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Life Sciences und
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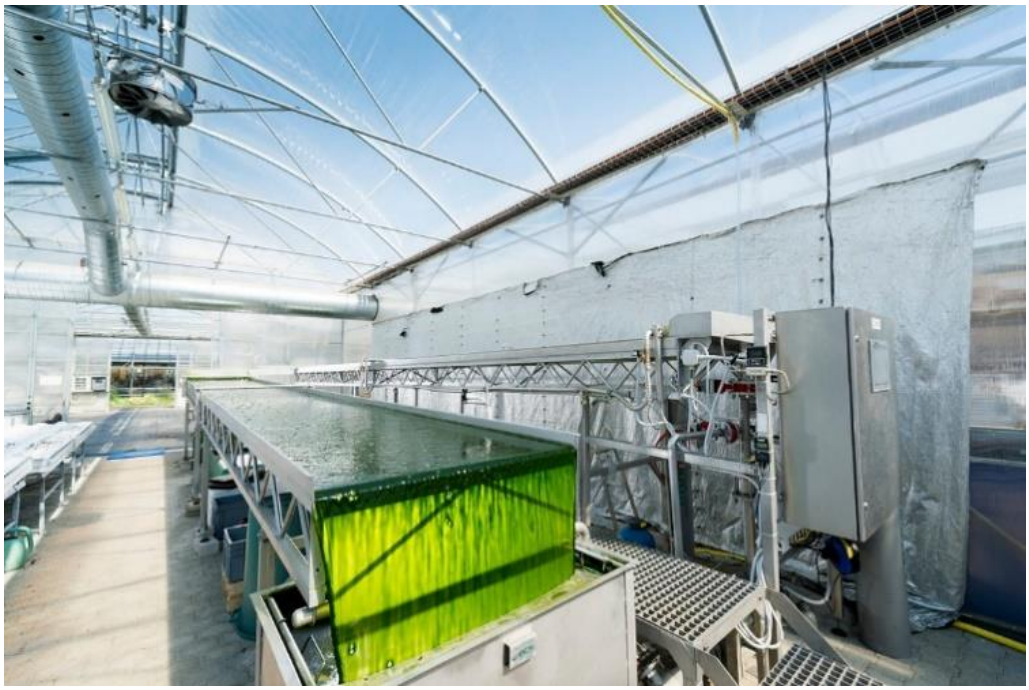
Life Cycle Assessment of Bio-based Materials

Environmental impacts of the value chain from cyanobacteria to PHB



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ABSTRACT

Considering the severe impacts of synthetic polymers on the environment as well as the human health, the development of biopolymers is becoming increasingly important. While biopolymers such as Bio-PA or Bio-PET address the issue of fossil resource depletion, they are not automatically biodegradable and can cause significant environmental impact in their respective value chains. One alternative to petroleum-based polymers while also being biodegradable is Polyhydroxybutyrate (PHB). Cyanobacteria is one of various bacterial strains capable of producing PHB. The aim of this study was to conduct a life cycle assessment (LCA) of cyanobacteria production on a thin-layer photobioreactor (PBR) pilot plant, and to extrapolate the data to an industrial-scale production scenario.

Primary data was collected on a pilot-scale thin-layer PBR, where cyanobacteria are cultivated using three different cultivation media being a Z medium (a standard mineral medium) and two variants of Z medium where the nitrogen source is replaced with either water from an aquaculture system or pre-processed liquid digestate. The functional unit was chosen as 1 kg of cyanobacteria biomass (dry matter). Processes included in the LCA reach from construction of the thin-layer PBR and its operation to the centrifugation of the cyanobacteria biomass. The environmental impacts of the cyanobacteria production were analysed using a selection of environmental impact categories according to the environmental footprint method, greenhouse gas (GHG) emissions according to the Intergovernmental Panel on Climate Change (IPCC) and the total environmental impact according to the ecological scarcity method.

Due to higher yields, cyanobacteria cultivation in the Z medium showed lower environmental impacts across all categories analysed. Main contributors to the environmental impacts of the pilot plant production were found to be the CO_2 fed to the cultures and electricity consumption of the pumps and centrifugation, across all three cultivation media. Extrapolation of the data from the pilot plant to an industrial-scale production scenario, where yield is increased and CO_2 input is decreased, showed a reduction potential of > 80 % across all environmental impacts analysed, leaving electricity consumption as the distinct primary environmental hotspot. In terms of climate change, the industrial-scale scenario showed a reduction of GHG emissions from 35 kg CO_2-eq to 4 kg CO_2-eq per kg of cyanobacteria for the Z medium. Regarding the aquaculture water and liquid digestate, GHG emissions per kg of cyanobacteria were reduced from 90 kg CO_2-eq to 8.7 kg CO_2-eq and 8.5 kg of CO_2-eq, respectively.

The present study thus shows, that from an environmental perspective, cyanobacteria production using waste streams would have to achieve an increase of productivity rates by a factor 3, if it were to

compete with production using a Z medium. Regardless of the medium used, focus for reducing the environmental impact of PHB production from cyanobacteria should lie on reducing the CO_2 input while aiming to maximise the areal productivity rates as well as decreasing electricity consumption, in order to compete with PHB produced from other feedstock as well as petroleum-based polymers.

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1 INTRODUCTION

Plastic is one of the most widely produced materials in the world. Production of the synthetic polymer has been consistently increasing over the past decades, reaching a world production of 370 Mt in 2020 (Plastics Europe, 2021). Along with the increasing production of plastics, their threat to the environment is becoming increasingly clear. Environmental impacts of plastics are primarily caused by the disposal processes, since a significant amount of the produced material serves an ephemeral purpose and is rapidly converted into waste (Kabir et al., 2020). A vast majority of plastic is incinerated at the end of its short life, which leads to large amounts of greenhouse gas emissions (CIEL et al., 2019). Another threat represents the plastic pollution of the environment through littering. As synthetic polymers are primarily petroleum-based, degradation of the materials is slow and leads to fragmentation into the smaller particles. These particles subsequently contaminate water, air and soil and lead to severe impacts on the environment as well as the human health (Ilyas et al., 2018). Finally, being petroleum-based, synthetic polymers further contribute to the depletion of non-renewable energy sources.

In this context, the development of biopolymers is becoming increasingly important, as they represent a bio-based alternative to the synthetic polymers. Most of the bioplastics on today's market are derived from agricultural-based feedstock such as Bio-PE, Bio-PET and Bio-PA, and present physiochemical and thermoplastic characteristics similar to those of petroleum-based polymers (Mercado et al., 2017). While thereby addressing the problem of fossil resource depletion, such bioplastics are not biodegradable and thus do not counteract the environmental impacts of synthetic plastics. Additionally, the production of agricultural feedstock leads to high environmental impacts, especially in terms of eutrophication potential (Kim & Dale, 2005). Reduction potential of the environmental impacts has been shown to be more significant when agricultural waste streams are used as feedstock for bioplastics, rather than agricultural feedstock itself (Samer et al., 2022).

An alternative to petroleum-based polymers while also being biodegradable is Polyhydroxybutyrate (PHB), which has physiochemical properties similar to those of polypropylene (PP) and polyethylene (PE) (McAdam et al., 2020). Cyanobacteria, a phylum of bacteria that obtain their energy through photosynthesis, are one of the microorganisms that are capable of producing PHB photoautotrophically (Markl et al., 2018). Opposed to heterotrophic cultivation of cyanobacteria, which generally yields higher PHB contents, photoautotrophic cultivation avoids the need of organic carbon and therefore eliminates the competition for primary sugars from food and feed production (Shahzad et al., 2020). In contrast, photoautotrophic cultivation of cyanobacteria requires inorganic

carbon in the form of CO_2 (Mariotto et al., 2022), which can have a substantial impact on the environment, depending on the source and production processes of the CO_2 (Koornneef et al., 2012).

