of temporary shutdowns for maintenance or cleaning is made with the assurance that it will not impact the ultimate outcomes.

### 2.3.2. Batch and Fed-Batch Mode

*A. platensis* requires essential nutrients for growth. To meet this condition, the inoculum was prepared using a nutrient-rich complex in a 7 L volume of SOT cultivation medium. The pH was controlled to maintain an alkalinity level of approximately 9 and the medium consisted of the following components (in  $\text{g}\cdot\text{L}^{-1}$ ):  $\text{NaHCO}_3$ , 16.8;  $\text{K}_2\text{HPO}_4$ , 0.5;  $\text{NaNO}_3$ , 2.5;  $\text{K}_2\text{SO}_4$  and  $\text{NaCl}$  1.00;  $\text{MgSO}_4\cdot 7\text{H}_2\text{O}$ , 0.20;  $\text{CaCl}_2\cdot 2\text{H}_2\text{O}$ , 0.04;  $\text{FeSO}_4\cdot 7\text{H}_2\text{O}$ , 0.01;  $\text{Na}_2\text{EDTA}\cdot 2\text{H}_2\text{O}$ , 0.08; water 6993 mL and A5 micro nutrients solution at 7 mL. The A5 composition comprises  $2.86\text{ g}\cdot\text{L}^{-1}$  of  $\text{H}_3\text{BO}_3$ ,  $2.5\text{ g}\cdot\text{L}^{-1}$  of  $\text{MnSO}_4\cdot 7\text{H}_2\text{O}$ ,  $0.222\text{ g}\cdot\text{L}^{-1}$  of  $\text{ZnSO}_4\cdot 7\text{H}_2\text{O}$ ,  $0.079\text{ g}\cdot\text{L}^{-1}$  of  $\text{CuSO}_4\cdot 5\text{H}_2\text{O}$  and  $0.021\text{ g}\cdot\text{L}^{-1}$  of  $\text{Na}_2\text{MoO}_4\cdot 2\text{H}_2\text{O}$ . This nutrient-rich medium serves as the essential foundation for the growth of *A. platensis* [33,35].

In addition, another fundamental determinant for fostering growth lies in the temperature and amount of supplied light. Therefore, as the project centers around outdoor cultivation, the assessment of light energy consumption is not included in LCA. The reactor temperature is regulated at a range of 30–35 °C. The exhaust air from the reactor is condensed by a cooling device (Batch circulator 426–1642, Thermo Scientific Haake, Schwerte, Germany) set to 20° [36]. At this stage, no additional heat source is required, as the biogas plant's reactor will supply a share of the CHP. Energy consumption is attributed to the operation of both cooler units and the compressor that powers the PBR airlift system. In this instance, the figures are based on two harvesting scenarios: a 20 L volume (Scenario I) and a 70 L volume (Scenario II). These variables fluctuate depending on the high and low light intensity levels, and as a result the number of harvesting days.

In fed-batch mode, 3.5 L of water is added, along with chemicals at half the concentration of nutrients used previously. Thereafter, regenerated media retrieved from the permselective dialysis outlet after the EPS concentration step are supplied to the feed stream. This will occur upon the completion of the initial process cycle, thereby ensuring the systematic recovery of resources and the establishment of sustainable process chain closure. The total electricity consumption consists of the combined energy usage of the 24/7 operation of the compressor and chiller over the course of 365 days, subtracted from the working days dedicated to batch mode.

### 2.3.3. Harvesting

A key aspect of this process step is to assess the appropriate technology for harvesting microalgae and cyanobacteria while attempting to ensure that a large volume of culture medium can be separated in a short period of time. Additionally, it is essential to underscore the significance of minimizing both cost and energy consumption to the greatest extent possible. These factors lead this process to a particularly significant degree of importance—in terms of parameter dependence—since almost all the variables are linked to the harvesting duration in days and the volume needed for harvesting. Additionally, it must be kept in mind that the highest product yield of both aimed-on products is in the stationary phase [32].

For the evaluation of both scenarios I and II, harvesting cycles were scheduled according to Table 1, which is dependent on the chosen volume, harvesting duration, and light intensity specified for each case. The culture broth was centrifuged (FJ 130 ERR Longlife, Janschitz GmbH, Althofen, Austria). The centrifuge's operational time per cycle quantified

at 105 kWh, considers both the duration of machine operation and the variable cleaning and washing time, which is contingent upon the volume of harvested material. The harvested output, as explained previously, will be directed along two distinct routes, each following specific treatment processes to maximize its full potential. In this regard, the remaining valuable stream, namely, the supernatant, undergoes filtration and concentration through DCF, while the biomass proceeds to the extraction process using PEF technology.

### 2.3.4. Exopolysaccharide Concentration

After the biomass is separated from the liquid during harvesting, the supernatant is further processed via DCF (CDR-01-152 SS, Novoflow GmbH, Rain, Germany) with a membrane pore size of 5 nm. The processing time depends on the harvesting volume, ranging from 3.3 h (20 L) to 4.05 h (70 L).

Using DCF for harvesting, EPS was described as a promising technology for concentration of EPS [37]. When compared with alternative mechanical and chemical techniques, its primary strengths lie in its remarkable energy efficiency, ranging from 0.38 to 2.06 kW·m<sup>-3</sup> for achieving 100–400 times concentration, as well as the cost-effectiveness related with initial pump outlay and the sporadic need for membrane replacements [38].

Regarding the input flows, the sole supplementary source is deionized water for washing, consistently set at a fixed volume of 30 L across all scenarios. The first product obtained from this added-value process is the EPS, whose EPS yield is dependent on process-specific parameters [32] and scenarios, as shown in Table 2.