To date, knowledge on the environmental impacts of PHB production from cyanobacteria is limited, with many uncertainties regarding the feasibility of an environmentally friendly production, especially on an industrial scale (Carpine et al., 2020). While environmental analyses of PHB production from other bacterial strains have been performed (Gupta et al., 2021; Vogli et al., 2020), no such analyses of PHB production from cyanobacteria are available. Goal of the present study was therefore to conduct a life cycle assessment (LCA) of cyanobacteria production on a thin-layer PBR pilot plant, and to extrapolate the data to an industrial-scale production scenario. The study thus gives an insight into the environmental performance of PHB production from cyanobacteria and identifies environmental improvement possibilities for future development of an industrial-scale application of this technology.

2 GOAL AND SCOPE DEFINITION

2.1 OVERVIEW AND GOALS

For the evaluation of the environmental impact of cyanobacteria production on a thin-layer PBR, a Life Cycle Assessment (LCA) was performed. The LCA of the cyanobacteria production allowed an analysis of the environmental performance of the studied system from raw material extraction to harvested and centrifuged cyanobacteria. According to the ISO 14040 standards (ISO, 2006), the following four stages were undertaken: goal and scope definition, inventory analysis, impact assessment and interpretation.

The goal of the present study was to assess the environmental impact of the PHB-producing cyanobacteria *Synechococcus leopoliensis*, cultivated on a thin-layer PBR using Z medium (a standard mineral medium) and two variants of Z medium where the nitrogen source is replaced with either water from an aquaculture system or pre-processed liquid digestate. Aim of the study was further to scale the cyanobacteria production from the pilot plant to industrial scale, in order to assess the environmental performance of industrial-scale PHB production from cyanobacteria.

2.2 FUNCTIONAL UNIT

The functional unit of the present LCA was chosen as 1 kg of produced cyanobacteria biomass (dry weight). To facilitate comparison to literature, results of the impact assessment were additionally calculated for the functional unit of 1 kg of PHB.

2.3 SYSTEM BOUNDARY

Processes included within the system boundaries are cyanobacteria cultivation, PHB accumulation and centrifugation (Figure 1). Cyanobacteria cultivation considers, on the one hand, the raw materials and resources needed for the construction of the thin-layer PBR. The plant is primarily made out of steel for the scaffolding, as well as glass plates creating the surface for the cyanobacteria production. The cultivation stage further includes the resources needed during the operation of the pilot plant. Main resources include the medium, being either Z medium, aquaculture water or liquid digestate, reactor and nutrient pumps, CO_2, nutrients, EDTA and electricity to operate the pumps. After a 14-day period of cyanobacteria cultivation, the cultures are transferred into a separate tank for the PHB

accumulation. During this process, no resources or energy are needed, with exception of some building materials for the tank. Once PHB is successfully accumulated, the cyanobacteria cultures are transferred to the centrifuge. Resources for this final process are steel, for the construction of the centrifuge, as well as electricity for the centrifugation of the cultures. PHB extraction was excluded from analysis.

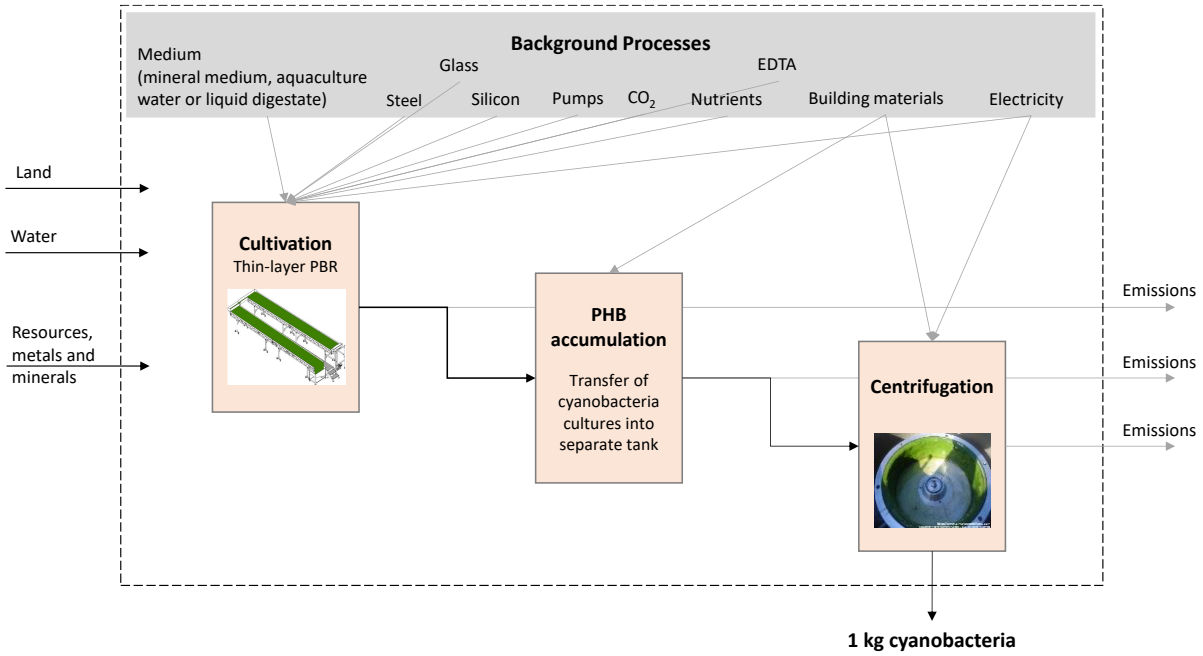

Figure 1 System model of cyanobacteria production on the thin-layer PBR at the Institute of Natural Resource Sciences of the Zurich University of Applied Sciences.

2.4 DATA COLLECTION

Primary data regarding the cultivation of cyanobacteria on the thin-layer PBR pilot plant have been collected with a questionnaire filled in by Marina Mariotto[1]. Yield and operation data stem from the period between July and September 2021. Data regarding the industrial-scale scenario of the cyanobacteria production were attained by extrapolating the pilot plant data, values for the adjusted parameters were derived from literature.

[1] Marina Mariotto, questionnaire, 19 November 2021, Institute of Natural Resource Sciences, ZHAW.

2.5 Allocation

Allocation describes the partitioning of input or output flows of processes or products in case of multi-output processes. The primary product system of the present LCA does not include the production of co-products, accordingly no allocation was necessary.

2.6 LCA Software and Databases

For the background data, the ecoinvent v3.8 database with the system mode *«allocation, cut-off by classification - unit»* was used (ecoinvent Centre, 2021). Modelling and analysis were performed using the life cycle assessment software SimaPro v9 (PRé Consultants, 2019).

2.7 Life Cycle Impact Assessment Methods

To assess the environmental impact of the cyanobacteria production in three different media, the impact assessment methods listed in Table 1 were applied. The selection of impact assessment methods is based on the recommendation of the Joint Research Council of the European Commission for the calculation of the Product Environmental Footprint (PEF), according to Fazio et al. (2018) and Hauschild et al. (2011). The selection of impact assessment methods includes twelve different environmental impacts: (1) climate change, (2) ionising radiation, (3) freshwater eutrophication, (4) marine eutrophication, (5) terrestrial eutrophication, (6) toxic emissions to humans and (7) ecosystems, and (8) non-renewable primary energy demand. In addition, the total environmental impact is shown in environmental impact points according to the ecological scarcity method (Frischknecht et al., 2013). The weighting of different environmental impacts into an aggregated figure is not in accordance with the ISO standards for life cycle assessments (ISO, 2006) and therefore the results according to the ecological scarcity method are described in a separate chapter.

Table 1 Overview of the impact assessment methods used.