**Table 2.** EPS parameters.

Light Intensity (I <sub>0</sub> )	Scenarios: I/II	
	EPS Unconcentrated (g·L <sup>-1</sup> )	EPS Yield (g) <sup>2</sup>
Low (I <sub>0</sub> )	0.04	0.62/2.18
High (I <sub>0</sub> )	0.06	0.94/3.28

<sup>2</sup> Filtration efficiency rate of 80%.

Along with this outflow, the residual fluid from the process is directed towards membrane electrodialysis for additional treatment, which will be elaborated upon in the following section.

### 2.3.5. Regenerated Medium

Following the preceding process step (2.3.4), the residual medium is regenerated by using the permselective dialysis method. With this technique, hydrogen ion molecules are transferred from a highly concentrated HCl solution to the used medium side through a monovalent cation membrane without the additional anion that would be present when using the direct addition of inorganic acids for the pH shift. Therefore, the carbonate of the used medium can be shifted to bicarbonate. The quantity of bicarbonate metabolized during the previous cultivation cycle is required to be replenished for subsequent use.

The ratio between the HCl solution and the used medium is set at 2.50 in both scenarios. This calculation considers the baseline water quantity and a fixed 30 L water input volume. The machine’s operational duration remains fixed at 7 h and depends on the amount of water introduced into the system. Therefore, the operational duration is 3.85 h for 20 L and 15.23 h for 70 L, respectively.

In terms of chemical product incorporation, 10 g·L<sup>-1</sup> of medium compounds is added to the regenerated medium. Aligned with each scenario-specific condition, these concentrations were specifically denoted as 170 g for scenario I and 650 g for scenario II, calculated based on a single process cycle.

### 2.3.6. Phycocyanin Extraction

The second route from the harvesting outflow involves using Pulsed Electric Field (PEF) technology (HVP5, Elea Technology GmbH, Quakenbrück, Germany) for opening of



the cell wall to extract PC. This extraction method offers both energy and process-related benefits when compared to conventional mechanical cell disruption techniques. Moreover, this process allows for a selective extraction of intracellular products, thereby preventing any loss in quality and the subsequent need for fractionation of impure extracts [14,39,40]. Furthermore, research findings demonstrate that employing this cell disruption approach yielded a remarkable 90% increase in the purity of PC extraction compared to ball milling. Equally significant—thereby streamlining downstream processing—is the discovery that the fractions obtained through this method not only exhibited higher purity but also boasted a significantly reduced environmental footprint, approximately half that of the alternative technique [16,41–43].

In this step, the material flows are limited to water and ethanol. It is important to highlight that the energy consumption of the PEF accounts for all unit operations involved, such as a heater, a pump, and the PEF itself. The operating time of the procedure is 1 h, including preparation and cleaning. Lastly, the amount of PC extracted from the biomass (specific concentration of gram PC per gram biomass) is independent of the scenarios, but dependent on the applied light intensities (see Table 3). In the extraction process, the biomass loses 50% of its water-soluble compounds. The residual biomass outflow from the process will be harnessed in a biogas plant, contributing to heat and power production. Hence, the quantities of PC extracted and the biomass residue per cycle for scenarios I and II are displayed in Table 3.

**Table 3.** Relevant outputs acquired from the PC extraction process.

Light Intensity ( $I_0$ )	Scenarios: I/II	
	Biomass Residue (g)	PC Extract (g)
Low ( $I_0$ )	9.20/32.20	3.68/12.88
High ( $I_0$ )	11.80/41.30	4.72/16.52

The final step in obtaining concentrated PC employs the same technology as in the EPS concentration step (2.3.4), namely, dynamic cross-flow filtration. This process adheres to identical conditions and material flow parameters concerning both the input and output of 30 L of water and 1 h of operation time. However, instead of treating supernatant as input, the system receives diluted PC extracted from the prior process.

### 2.3.7. Biogas Plant

To develop a sustainable supply network by integrating recycling and efficient technologies into production processes, the present project includes a unique feature: the integration of the value chain with a biogas plant. Biogas plants are usually linked to CHP units, granting efficient operation and the provision of waste heat for various processes. This strategic cascade connection with a biogas facility provides process heat for algae culture.

Consequently, the biomass residue resulting from PC extraction contributes to the biogas plant’s energy supply, generating the highest methane figures of  $4.64 \text{ MJ}\cdot\text{a}^{-1}$  and  $5.41 \text{ MJ}\cdot\text{a}^{-1}$  in scenarios I and II, respectively, under high light intensity conditions. The preceding figures were obtained through experimental data, which yielded a 20% conversion rate for every 300 mL of gas [44]. The thermal energy released from the plant is then reintegrated into the cascade to feed the PBR production, effectively closing the process loop.

In summary, this integrated approach not only underscores the project’s commitment to sustainability but also positions it favorably to achieve significant economic gains, making it a promising venture with multifaceted potential outcomes.

### 2.4. Inventory Analysis

In the second phase of the LCA, the focus shifts to compile and precisely quantify the inputs and outputs related to the product throughout its entire life cycle within the project boundaries [18,29]. Therefore, data collection was thoroughly performed through on-site experiments conducted in the pilot-scale experiments. The inventory analysis was performed using ELCD database and Eco-invent to perform the LCIA. Furthermore, this study provides a supplementary spreadsheet with the parameters of the processes. Within this table are unlocked cells where variables can be entered, and the system will perform automatic calculations for either input or output values. This configuration provides a foundational framework that can be seamlessly applied to various processes, irrespective of differences in material flows, end products, or technologies. Table 4 summarizes the parameters selected for the sensitivity analysis. Here, the most relevant inputs (I) and outputs (O) are attributed to the cultivation, harvesting, and downstream processes. I/O represents both the material that functions as the input for a process and the resulting output.

**Table 4.** Parameters of the processes throughout the project’s life cycle.

Process Step	Technology	(I/O)	Material Flows [Unit]	Unit
Cultivation Batch mode	PBR	I	Batch operation time	$\text{h}\cdot\text{d}^{-1}$
		I	Cooler electricity	$\text{kWh}\cdot\text{d}^{-1}$
		I	Compressor electricity	$\text{kWh}\cdot\text{d}^{-1}$
Cultivation Fed-batch	PBR	I	Cooler electricity	$\text{kWh}\cdot\text{d}^{-1}$
		I	Compressor electricity	$\text{kWh}\cdot\text{d}^{-1}$
		I	Chemicals	
		I	Water	L
Harvesting	Centrifuge	I	Harvesting duration	d
		I	Harvesting volume	L
		I	Centrifuge electricity	$\text{kWh}\cdot\text{d}^{-1}$
		I	Centrifuge power	$\text{kWh}\cdot\text{d}^{-1}$
		I/O	Water	L
		O	Supernatant	L
EPS Concentration	Dynamics Cross Flow Filtration	O	Dried biomass	kg
		I/O	Water	L
		I	DCF electricity	$\text{kWh}\cdot\text{d}^{-1}$
		O	Wastewater	L
		O	EPS unconcentrated	$\text{g}\cdot\text{L}^{-1}$
Regenerate medium	Permselective dialysis	O	Rest medium	L
		I/O	Water	L
		I	Ratio—Used medium and HCl	No unit
		I	Chemicals	g
		I	HCl	g
PC Extraction	PEF & Dynamics Cross Flow Filtration	I	Permselective dialysis electricity	$\text{kWh}\cdot\text{d}^{-1}$
		I/O	Water PEF	L
		I	PEF electricity	$\text{kWh}\cdot\text{d}^{-1}$
		I	Pump electricity	$\text{kWh}\cdot\text{d}^{-1}$
		I	Heater electricity	$\text{kWh}\cdot\text{d}^{-1}$
		O	Biomass residue	g
Biogas Plant		I/O	Water Filtration	L
		I	DCF Electricity	W
		I	PC extract	g
		O	Methane	$\text{mL}\cdot\text{g}^{-1}$ biomass

Table 5 displays the inventory of the processes being investigated. The machinery’s electricity consumption is composed of power intake in watts (W) multiplied by operational time in hours and days.

**Table 5.** Inventory of the upstreaming, harvesting, and downstream processes.

Process Step	Technology	(I/O)	Material Flows [Unit a <sup>-1</sup> ]	Scenarios			
				Low I <sub>0</sub> Hv 20 L	Low I <sub>0</sub> Hv 70 L	High I <sub>0</sub> Hv 20 L	High I <sub>0</sub> Hv 70 L
Cultivation Batch mode	PBR	I	Media (g·L <sup>-1</sup> )	22.13	22.13	22.13	22.13
		I	Water (L)	75	75	75	75
		I	Electricity (kWh)	2310	2310	726	726
		I	Heat (MJ) <sup>3</sup>	--	--	--	--
Cultivation Fed batch	PBR	I	Rest medium regenerated (L)	1505	1488	3011	3969
		I	Water (L)	319.38	79.84	638.75	212.92
		I	Electricity (kWh)	21,780	21,780	23,364	23,364
		I	Heat (MJ) <sup>3</sup>	--	--	--	--
		I	Chemicals (kg)	20.2	17.7	40.4	47.11
Harvesting	Centrifuge	I	Culture broth (L)	20	70	20	70
		I/O	Water (L)	912.5	798.4	1825	2129
		I	Electricity (kWh)	9.58	8.38	19.16	22.35
		O	Dry biomass (kg)	1.68	1.47	4.31	5.02
		O	Supernatant (L)	1779	1556	3558	4151
EPS Concentration	Dynamic Cross-Flow Filtration	I	Supernatant (L)	1779	1556	3558	4151
		I	Electricity (kWh)	34.93	10.71	69.86	28.56
		I/O	Water (L)	2737	684.4	5475	1825
		O	EPS (g)	56.94	49.82	170.82	199.29
		O	Rest medium (L)	1505	1488	3011	3969
Regenerate medium	Permselective dialysis	I	Rest medium (L)	1505	1488	3011	3969
		I	Electricity (kWh)	60.07	59.39	120.15	158.38
		I	Water (L)	41.25	163.13	41.25	163.13
		I	HCl (kg)	155.58	153.81	311.16	410.17
		I	Chemicals (kg)	15.06	14.89	30.11	39.69
		O	Regenerated medium (L)	1505	1488	3011	3969
		O	Water—recycled (L)	41.25	163.13	41.25	163.13
PC Extraction	PEF and Dynamic Cross-Flow Filtration	I	Dry biomass (kg)	1.68	1.47	4.31	5.02
		I	Ethanol (L)	2.00	2.00	2.00	2.00
		I	Electricity PEF (kWh)	182.50	136.87	365.00	364.98
		I	Electricity DCF (kWh)	34.93	10.71	69.81	28.56
		I/O	Water PEF (L)	912.50	228.12	1825	608.33
		I/O	Water DCF (L)	2737	684.4	5475	1825
		O	Biomass residue (g)	839.50	734.56	2153	2512
		O	PC (g)	335.80	293.83	861.40	1004
Biogas Plant		I	Biomass residue (g)	839.50	734.56	2153	2512
		O	Biogas energy (MJ)	1.81	1.58	4.64	5.41

<sup>3</sup> The assessment of heat input in the cultivation process is considered for potential project configurations. However, the calculation thereof is beyond the scope of this project.