Indicator	Method	Description
Climate change	IPCC (2021)	The impact category climate change according to IPCC 2013 takes into account all emissions that contribute to climate change. The potential climate impact of a greenhouse gas is compared with the climate impact of CO_2 and expressed in CO_2 equivalents.
Ionising radiation	Frischknecht et al. (2000)	The impact category ionising radiation considers the effects of ionizing radiation on human health and is indicated in kBq U-235 eq.
Eutrophication, freshwater	Goedkoop et al. (2009)	Freshwater eutrophication assesses nutrients in freshwater (phosphorus as limiting nutrient), unit: kg P eq.
Eutrophication, marine	Goedkoop et al. (2009)	Marine eutrophication assesses nutrients in marine waters (N as a limiting nutrient), unit: kg N eq.
Eutrophication, terrestrial	Posch et al. (2008); Seppälä et al. (2006)	Terrestrial eutrophication assesses the effect of nutrients in sensitive terrestrial ecosystems, unit: mol N eq.
USETox, human health USEtox, ecosystem	Rosenbaum et al. (2011)	USEtox is an impact assessment method for characterizing toxic effects of chemicals on human health as well as ecosystems, indicated in disease increase and PAF/m^3/d. USETox is the recommended method according to the UNEP/SETAC Life Cycle Initiative.
Primary energy, non-renewable	Van Oers et al. (2002)	This impact assessment method assesses the extraction of abiotic, non-renewable resources from nature. Results are indicated in MJ.
Total environmental impact, according to the ecological scarcity method 2013	Frischknecht et al. (2021)	The ecological scarcity method weights emissions and resource consumption according to policy targets. The result is expressed in eco-points. This impact assessment method involves weighting and is therefore not ISO-compliant. The results are therefore reported in a separate chapter.

2.8 SENSITIVITY ANALYSIS

In order to test the robustness and reliability of the results in LCA studies, sensitivity analyses can be carried out. In the present study two aspects were investigated in a sensitivity analysis, being the yield difference between the media on the one hand, and potential benefits of using wastewater on the other hand:

- Yield: assuming equal yields across the three media used
- Wastewater: analysing potential benefits of using aquaculture water in terms of avoided environmental impacts

The sensitivity analysis regarding equal yields is relevant for the present study, as yield represents a crucial reference value. Additionally, data of this study is based on early-stage experiments and extrapolation of the data therefore contains a certain degree of uncertainty. Assuming a harmonised yield between the different media allows an investigation of environmental impacts between the

different cultivation media while disregarding heterogeneities in yield, which may alter in further experiments.

The sensitivity analysis regarding benefits of using aquaculture water as the cultivation medium considers the additional function of the cyanobacteria cultivation as a wastewater treatment plant. Although not its main function in this context, the thin-layer PBR simultaneously serves as a wastewater treatment plant. For this reason, three different avoidances are considered. Firstly, avoided environmental impacts are analysed, in the case of discharging aquaculture water directly into natural waters. This is the most common disposal method of aquaculture wastewater and must be in accordance to the Swiss water protection ordinance (GSchV, 2021). For the present study, the most relevant emissions of direct discharge are nitrate emissions, which were calculated as an avoided emission according to the amount of nitrogen taken up by the cyanobacteria biomass. This scenario is described as the discharge of low-polluted water. Further, the direct discharge of higher-polluted water is analysed, where next to nitrate additional emissions of the chemical oxygen demand (COD), phosphorus and ammonium are considered. Data for this scenario was based on an existing dataset regarding wastewater from a salmon recirculation system. Lastly, an avoided wastewater treatment is analysed, in case of discharging aquaculture wastewater into a wastewater treatment plant. Data for this scenario was based on an average wastewater treatment in Switzerland, according to the ecoinvent v3.8 database.

3 LIFE CYCLE INVENTORY

3.1 THIN-LAYER PHOTOBIOREACTOR

All data regarding the cyanobacteria production stem from experiments performed on an open thin-layer photobioreactor (PBR) at the Zurich University of Applied Sciences in Wädenswil, Switzerland (Figure 2). The pilot plant is placed in a greenhouse and has a sun-exposed surface of 18 m^2, holding a volume of 200 l. *Synechococcus leopoliensis*, which through experiments proved the most reliable growth with the highest PHB content, was cultivated in three different media. The first medium is a Z medium, where $NaNO_3$ is the main nitrogen source. In addition, two different types of wastewaters are used, to equimolarly replace the nitrogen in the Z medium, being water from aquaculture systems (hereinafter referred to as aquaculture water) and pre-processed liquid digestate (hereinafter referred to as liquid digestate). Cultivations of the cyanobacteria last for an average of 14 days, describing one growth cycle of the strain. CO_2 is continuously supplied to the system at a partial pressure of 5-10 mbar during the day, while CO_2 supply is stopped overnight. Nutrients for the cultivation in the Z medium are added batchwise and always before becoming limiting. The aquaculture water is added using a pump and based on the amount of water evaporated from the system, while the liquid digestate is continuously fed to the system via a peristaltic pump. After the 14-day cultivation period on the PBR, the cyanobacteria cultures are transferred into nutrient-depleted mineral medium to accumulate PHB. Subsequently, the cultures are centrifuged and resuspended in the nutrient-depleted media (Mariotto et al., 2022).

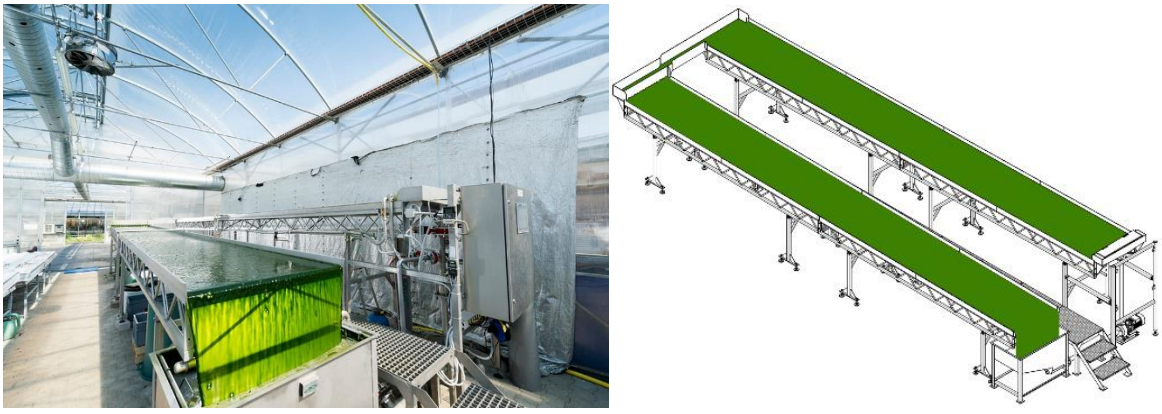

Figure 2 Thin-layer PBR for cyanobacteria production, located at the Institute of Natural Resource Sciences of the Zurich University of Applied Sciences (Mariotto et al., 2022).