### 2.5. Life Cycle Impact Assessment (LCIA)

The environmental impact assessment in this study was carried out employing a range of approaches, such as ReCiPe 2016 Midpoint (H), IPCC (Intergovernmental Panel on Climate Change), and Cumulative Energy Demand (CEM). These methodologies are renowned for their capacity to comprehensively quantify and analyze the environmental impacts associated with diverse processes. The ReCiPe methodology, developed in 2008 through collaboration between RIVM, Radboud University Nijmegen, Leiden University, and Pré Consultants, remains instrumental in evaluating environmental implications and provides harmonized characterization factors at midpoint and endpoint levels [20].

Both midpoint and endpoint indicators were integrated to enhance the categorization of impacts. This approach was adopted to provide a more holistic assessment of the environmental performance of the product systems. It guarantees a well-rounded evaluation, providing insights into diverse sustainability aspects, ranging from resource consumption to potential impacts on ecosystems and human well-being. In essence, it delivers a holistic perspective on the overall consequences [20,21]. In this analysis, the outcomes are obtained

by aggregating elementary flows from all processes within the LCI model, considering a cause-effect chain impact category [21,45]

Hence, the process's contribution to potential impacts was assessed by selecting the most critical impacts for EPS and PC extraction. Therefore, the scope of the application using ReciPe Midpoint as a reference will measure the impact of water consumption ( $\text{m}^3$ ), and marine eutrophication ( $\text{kg N eq}$ ). When evaluating at the endpoint level (ReciPe Endpoint H), the influences are linked to their impact on human health due to global warming. This impact, quantified in terms of disability-adjusted life years (DALYs), becomes evident through examples such as increased disease susceptibility to malnutrition, malaria, and diarrhea, along with an elevated likelihood of experiencing more frequent and severe flooding events [21,46].

The long-term climate impact is assessed through GHG emissions, which are quantified using global warming potential (GWP) metrics. Achieved through the approach established by the IPCC, this metric, expressed in kilograms of  $\text{CO}_2$  equivalent ( $\text{CO}_2\text{-eq}\cdot\text{kg}^{-1}$ ), considers emissions of carbon dioxide ( $\text{CO}_2$ ), methane ( $\text{CH}_4$ ), and nitrous oxide ( $\text{N}_2\text{O}$ ) while accounting for their respective GWP values over a century-long timeframe. This specific time period was selected based on the guidance of the IPCC, as it provides a robust foundation for meaningful comparisons. Additionally, it is acknowledged as a benchmark metric within the realm of scientific consensus [22,47].

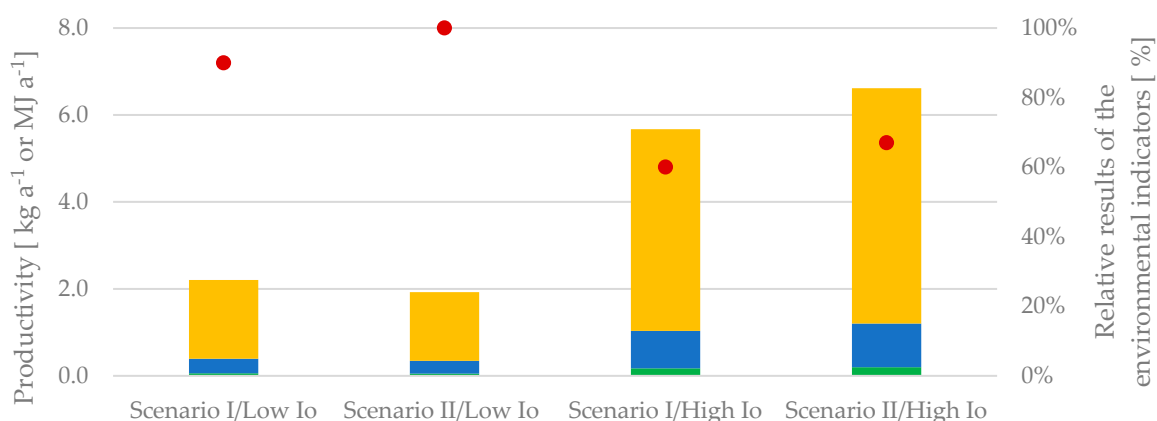
Lastly, the CEM method was applied to compute the energy consumption and the utilization of both natural and fossil resources measured in  $\text{MJ-eq}$  throughout the life cycle of the value-added product extraction process [48,49].

### 3. Results

The emissions and resource flows underwent mapping during the inventory analysis (2.4), aligning seamlessly with the specific impact methodologies (2.5), marking the culmination of the life cycle assessment step. The calculations for algae productivity, mass, energy, and water consumption were executed, accounting for the two scenarios. Subsequently, these data were transposed into the OpenLCA software, enabling the detailed assessment of environmental impacts.

Thus, the study findings unveil insights into the environmental impact of various circumstances. For this purpose, the light intensity variables are devised to establish boundary conditions, ensuring a representation of realistic environments that are subject to seasonal variations throughout the year. These variations encompass diverse factors such as temperature and sunlight intensity fluctuations. Consequently, the research presents extreme scenarios, with one depicting high sunlight conditions representing sunny days, while the low light scenario indicates overcast conditions or environments in different countries with distinct geographic locations. By incorporating such boundaries, the study provides a comprehensive understanding of the impact of varying environmental conditions on algae cultivation, offering insights that align with the dynamic nature of real-life scenarios.

Considering the entirety and proportional averages of environmental impact outcomes, the scenarios with the most significant impact are those characterized by receiving the least amount of sunlight (low  $I_0$ ) and the one operating the system with the highest volume of culture harvested (Scenario II), as seen in Figure 3. The most favorable situation is found in Scenario I under high light intensity, in both regards: achieving higher productivity when comparing conditions within the same scenario and minimizing environmental harm.