3.2 PILOT PLANT

Table 2 shows an overview of the most relevant input and output parameters for the cyanobacteria production by the pilot plant, for 1 year. The cyanobacteria production includes the cultivation, harvest and centrifugation of the cyanobacteria biomass. All weight specifications refer to cyanobacteria dry weight (DW). The cyanobacteria cultivation is performed in three different media being a Z medium, aquaculture water and liquid digestate. The thin-layer PBR has a surface 18 m^2. Highest areal productivity rate was achieved in the Z medium, with 4.8 g m^{-2} d^{-1}. In the aquaculture water and liquid digestate, areal productivity rate was around three times lower, producing 1.6 g m^{-2} d^{-1}. These yield figures represent maximum values that were achieved during the summer. Extrapolating the values to one year, cultivation in the Z medium yields a yearly production volume of 18 kg, while yields in the aquaculture water and liquid digestate amount to 6 kg per year. The yearly CO_2 input amounts to 456 kg for all three media. For the Z medium, the carbon uptake rate of the cyanobacteria biomass is consequently at 6 %. Carbon uptake rate of cyanobacteria cultivated in aquaculture water and liquid digestate is at 2 %. Energy input includes energy demand of the pumping system (reactor pump and nutrient pump) and centrifugation. As in the Z medium no nutrient pump is necessary, energy input is lower compared to the other two media with 1485 kWh. Energy input for cultivation in the aquaculture water and liquid digestate is 1517 kWh and 1638 kWh, respectively. The Z medium contains $NaNO_3$ and $Ca(NO_3)_2$. The yearly amount of N added to the cultures adds up to 2.56 kg. The aquaculture water itself contains a concentration of NO_3^--N of 35.3 mg L^{-1}, equalling at a yearly N input of 0.45 kg. Liquid digestate contains a concentration of N of 6419 mg L^{-1}, which is equally completed by adding $Ca(NO_3)_2$, resulting in a yearly N input of 1.26 kg. Of the total N input, between 17-52 % is lost to the air, while 2-5 % remains in the water. Regarding the Z medium and aquaculture water, N was primarily present as nitrate (NO_3^-). All N losses to the air were assumed to be in the form of N_2 through denitrification, amounting to 0.42 kg for the Z medium and 0.11 kg for the aquaculture water. As for the liquid digestate, N was primarily present as ammonium (NH_4^+) and N losses to air were therefore assumed to be in the form of ammonia (NH_3), resulting in yearly N emissions as NH_3 of 0.65 kg. The percentage of N lost to the water was assumed to be in the form of NO_3^- for the Z medium and aquaculture water and NH_4^+ for the liquid digestate, for all three media and equals in N emissions of 0.12 kg, 0.02 kg and 0.1 kg, respectively.

Table 2 Overview of most relevant input and output parameters of the production of cyanobacteria by the pilot plant. All values refer to the operation of the bioreactor for 1 year. The parameters are indicated for the three media, Z medium, aquaculture water and liquid digestate.

Input Cyanobacteria production	**Unit**	**Z medium**	**Aquaculture water**	**Liquid digestate**
Reactor size	m^2	18	18	18
Areal productivity rate	g m^{-2} d^{-1}	4.76	1.59	1.59
Operation time of reactor	d/a	213	213	213
Yearly production volume	kg	18.25	6.08	6.08
PHB content	%	5	5	5
CO_2 input	kg	456.25	456.25	456.25
Energy input	kWh	1484.76	1516.70	1638.06
Nutrients				
N, total	kg	2.56	0.45	1.26
N, in biomass	kg	2.01	0.33	0.58
	%	78.6	72.3	46.2
N losses to air	%	16.6	23.6	51.5
N losses to water	%	4.8	4.1	2.3
Emissions	**Unit**			
N in N_2 to air	kg	0.42	0.11	-
N in NH_3 to air	kg	-	-	0.65
N in NO_3^- to water	kg	0.12	0.02	-
N in NH_4^+ to water	kg	-	-	0.1

Table 3 shows the same input and output parameters as described above, with specific values for 1 kg of cyanobacteria DW and 1 kg of PHB. CO_2 input in the Z medium is 28 kg per kg of cyanobacteria and 556 kg per kg of PHB. Due to the lower productivity rate in the aquaculture water and liquid digestate, CO_2 input per kg of cyanobacteria and per kg of PHB is three times higher in these two media, being 83 kg and 1667 kg, respectively. Energy input in the Z medium is at 90 kWh per kg of cyanobacteria and at 277 kWh and 299 kWh in the aquaculture water and liquid digestate, respectively. The considerably lower energy input for cyanobacteria cultivation in the Z medium is explained by the higher productivity rate as well as the lower energy demand compared to the other two media, since no nutrient pump is used. Total N input is highest in the liquid digestate with 0.23 kg per kg of cyanobacteria, followed by Z medium with 0.16 kg and aquaculture water with 0.08 kg. Accordingly, N emissions as N_2 and NO_3^- are also highest in the cultivation in the Z medium. See Table 7 in Appendix I for complete inventory data per kg of cyanobacteria.

Table 3 Overview of most relevant input and output parameters of the production of cyanobacteria, indicated per kg of cyanobacteria as well as per kg of PHB. The parameters are indicated for the three media, Z medium, aquaculture water and liquid digestate.

		per kg cyanobacteria			per kg PHB		
Input	**Unit**	**Z medium**	**Aquaculture water**	**Liquid digestate**	**Z medium**	**Aquaculture water**	**Liquid digestate**
Reactor size	m^2	18	18	18	18	18	18
Areal productivity rate	g m^{-2} d^{-1}	4.76	1.59	1.59	4.76	1.59	1.59
Yearly production volume	kg	18.25	6.08	6.08	18.25	6.08	6.08
PHB content	%	5	5	5	5	5	5
Operation time of reactor	d/a	213	213	213	213	213	213
CO_2 input	kg	27.78	83.33	83.33	555.56	1666.67	1666.67
Energy input	kWh	90.40	277.02	299.19	1807.93	5540.46	5983.79
Nutrients							
N, total	kg	0.16	0.08	0.23	3.11	1.66	4.59
N, in biomass	kg	0.12	0.06	0.11	2.45	1.20	2.12
Emissions	**Unit**						
N in N_2 to air	kg	2.58E-02	1.95E-02	-	5.16E-01	3.91E-01	-
N in NH_3 to air	kg	-	-	1.18E-01	-	-	2.36E+00
N in NO_3^- to water	kg	7.47E-03	3.39E-03	-	1.49E-01	6.79E-02	-
N in NH_4^+ to water	kg	-	-	1.81E-02	-	-	3.63E-01

3.3 UPSCALING OF PILOT PLANT TO INDUSTRIAL SCALE

An overview of the most relevant parameters for the industrial-scale scenario of the cyanobacteria production are depicted in Table 4. Values are indicated for the operation of the bioreactor for 1 year. The size of the reactor was upscaled to 9863 m^2, compared to the 18 m^2 of the pilot plant. This upsizing equals in an upscaling factor of 548. When the areal productivity rate in the Z medium is kept at the same values as for the pilot plant, 4.76 g m^{-2} d^{-1}, a yearly production volume of 10 tonnes can be achieved. It is indicated by Borowitzka & Vonshak (2017) that 10 tonnes per year is the lowest limit for an industrial-scale production of cyanobacteria. For the aquaculture water and liquid digestate, with a productivity rate of 1.59 g m^{-2} d^{-1}, the production volume amounts to 3.34 tonnes of cyanobacteria per year. CO_2 input per year accordingly amounts to 250 tonnes, for all three media. For the Z medium, energy input is 814 MWh, slightly lower compared to 831 MWh for the aquaculture water and 898 MWh for the liquid digestate. Total N input per year is 1.4 tonnes in the Z medium, compared to 248 kg in the aquaculture water and 688 kg in the liquid digestate. In terms of emissions, this results in 232 kg of N_2 emissions in the Z medium and 59 kg in the aquaculture water, per year. Yearly NH_3

emissions in the liquid digestate amount to 354 kg, while between 10 kg and 67 kg of NO_3^- and NH_4^+ emissions arise between the different media.

Table 4 Overview of most relevant input and output parameters for the cyanobacteria production on industrial scale. All values refer to the operation of the bioreactor for 1 year. The parameters are indicated for the three media, Z medium, aquaculture water and liquid digestate.

Input Cyanobacteria production	Unit	Z medium, industrial scale	Aquaculture water, industrial scale	Liquid digestate, industrial scale
Reactor size	m^2	9863	9863	9863
Areal productivity rate	g m^{-2} d^{-1}	4.76	1.59	1.59
Yearly production volume	t	10	3.34	3.35
PHB content	%	5	5	5
Operation time of reactor	d/a	213	213	213
CO_2 input	t	250	250	250
Energy input	MWh	814	831	898
Nutrients				
N, total	kg	1400	248.3	688.3
N, in biomass	kg	1100	179.6	318.0
Emissions	**Unit**			
N in N_2 to air	kg	232.40	58.61	-
N in NH_3 to air	kg	-	-	354.49
N in NO_3^- to water	kg	67.20	10.18	-
N in NH_4^+ to water	kg	-	-	54.42

In addition to the upscaling of the pilot plant, a few parameters were adjusted in a second step, shown in Table 5. The parameters which were adjusted for the upscaling include areal productivity rate, PHB content and CO_2 input. The areal productivity rate in the Z medium was raised from 4.8 g m^{-2} d^{-1} to 20 g m^{-2} d^{-1}, as this value is described as the upper limit of potential productivity in microalgal cultivation systems (Borowitzka & Vonshak, 2017; Grobbelaar, 2012). According to the lower productivity rate by two thirds in the other two media, cyanobacteria production in aquaculture water and liquid digestate was set at 6.67 g m^{-2} d^{-1}. With a plant size of 9863 m^2 and the increased areal productivity rates, the yearly production volumes amount to 42 tonnes in the Z medium and 14 tonnes in the other two media. The second parameter, which was adjusted for the upscaling is the PHB content of the cyanobacteria. PHB content was raised from 5 % to 60 %, as Drosg et al. (2019) indicate 60 % as the upper realistic limit for PHB content. Due to the increase in yield, the CO_2 input (baseline) is decreased by a factor 4.2, resulting in 6.6 kg of CO_2 per kg cyanobacteria in the Z medium and 19.8 kg in the other two media. CO_2 input was moreover the third parameter which was adjusted for the upscaling. Doucha

& Lívanský (2014) state in their paper that CO_2 utilization by algal cultures is at about 70 % in thin-layer systems, this value was therefore chosen as the upper limit. A 70 % CO_2 utilization in the Z medium leads to a CO_2 input of 2.3 kg per kg of cyanobacteria. With the areal productivity rate being 3 times lower in the aquaculture water and liquid digestate, CO_2 input per kg cyanobacteria amounts to 7 kg in these two media. Combining the yield increase and the increase in CO_2 uptake, the CO_2 input is roughly 50 times lower compared to the pilot plant scenario. Energy demand is decreased to 29 kWh per kg of produced cyanobacteria for the Z medium, 99 kWh for aquaculture water and 92 kWh for liquid digestate. Total N input, as well as all emissions are accordingly decreased compared to the cyanobacteria production of the pilot plant scenario, by a factor 4.2 for all three different media. For more details see Table 8 in Appendix II.

Table 5 Overview of most relevant input and output parameters adjusted for the cyanobacteria production on industrial scale. Values are indicated per kg of cyanobacteria as well as per kg of PHB and refer to the increased areal productivity rate. The parameters are indicated for the three media, Z medium, aquaculture water and liquid digestate.

		per kg cyanobacteria			per kg PHB		
Input	**Unit**	**Z medium**	**Aquaculture water**	**Liquid digestate**	**Z medium**	**Aquaculture water**	**Liquid digestate**
Reactor size	m^2	9863	9863	9863	9863	9863	9863
Areal productivity rate	g m^{-2} d^{-1}	20	6.67	6.67	20	6.67	6.67
Yearly production volume	t	42	14	14	42	14	14
PHB content	%	60	60	60	60	60	60
Operation time of reactor	d/a	213	213	213	213	213	213
CO_2 input, baseline	kg	6.61	19.8	19.8	33.1	99.2	99.2
CO_2 input, increased uptake	kg	2.3	7.0	7.0	11.6	34.9	34.9
Energy input	kWh	28.5	99.4	92.2	142.5	497.1	460.9
Nutrients							
N, total	kg	0.04	0.02	0.05	0.19	0.10	0.27
N, in biomass	kg	0.03	0.01	0.03	0.15	0.07	0.13
Emissions	**Unit**						
N in N_2 to air	kg	6.15E-03	4.65E-03	-	3.07E-02	2.33E-02	-
N in NH_3 to air	kg	-	-	2.81E-02	-	-	1.41E-01
N in NO_3^- to water	kg	1.78E-03	8.08E-04	1.26E-03	8.89E-03	4.04E-03	6.28E-03
N in NH_4^+ to water	kg	-	-	4.32E-03	-	-	2.16E-02

4 RESULTS

4.1 ENVIRONMENTAL IMPACT ON MIDPOINT LEVEL

Figure 3 depicts the environmental impact of nine midpoint categories, caused by the production of 1 kg of cyanobacteria by the pilot plant (baseline) with an areal surface of 18 m^2, for the cultivation in the three media Z medium, aquaculture water and liquid digestate. Also indicated are the environmental impacts of 1 kg of cyanobacteria from an upscaled PBR with a surface of 9'863 m^2, including an increased yield of 20 g m^{-2} d^{-1} (Z medium) and 6.67 g m^{-2} d^{-1} (aquaculture water and liquid digestate), as well as a decreased CO_2 input of 2.3 kg and 7 kg respectively, as indicated in Table 5. For all midpoint categories, for the baseline as well as the scaled scenarios, cyanobacteria production in the Z medium shows the lowest environmental impact, with on average between 30-40 % of the impacts of the other two media. This general outcome can mainly be explained by the higher yield (3 times higher) compared to the production in the other two media. Highest environmental impacts are caused either by the production in aquaculture water or liquid digestate, depending on the impact category in question. For most impact categories, production in the latter two media show very similar results. Regarding the scaled scenarios of the Z medium, environmental impacts are between 3.5-8.5 times lower compared to the baseline scenario. Regarding the aquaculture water and liquid digestate, upscaling of the baseline scenario reduces environmental impacts by 3.5-10.5 times, depending on the impact category.

In the baseline scenario, the impact categories greenhouse gas (GHG) emissions, freshwater and marine eutrophication, human toxicity (non-cancerous effects) and freshwater ecotoxicity are dominated by the CO_2 input, due to the high energy input and direct emissions during the production of the CO_2. Ionising radiation and fossil resource use are dominated by the electricity input in all three media. GHG emissions for the production of 1 kg of cyanobacteria in the Z medium amount to 34.5 kg CO_2-eq, while production in the aquaculture water and liquid digestate lead to 90 kg CO_2-eq per kilogram of cyanobacteria. Next to the CO_2 input as main contributor to GHG emissions, accounting for between 60-68 % of total GHG emissions depending on the medium, further contributors are electricity input (11-14 %) and steel input for the scaffolding (8-9 %). In contrast, the production of 1 kg of cyanobacteria in the Z medium in the industrial-scale scenario leads to GHG emissions of 4 kg CO_2-eq, while production in the aquaculture water and liquid digestate are at 8.7 kg CO_2-eq and 8.5 kg of CO_2-eq per kilogram of cyanobacteria. Compared to the baseline scenario, GHG emissions are thus reduced by a factor 9 for the Z medium, and a factor 10.5 for the aquaculture water and liquid

digestate. In contrast to production by the pilot plant, main contributor to GHG emissions of the industrial scale production scenario is the electricity input for all cultivation media, accounting for 31 %, 42 % and 46 % of the total GHG emissions, respectively. Despite absolute numbers being considerably lower compared to the cyanobacteria production by the pilot plant, the general relationship between the three media remains very similar in both scenarios. The main difference is that in contrast to the baseline scenario, where CO_2 input is a main contributor to most environmental categories, the environmental impact in the industrial-scale production scenario is primarily dominated by the electricity input for most categories considered.