**Figure 3.** The efficiency of products generated from the system and their environmental impact contributions across diverse project variants. The primary *y*-axis (left) shows the productivity of EPS in kg a<sup>-1</sup> (■), phycocyanin in kg a<sup>-1</sup> (■), and methane in MJ a<sup>-1</sup> (■) under the different scenario settings. The red dots (●) on the secondary *y*-axis (right) illustrate the relative results of the environmental indicators. For each impact indicator, the maximum result is set to 100%, and the outcomes of the other variants are presented concerning this benchmark.

### 3.1. Environmental Impact Analysis for Exopolysaccharide Extraction

Overall, for producing 1 kg of EPS, the electricity demand of the system varies from 0.69 to 1.28 MJ a<sup>-1</sup>, wherein the energy consumption is assigned to the operation of the machinery. Within this process, the study highlights that the predominant portion of electricity usage occurs during upstream processes. Furthermore, the regenerated medium process, integrated into the system, involves the calculation of energy flow using negative values due to its recycling and loop closure.

The extraction process stands out as the major driver of water consumption, representing a share of 69.78%, with harvesting contributing 30.22%. Additionally, the fed-batch stage accounts for 6.97% of the total. The operation of dynamic cross-flow filtration, in this process step (2.3.4), demands a significant volume of water, leading to this result. The water impact unfolds similarly, with the extraction phase accounting for nearly the entirety of the environmental impact, succeeded by a minor contribution from the upstream process.

Table 6 displays the environmental impact results and the consistency in absolute deviation across various impacts. Regardless of the specific categories, a 12% deviation is consistently observed in all four scenarios.

**Table 6.** Sensitivity analysis for exopolysaccharide extraction.

LCIA Methodology	Impact Category	Abbr.	Units	Scenario I (20 L)		Scenario II (70 L)	
				Low I <sub>0</sub>	High I <sub>0</sub>	Low I <sub>0</sub>	High I <sub>0</sub>
ReCiPe 2016 Midpoint (H)	Marine eutrophication Water consumption	MEP	N-eq·kg <sup>-1</sup>	1.52 × 10 <sup>4</sup>	1.01 × 10 <sup>4</sup>	1.71 × 10 <sup>4</sup>	1.14 × 10 <sup>4</sup>
		FETP	m <sup>3</sup>	7.14 × 10 <sup>6</sup>	4.68 × 10 <sup>6</sup>	8.27 × 10 <sup>6</sup>	5.29 × 10 <sup>6</sup>
ReCiPe 2016 Endpoint	Water consumption, Aquatic ecosystems	WCP-aq	species a <sup>-1</sup>	4.27 × 10 <sup>-6</sup>	2.40 × 10 <sup>-6</sup>	3.67 × 10 <sup>-6</sup>	2.71 × 10 <sup>-6</sup>
	Water consumption, Human health <sup>4</sup>	WCP-hh	d <sup>-1</sup>	1.58 × 10 <sup>1</sup>	8.87 × 10 <sup>0</sup>	1.36 × 10 <sup>1</sup>	1.00 × 10 <sup>1</sup>



Table 6. Cont.

LCIA Methodology	Impact Category	Abbr.	Units	Scenario I (20 L)		Scenario II (70 L)	
				Low I <sub>0</sub>	High I <sub>0</sub>	Low I <sub>0</sub>	High I <sub>0</sub>
IPCC Climate change	Biogenic	GWP-b	CO <sub>2</sub> -eq·kg <sup>-1</sup>	1.73 × 10 <sup>7</sup>	1.01 × 10 <sup>7</sup>	1.53 × 10 <sup>7</sup>	1.14 × 10 <sup>7</sup>
	Fossil	GWP	CO <sub>2</sub> -eq·kg <sup>-1</sup>	2.04 × 10 <sup>8</sup>	1.20 × 10 <sup>8</sup>	1.81 × 10 <sup>8</sup>	1.36 × 10 <sup>8</sup>
Cumulative Energy Demand (CED)	Non-renewable resources (fossil)	NR-f	MJ	1.89 × 10 <sup>9</sup>	1.26 × 10 <sup>9</sup>	2.14 × 10 <sup>9</sup>	1.42 × 10 <sup>9</sup>
	Renewable resources <sup>5</sup>	RN-b	MJ	1.54 × 10 <sup>8</sup>	1.02 × 10 <sup>8</sup>	1.73 × 10 <sup>8</sup>	1.15 × 10 <sup>8</sup>

<sup>4</sup> Damage to human health calculated in disability-adjusted life years; <sup>5</sup> category that quantifies the environmental impact related to the consumption of renewable biomass resources within a particular process or product life cycle.

Despite the overarching tendency shown previously (Figure 3), which indicates that Scenario II causes a greater environmental impact, the analysis of EPS output (Figure 4) yields different results, specifically for the categories of water consumption related to aquatic ecosystems and human health, as well as for impacts associated to biogenic carbon in climate change and the GWP 100a of fossil fuels. These disparities highlight the complexity of the variables and system parameterization between harvesting volume and duration, light intensity, and its peculiarities within the entire extraction process.