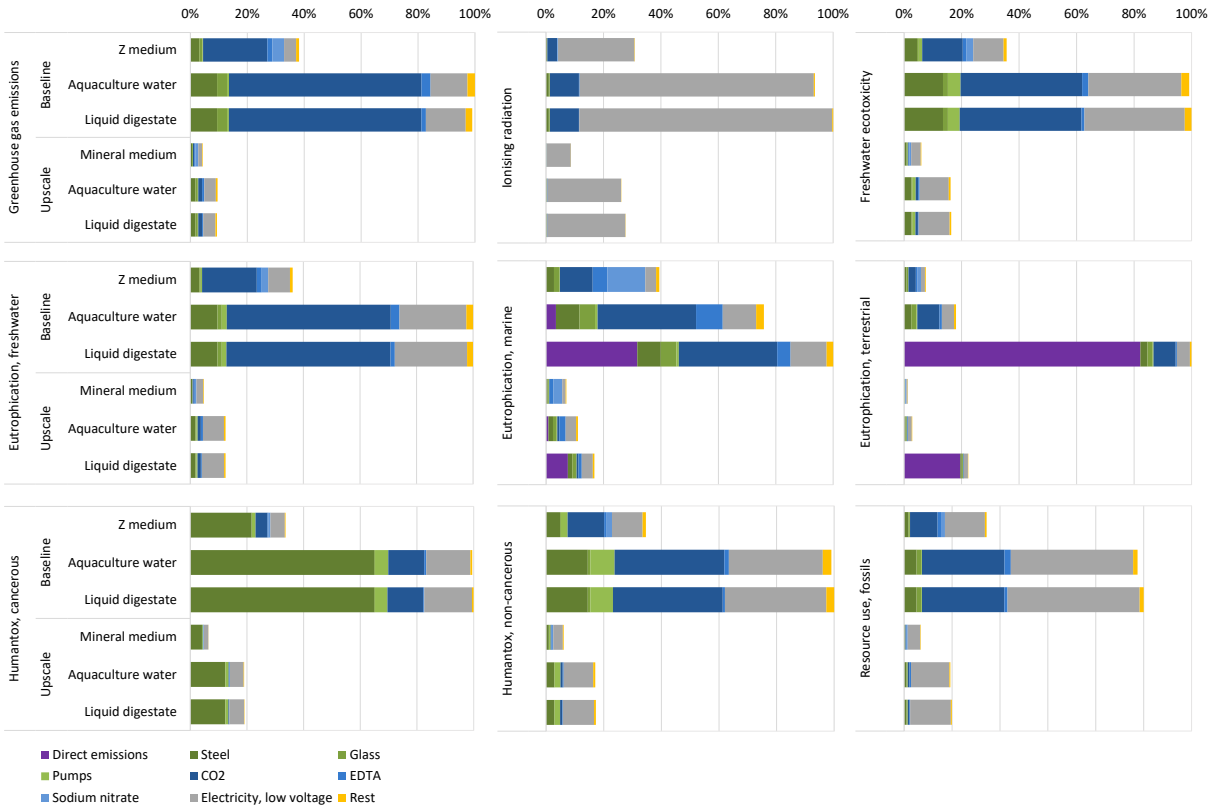

Figure 3 Environmental impacts of 1 kg of cyanobacteria, for the baseline and the upscale scenario. Results are shown for different mid-point categories are according to EF 3.0 method (Fazio et al., 2018), greenhouse gas emissions according to IPCC (2021), as well as human- and ecotoxicity according to USETox (Rosenbaum et al., 2011).

A distinctly different picture between the three media can be seen in the impact category terrestrial eutrophication. Cyanobacteria production in the liquid digestate shows the highest terrestrial eutrophication potential, being 82-94 % higher compared to the production in the Z medium and aquaculture water. The large difference between the media can primarily be explained by the NH_3

emissions during cultivation in the liquid digestate, accounting for 82 % (baseline) and 88 % (upscale) of the total terrestrial eutrophication potential.

4.2 TOTAL ENVIRONMENTAL IMPACT

The total environmental impact of the production of 1 kg of cyanobacteria by the pilot plant as well as the scaled scenarios are depicted in Figure 4.

Regarding the baseline scenarios, total environmental impact per kilogram of cyanobacteria is the lowest for the cultivation in the Z medium, with 91'300 eco-points. The total environmental impact for the cyanobacteria cultivation in aquaculture water and liquid digestate is 2.8-2.9 times higher compared to the Z medium, with 254'000 and 265'000 eco-points respectively. Considering the different life cycles stages of the cyanobacteria production, the operation of the plant causes the majority of the environmental impacts (blue shades). The process dominating the environmental impacts of the operation phase is the CO_2 input. In the Z medium, the CO_2 input accounts for 40 % of total environmental impacts, 43 % in the aquaculture water and 42 % in the liquid digestate. Another process of the operation phase with a high contribution to total environmental impacts is the electricity consumption, including electricity demand of reactor pump, nutrient pumps and centrifuge. Of total environmental impacts, electricity consumption accounts for 35 % in cyanobacteria cultivation in the Z medium, 39 % in aquaculture water and 40 % in liquid digestate. Production of the infrastructure of the bioreactor is responsible for 12 % of total environmental impacts in cyanobacteria cultivation in the Z medium, and 13 % in the aquaculture water and liquid digestate. Primary process contributing to the impacts of the infrastructure is the production of steel for the scaffolding of the reactor. Direct emissions, shown in purple, account for 2.6 % of total environmental impacts in the liquid digestate and < 1 % in the Z medium and aquaculture water. The direct emissions in the liquid digestate are largely caused by the NH_3 emissions, as explained in chapter 3.2.

Regarding the industrial-scale scenarios, the Z medium shows the lowest environmental impacts among the three different media, which is in accordance with the baseline scenarios. In the Z medium, the decreased CO_2 input leads to a decrease of the total environmental impact to 55'800 eco-points, being 39 % lower compared to the baseline scenario. This difference is almost exclusively explained by a lower impact from CO_2, as compared to the baseline scenario the environmental impact from the CO_2 is 7 times lower. Regarding cyanobacteria production in the aquaculture water, the decreased CO_2 input leads to a reduction of the total environmental impact to 147'000 eco-points, being 42 % lower compared to the baseline scenario. Similarly, decrease of the CO_2 input in the liquid digestate leads to

a reduction of the total environmental impact by 40 %, compared to its baseline scenario. Even larger is the effect of the third scenario, representing an increase in yield due to a higher areal productivity rate. In contrast to the CO_2 decrease, which merely reduced the environmental impact of one process, the yield increase affects all processes. In the Z medium, the total environmental impact is decreased to 24'000 eco-points, which is 74 % lower compared to the baseline scenario and 57 % lower compared

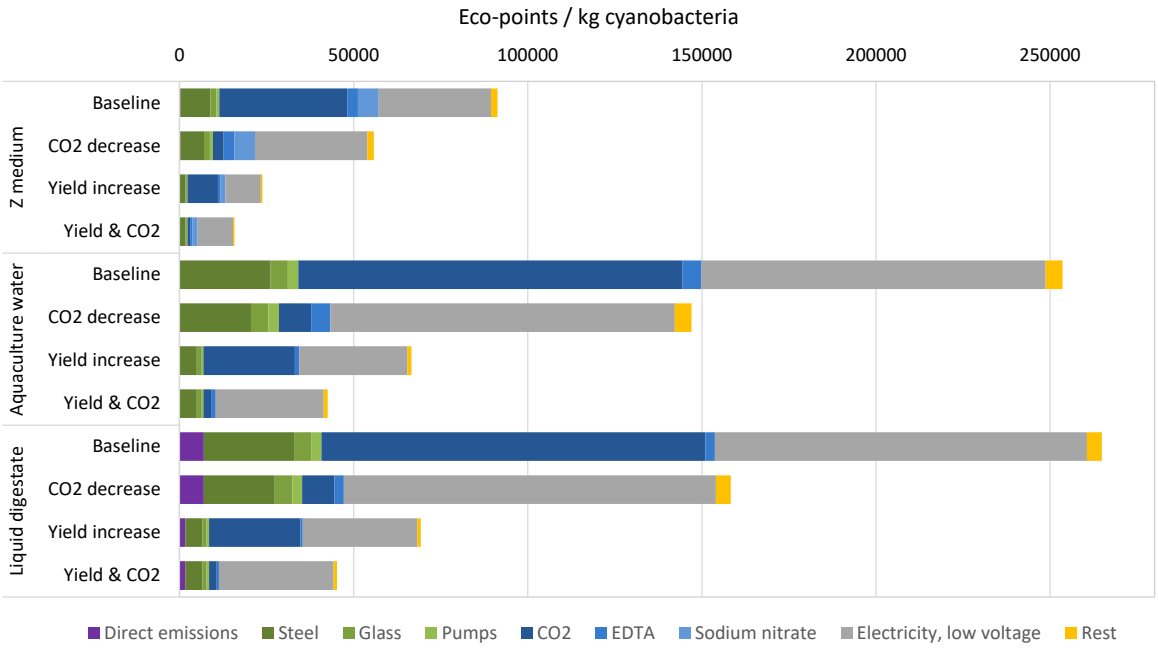

Figure 4 Total environmental impact per kilogram of cyanobacteria, for different scenarios and expressed in eco-points and according to the ecological scarcity method (Frischknecht et al., 2021). Baseline represents the production on the pilot plant, the other 3 scenarios are upscaled with parameter adjustments. Impacts are divided into direct emissions (purple), infrastructure of the plant (green shades), operation of the plant (blue shades) and rest / remaining processes (yellow).

to the decreased CO_2 scenario. Dominating processes in the present scenario are CO_2 and electricity input, which together account for 80 % of the total environmental impact. For cyanobacteria production in the aquaculture water and liquid digestate, the increased yield leads to a reduction of the total environmental impact by 74 % compared to their baseline scenarios, and 55-56 % compared to the CO_2 scenario. CO_2 and electricity input account for 85-86 % of the total environmental impact. The fourth scenario, combining the CO_2 decrease and the yield increase, reduces the total environmental impact by 83 % in all three media to 15'800 eco-points, 42'600 eco-points and 45'200 eco-points, respectively. Dominating process in the combining scenario in the Z medium are electricity input (64 %), steel for the scaffolding (10 %) and sodium nitrate input (9 %). In the

aquaculture water and liquid digestate dominating processes are electricity input with 73 %, steel input with 11-12 % and CO_2 input with 5 %.

5 DISCUSSION

Impact assessment results showed similar outcomes across the analysed midpoint categories as well as the total environmental impact. For all midpoint categories analysed, cyanobacteria production in the Z medium showed the lowest environmental impacts. For the majority of categories analysed, cyanobacteria production in the aquaculture water and liquid digestate showed very similar environmental impacts. Cyanobacteria production by the pilot plant leads to GHG emissions of 34.5 kg CO_2-eq in the Z medium and 90 kg CO_2-eq in the aquaculture water and liquid digestate, per kg of cyanobacteria. The industrial-scale scenarios, with adjustment of the parameters CO_2 input and areal productivity rate, showed that GHG emissions could potentially be reduced to 4 kg CO_2-eq (Z medium), 8.7 kg CO_2-eq (aquaculture water) and 8.5 kg CO_2-eq (liquid digestate), per kg of cyanobacteria. When referring these numbers to the PHB content of the cyanobacteria, GHG emissions amount to 6.6 kg CO_2-eq, 14.5 kg CO_2-eq and 14.1 kg CO_2-eq per kg of PHB, respectively.

Total environmental impact of cyanobacteria production by the pilot plant in the Z medium amounts to 91’300 eco-points per kilogram of cyanobacteria. In the aquaculture water, total environmental impact counts 254’000 eco-points and in the liquid digestate 264’000 eco-points per kilogram of cyanobacteria. By scaling the pilot plant to an industrial scale and adjustments of the parameters CO_2 input and areal productivity rate showed a maximum potential decrease of the total environmental impact to 15’800 eco-points in the Z medium, 42’600 eco-points in the aquaculture water and 45’100 eco-points in the liquid digestate, per kilogram of cyanobacteria.

5.1 COMPARISON WITH LITERATURE

Results of the present study are subsequently compared to other LCA studies of (bio)polymers, regarding GHG emissions (Figure 5). Regarding PHB from biomass, a PHB content of 60 % was consistently assumed for all scenarios and studies, in order to facilitate comparison. PHB extraction was not considered in the scenarios of the present study, due to lack of data. For the industrial-scale scenario of PHB production of the present study as well as the petroleum-based polymers, disposal of the polymer was additionally considered.

Outcomes of the present study are displayed for the pilot scale as well as the scaled scenarios to industrial scale. Comparisons to PHB from algae biomass, PHA/PHB from sugarcane and sewage sludge, and petroleum-based polymers PP, LDPE and PET were made. Regarding the pilot scale, Pérez-López and colleagues (2017) performed a comparative LCA study on microalgae cultivation on different pilot

reactors. The authors compared microalgae cultivation in a horizontal PBR, vertical PBR and open raceway pond (ORP), during summer, fall and winter. The areal productivity reached 10.5 g m^{-2} d^{-1} (ORP), 12.1 g m^{-2} d^{-1} (horizontal PBR) and 19.4 g m^{-2} d^{-1} (vertical PBR). For microalgae production in summer, GHG emissions of the different cultivation systems varied between 214 kg CO_2-eq (vertical PBR), 216 kg CO_2-eq (horizontal PBR) and 256 kg CO_2-eq (ORP) per kg of microalgal biomass (DW). The production of 1 kg of PHB from microalgal biomass would therefore yield GHG emissions of 360-430 kg of CO_2-eq, depending on the cultivation system. Those values are 6-7 times higher compared to PHB from cyanobacteria cultivated in the Z medium. Regarding aquaculture water and liquid digestate, GHG emissions of PHB from microalgal biomass are 2-3 times higher.