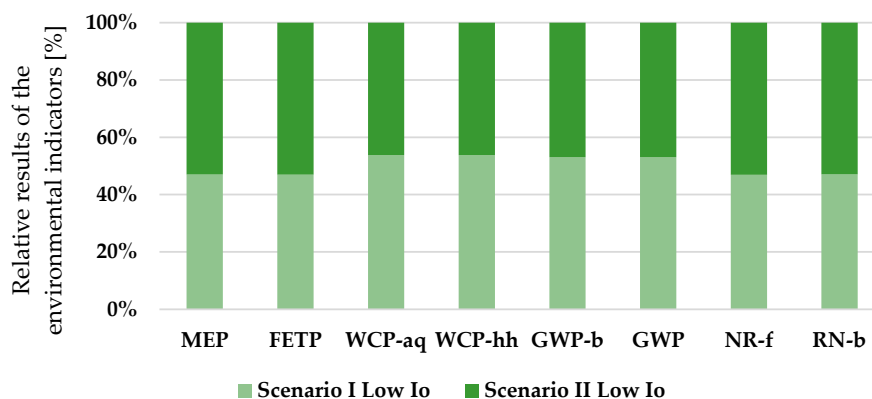


Figure 4. Comparative environmental impacts at low light intensity for Scenarios I and II, considering the functional unit of 1 kg EPS.

### 3.2. Environmental Impact Analysis for Phycocyanin Extraction

Environmental impact categories were likewise scrutinized in the assessment for the extraction of 1 kg of PC. The relative contributions of each life cycle scenario to ecological impacts are presented in Figure 5 and Table 7.

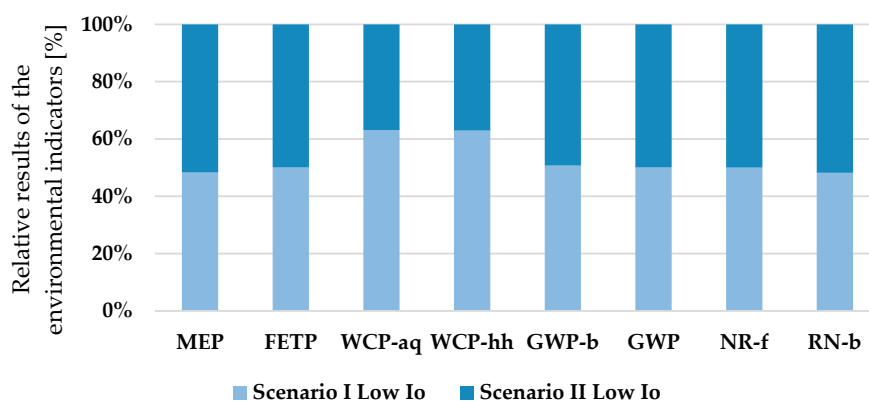


Figure 5. Comparative environmental impacts at low light intensity for Scenarios I and II, considering the functional unit of 1 kg PC.

**Table 7.** Sensitivity analysis for phycocyanin extraction.

LCIA Methodology	Impact Category	Abbr.	Units	Scenario I (20 L)		Scenario II (70 L)	
				Low I <sub>0</sub>	High I <sub>0</sub>	Low I <sub>0</sub>	High I <sub>0</sub>
ReCiPe 2016 Midpoint (H)	Marine eutrophication	MEP	N-eq·kg <sup>-1</sup>	5.32 × 10 <sup>3</sup>	2.12 × 10 <sup>6</sup>	5.69 × 10 <sup>3</sup>	2.11 × 10 <sup>6</sup>
	Water consumption	FETP	m <sup>3</sup>	6.67 × 10 <sup>5</sup>	5.01 × 10 <sup>5</sup>	4.63 × 10 <sup>5</sup>	3.36 × 10 <sup>5</sup>
ReCiPe 2016 Endpoint	Water consumption, Aquatic ecosystems	WCP-aq	species·yr	2.96 × 10 <sup>-7</sup>	2.19 × 10 <sup>-7</sup>	1.73 × 10 <sup>-7</sup>	1.20 × 10 <sup>-7</sup>
	Water consumption, Human health <sup>4</sup>	WCP-hh	DALY	1.10 × 10 <sup>0</sup>	8.17 × 10 <sup>-1</sup>	6.49 × 10 <sup>-1</sup>	4.52 × 10 <sup>-1</sup>
IPCC Climate change	Biogenic	GWP-b	CO <sub>2</sub> -eq·kg <sup>-1</sup>	2.44 × 10 <sup>6</sup>	1.01 × 10 <sup>7</sup>	2.37 × 10 <sup>6</sup>	1.14 × 10 <sup>7</sup>
	Fossil	GWP	CO <sub>2</sub> -eq·kg <sup>-1</sup>	3.09 × 10 <sup>7</sup>	1.20 × 10 <sup>8</sup>	3.08 × 10 <sup>7</sup>	1.36 × 10 <sup>8</sup>
Cumulative Energy Demand (CED)	Non-renewable resources (fossil)	NR-f	MJ	3.23 × 10 <sup>8</sup>	2.49 × 10 <sup>8</sup>	3.23 × 10 <sup>8</sup>	2.48 × 10 <sup>8</sup>
	Renewable resources <sup>5</sup>	RN-b	MJ	5.88 × 10 <sup>7</sup>	3.29 × 10 <sup>7</sup>	6.33 × 10 <sup>7</sup>	3.09 × 10 <sup>7</sup>

<sup>4</sup> Damage to human health calculated in disability-adjusted life years; <sup>5</sup> category that quantifies the environmental impact related to the consumption of renewable biomass resources within a particular process or product life cycle.

Despite the array of equipment utilized in PC extraction and concentration, the focal point of energy demand lies in the initial processes. To delve into specifics, within the batch and fed-batch steps, energy demands are primarily driven by the electricity required for algae culture circulation and the embedded energy within the cultivation process. This is warranted as the compressor and chiller operate daily throughout the year, showcasing a consistent pattern in EPS production.

The results show significant variations in the water consumption impact category, with Scenario II exhibiting a 36.2% reduction compared to Scenario I, both under low light intensity. Additionally, water-related impacts on aquatic ecosystems and human health represent more than 40% in deviation. This evidence points to an intensified harvesting schedule in the scenario where each cycle harvests 20 L—the total duration extends to approximately 91 d·a<sup>-1</sup>, while opting for 70 L per cycle reduces this timeframe to 23 d—leading to increased water demand, which is correlated with increased environmental repercussions.

In the realm of other impacts, the deviation remains consistent at 12%. Therefore, under high sunlight conditions, water consumption, marine eutrophication, and overall climate change impacts increase at the same rate in Scenario II. The environmental impact performance in high light intensity scenarios exhibits a similar correlation between deviations, albeit with elevated figures.

#### 4. Discussion

This environmental assessment reveals that scenarios with lower light intensity and higher water volume demonstrate greater environmental impacts, emphasizing the relationship between harvesting volume, duration, and environmental repercussions. Despite higher environmental impacts in Scenario II, a nuanced analysis of EPS and PC extraction shows variations, highlighting the complexity of system parameters. As per the findings by Wenzel et al. [50] study, the data quality can be classified as “very high” in terms of specificity, as they are either directly measured at the specific process site or accurately scaled from measurements. Henceforth, a systematic identification and quantification of material and energy flows took place for each of the eight process steps.

The project's environmental impact is notably influenced by sunlight intensity and harvesting practices. Moreover, the corresponding impacts and resource demand are closely tied to the productivity levels of the high-value products considered within this project's scope. In examining the overall environmental impacts, encompassing both midpoint and endpoint analyses, the study underscores critical environmental effects induced by the system. The research reveals that the primary environmental impacts stem from marine ecotoxicity, fossil resource depletion, and climate change-fossil. Significantly, the stages exerting the most substantial influence on these environmental aspects are upstreaming and harvesting. This correlation can be traced back to the considerable energy requirements and nutrient consumption intrinsic to algae cultivation.

Pierre et al. (2019) highlight the connection between microalgae biomass production and factors such as light intensity and aeration levels, underscoring a direct correlation with photosynthesis activity [37,51–54]. Notably, the enhanced biomass production of diverse microalgae species, including *Cyanospira capsulata*, *Porphyridium*, and *Synechococcus*, as well as the researched cyanobacteria *A. platensis*, is particularly notable under conditions of high continuous light irradiance [25–28]. This becomes apparent in the study setups when simulated for a situation characterized by increased solar exposure. The PC content at the light intensities of  $450 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  showed a significant rise relative to that at intensities of  $100 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ . This fact is likewise noted in the studies from [55]. Soni et al. (2019) further indicate that the greatest biomass productivity occurred in the summer months, coinciding with the highest recorded chlorophyll content [56].

The highest PC yield was 16.52 g per cycle under 70 L harvesting volume, while the lowest PC content was 3.68 g per cycle at low light intensity and 20 L of harvested water. Similarly, this holds true for EPS, showcasing peak values (3.28 g) in simulations conducted in regions with abundant sunlight and Scenario II configuration. In contrast, the lowest value is observed in the contrasting scenario, registering 0.62 g. Regarding biogas production, Scenario II exhibits the highest value at high light intensities, followed by Scenario II with low light intensities. Irrespective of the harvesting scenarios, processes under high light intensities demonstrate a higher yearly yield, correlating with the elevated growth kinetics. However, when comparing the yield of a single batch, harvesting 70 L yields a superior outcome. This discrepancy arises from the comparison between yield (per batch) and space-time yield (per year).

In the present study, the environmental impact of PC extraction concerning climate change impact, the authors report lower values with water as the solvent ( $1.25 \times 10^3$  kg CO<sub>2</sub> eq) compared to ethanol ( $3.26 \times 10^5$  kg CO<sub>2</sub> eq). In contrast, the current investigation, conducted under varying light intensities, revealed divergent GWP100 values. In low light settings, it was  $3.09 \times 10^7$  kg CO<sub>2</sub> eq, while scenarios of high light intensity demonstrated higher GWP100 values, ranging from  $1.20 \times 10^8$  kg CO<sub>2</sub> eq in Scenario I to  $1.36 \times 10^8$  kg CO<sub>2</sub> eq in Scenario II.

Consistent with this observation, the LCA results reaffirm that sodium bicarbonate, representing the bulk of the SOT compound, emerges as the primary stressor to this environmental impact. Furthermore, within the ReCiPe midpoint, this impact has the utmost magnitude after applying the normalization.

For the majority of the impacts analyzed, the cultivation process exerts a considerable environmental footprint. Therefore, the system is designed to recycle the regenerated media from the output of the EPS concentration process, lowering the need for additional resources. This approach enhances supply chain management, contributing to a reduction in the depletion of natural resources and minimizing environmental impacts throughout nutrient production and utilization [6,57,58].

The outcomes obtained from the IPCC (GWP 100a) methodology highlight the most significant environmental impact in the climate change categories, specifically climate change-fossil for the former and non-renewable fossil for the latter. This outcome primarily stems from the electricity consumption required for machinery operations. Potential

improvements to mitigate this impact involve exploring alternatives, such as shifting to renewable energy sources in lieu of grid electricity.

Conversely, employing microalgae and cyanobacteria yields positive outcomes for carbon mitigation and encompasses the capacity to extract nutrients from wastewater and various gaseous emissions. Studies have shown that the production of 1 kg of dry biomass requires approximately 1.8 kg of CO<sub>2</sub>. Combined with their fast growth, their CO<sub>2</sub> fixation efficiency is 10–50 times greater compared to terrestrial plants [37,51,52,55,58]. Likewise, phototrophic organisms have the capability to capture around 1.3 kg of carbon dioxide in the process of generating 1 kg of biomass. However, it is essential to consider the overall carbon balance, including not just the fixation of CO<sub>2</sub> during growth but also the subsequent release of CO<sub>2</sub> upon product utilization or degradation. For this reason, in this study, the boundaries were chosen so that the fixation of CO<sub>2</sub> and the subsequent release are net zero.

Looking at it from an endpoint methodology standpoint and considering the factors contributing to the process, the primary stressor impacting both water resources and human health is focused on the media components. These components are once again associated with the production of sodium bicarbonate. Water consumption during manufacturing poses challenges to water availability and quality, contributing to environmental stress and potential contamination. This is in line with the concerns raised by the World Health Organization regarding the implications of industrial activities on water quality and accessibility [59]. Additionally, the discharge of industrial by-products may introduce pollutants into water bodies, further compromising water quality. The compromised water quality resulting poses risks to human health, consistent with studies highlighting the correlation between water quality and public health [60–62]. Therefore, the management of material flows and impact factors stands out as a key area that demands attention in future technological advancements.

Responsible water management practices are fundamental to mitigate adverse effects on aquatic environments. Implementing efficient water recirculation systems and employing nutrient recovery techniques in microalgae and cyanobacteria cultivation is essential for responsible water management, helping minimize environmental impact by reducing water consumption and nutrient runoff [7,60,61,63]. Moreover, adopting precision technologies and optimizing cultivation parameters can enhance the sustainability of microalgae and cyanobacteria production, ensuring a balance between resource utilization and environmental conservation. Integrating microalgae and cyanobacteria production with photovoltaic panels presents numerous benefits, with the primary advantage being the utilization of surplus solar energy to fulfill the substantial energy requirements of biodiesel production [62]. Alternatively, research indicates that optimizing algal biomass production could be achieved by combining open ponds and photobioreactors, creating a hybrid system and ensuring that the unique advantages of each methodology are appropriately utilized and valued [64,65].

Recognizing the intricacy of the situation emphasizes the importance of a dual approach utilizing both midpoint and endpoint indicators. This comprehensive method is essential for capturing the diverse aspects of environmental impacts, considering their implications on ecosystems and human well-being, and providing globally representative characterization factors [21,47,66].

The economic outlook of exploring algae cultivation for economic benefits reveals significant promise, particularly in addressing environmental and human health concerns. Nevertheless, the hurdles to achieving project scalability while ensuring economic viability persist. Therefore, a key objective of this project is to overcome these challenges by concurrently generating two high-value products for the market.

Moreover, generating high-value products, such as PC at market price, market prices at 170–280 USD·kg<sup>-1</sup> [67], and EPS at 300 USD·kg<sup>-1</sup> [37], distinguishes this initiative from conventional bioenergy-oriented projects. The strategic emphasis on products with significant market value aims to offer a unique approach to mitigating costs. Moreover,

findings reported by Chalermthai et al. (2023) indicate that the simultaneous production of spirulina powder and bioplastic represents a promising endeavor, demonstrating a payback time (PBT) as brief as 2.6 years and a return on investment (ROI) reaching a noteworthy 38.5% [68].

The project's potential is clarified by examining the energy demand relationship between the processes of extracting both products compared to the overall energy consumption for cultivation and harvesting. In essence, when we allocate the cultivation and extraction stages as the system's core elements, the extraction processes for by-products exert minimal influence on the total energy demand. To emphasize, even if EPS and PC were neglected from the system, their absence would not significantly affect the overall impact. This resilience highlights the potential economic viability of the system. As a result, the project's dual focus on environmental sustainability and the production of high-value goods positions it as a promising avenue for economic progress and resource optimization.

Finally, performing a comprehensive LCA is paramount when evaluating cyanobacteria cultivation projects' environmental impacts. However, it is essential to acknowledge the inherent limitations of comparing studies in this field. Numerous variables come into play, including the specific species of microorganisms being cultivated, technology employed, cultivation conditions, and geographical factors. Each of these variables can significantly influence the outcomes and make direct comparisons challenging. Moreover, the diversity in methodologies used for analysis across studies introduces an additional layer of complexity. Recognizing these limitations underscores the importance of cautiously approaching the assessment and advocating for standardized methods to enhance the reliability and comparability of results in the evolving field of algae cultivation.

## 5. Conclusions

This environmental assessment reveals that scenarios with higher light intensity and lower water volume demonstrate the lowest environmental impacts, emphasizing the intricate relationship between harvesting volume, duration, and environmental repercussions. Despite higher environmental impacts in Scenario II, a nuanced analysis of EPS and PC extraction shows variations, highlighting the complexity of system parameters.

The project exhibits a multifaceted landscape of both favorable and challenging aspects. Highlighting the optimistic aspect, the incorporation of a biogas plant not only produces methane but also strategically supplies process heat for the algae culture, effectively addressing resource efficiency. The cyanobacteria cultivation for biofuel production represents a significant stride towards a more sustainable economy, aligning with the global effort to address fossil fuel scarcity. However, this project distinguishes itself by engaging in biomass generation for energy and incorporating the extraction of two high-value products, PC and EPS. A closer examination of this unique approach reveals its potential, particularly in terms of environmental impact. Interestingly, the extraction and concentration steps, often considered environmentally burdensome, play a minor role compared to the cultivation and harvesting process—the primary contributors to resource demand. This nuanced insight positions the project as an environmentally conscientious initiative, strategically minimizing its ecological footprint where it matters most. Beyond its eco-friendly attributes, the inclusion of PC and EPS in the system offers substantial economic advantages, given their high market value. This holistic integration of economic, environmental, and societal benefits underscores the project's significant contribution to a more balanced and sustainable future.

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