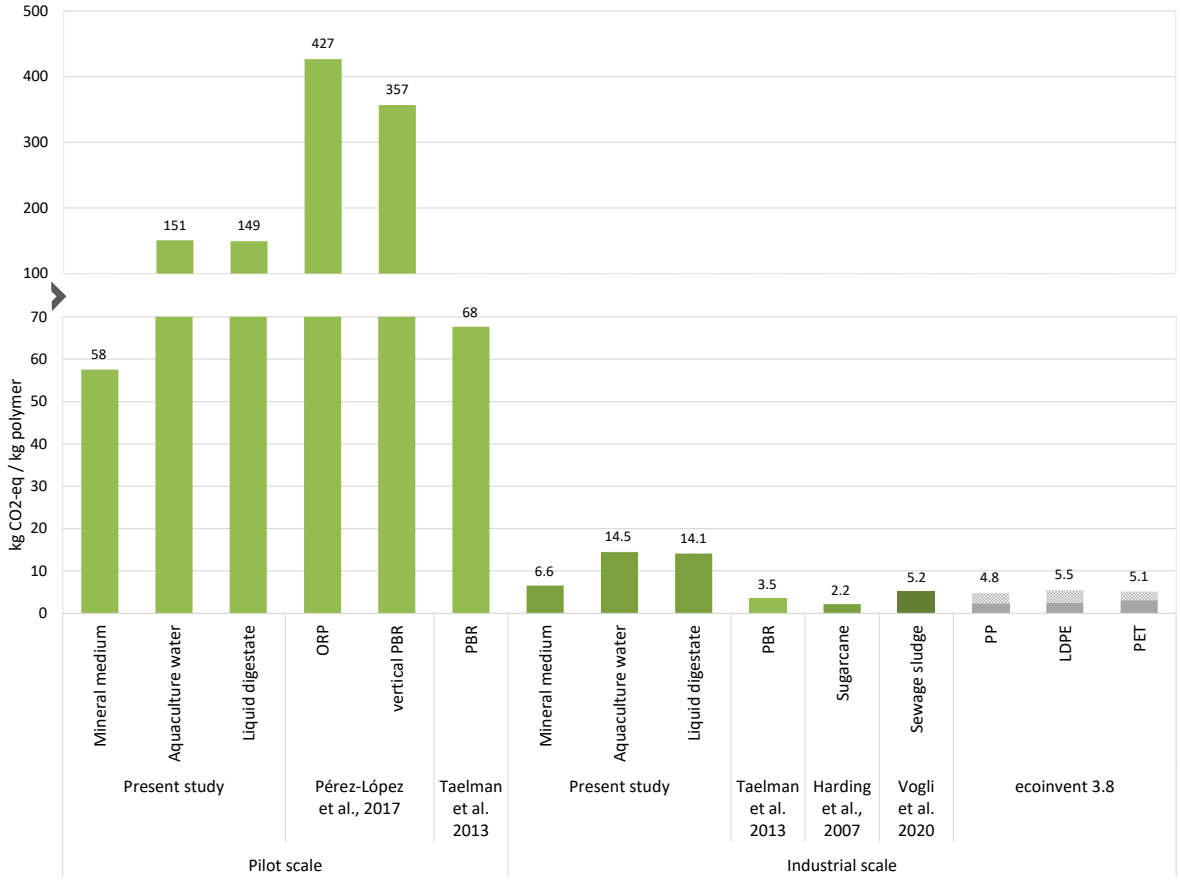

Figure 5 **Comparison of GHG emissions of 1 kg polymer from the present study to results from literature. Outcomes are depicted for pilot-scale as well as industrial-scale values. For PHB from biomass, a PHB content of 60 % was consistently assumed. PHB produced from cyanobacteria or algae biomass are marked in light green. Dark green represents PHB produced from biomass other than algae. Petroleum-based polymers are shown in grey (dark grey: production, light grey: disposal).**

The overall main environmental burden in the production of the algal biomass is electricity production for cultivation. A primary difference compared to the cyanobacteria cultivation in the present study, is that in the study by Pérez-López et al. (2017) heating and cooling during algae cultivation was used. For algae cultivation in summer, heating and cooling requires between 56 % and 62 % of the electricity consumption during cultivation. This main difference, and the energy-intensiveness of heating and cooling therefore largely explains the higher GHG emissions of the microalgae production, compared to the cyanobacteria production. This difference is also reflected in the non-renewable energy (NRE) demand, with microalgae production requiring 5-6 GJ for the production of 1 kg PHB, while in the present study 0.6-1.5 GJ of fossil energy are required per kg of PHB.

Another study on algae production by Taelman et al. (2013) describes, similar to the present study, a pilot-scale scenario as well as an extrapolated industrial-scale scenario. The pilot-scale scenario describes a PBR with an areal productivity of 4.59 g m^{-2} d^{-1}, which is comparable to that of the cyanobacteria production in the Z medium (4.76 g m^{-2} d^{-1}). When converted to the functional unit of 1 kg PHB, a total of 68 kg of CO_2-eq are emitted. GHG emissions are thus 15 % higher compared to the PHB production from cyanobacteria in the Z medium. Largest contribution to GHG emissions in the study by Taelman et al. (2013) derives from the electricity consumption of a freeze dryer, used for the drying of the algal biomass (55 % of total impact), and the electricity consumption of a fan, used for the supply of CO_2 (22 % of total impact). Neither a freeze dryer nor a fan is used in the PHB production from cyanobacteria and electricity consumption has a considerably smaller contribution to total GHG emissions (8 %). In contrast, the CO_2 input represents the main contributor to total GHG emissions in the cyanobacteria production (45 %), while in the microalgae production only 4 % of total impacts are due to the CO_2 input. This difference can primarily be explained by the difference in CO_2 supplied to the system, being much higher in the cyanobacteria cultivation with 2.1 kg per day, compared to 0.1 kg of CO_2 per day supplied to the microalgae cultivation. For the industrial-scale scenario, Taelman and colleagues assumed an areal productivity of 15.1 g m^{-2} d^{-1}, which is slightly lower compared to the 20 g m^{-2} d^{-1} assumed for the scaled cyanobacteria production in the Z medium. The industrial-scale scenario results in 3.5 kg of CO_2-eq emitted per kg of PHB, with main contributors being electricity for drying, nutrient production and electricity to operate the fan. GHG emissions are therefore 2 times lower than the upscaled cyanobacteria production in the Z medium. This difference is mainly due to a higher energy demand for operation of the system (mainly pumps), with NRE demand amounting to 75 MJ per kg PHB in the Z medium of the present study, compared to 48 MJ in the microalgae production.

Concerning the industrial-scale production of PHB from biomass other than algae, Harding et al. (2007) investigated PHB produced from sugarcane. GHG emissions per kg of PHB amount to 2.2 kg CO_2-eq.

This value is lower than values found for the petroleum-based polymers PP, LDPE and PET (4.8 kg CO_2-eq, 5.5 kg CO_2-eq and 5.1 kg CO_2-eq, respectively). Regarding the industrial-scale scenario of the present study, GHG emissions are 3 times higher for PHB from cyanobacteria produced in the Z medium, compared to PHB from sugarcane. Primary reason for this difference is the NRE demand, which is 1.6-3.7 times higher in the present study, with 75-175 MJ per kg PHB, compared to 48 MJ per kg PHB from sugarcane. The comparatively high NRE demand for PHB produced from cyanobacteria therefore largely explains the higher impacts for the industrial-scale scenario of the present study.

Vogli et al. (2020) performed an LCA of PHA produced by microbial cultures using anaerobically digested sewage sludge as feedstock. The authors extrapolated, similar as in the present study, primary pilot-scale data to an industrial scale scenario. Per kg of PHA produced, the GHG emissions amount to 5.2 kg CO_2-eq. This value is comparable to the industrial-scale Z medium scenario of the present study of 6.6 kg CO_2-eq. Opposed to the present study, Vogli and colleagues additionally included the PHA extraction process, which accounts for roughly 10 % of the overall GHG emissions. Energy consumption of PHA from sewage sludge amounts to 70 MJ per kg of PHB, which is slightly lower compared to the 75 MJ for PHB from cyanobacteria in the Z medium. The higher energy consumption, along with higher impacts from CO_2 input and production of infrastructure in the present study likely explains the slightly higher overall GHG emissions per kg of polymer.

5.2 UNCERTAINTIES AND DATA QUALITY

Generally, the data on the cyanobacteria production on the baseline scenario is of high quality, as it stems from first-hand experiments from a thin-layer PBR pilot plant at the Zurich University of Applied Sciences. The production of materials used for the construction and operation of the pilot plant is based on background data from the ecoinvent database. An estimation concerning specific data was made for the direct emissions during cyanobacteria cultivation. The loss of nitrogen during the cultivation of cyanobacteria is scarcely researched and the assumed losses therefore remain assumptions.

The data regarding the industrial-scale scenario is based on an extrapolation of the pilot plant data. The two parameters, CO_2 input and areal productivity were artificially modified. The values for those parameters are thus theoretical. However, since they are based on justified indications found in literature, they are considered as realistic.

An uncertainty in the data further regards the yearly production volume of the pilot plant. Indicated yields refer to maximum values measured during midsummer. Production volumes during spring and

fall were found to be 30-50 % lower compared to summer[2]. It therefore needs to be stated that yearly production in the geography of Switzerland would accordingly be lower in reality. The production volumes used in the present study might refer to a geography with a longer summer (e.g. Spain). Regarding the two wastewater media, yields are generally assumed to be underestimated. Primary data was obtained from early-stage experiments, where productivity of cyanobacteria in the wastewater media were limited by either nutrient supply or light (Mariotto et al., 2022). Productivity rates with the wastewater media are therefore expected to increase in the future.

5.3 Sensitivity Analysis

5.3.1 Harmonised yield between cultivation media

In order to assess the total environmental impact of the cyanobacteria production in the three cultivation media while disregarding the differences in productivity, the yield was harmonised between the cultivation media. The annual yield of the cyanobacteria cultivation in the Z medium was applied to both wastewater media, representing 18 kg of cyanobacteria biomass for the pilot scale and 42 tonnes for the industrial scale (see Table 5).

When assuming equal yields across all media, the cyanobacteria production in the aquaculture water shows the lowest total environmental impact on both pilot and industrial scale, followed by the liquid digestate and Z medium, as shown in Figure 6. These results are in contrast to the results in chapter 4, confirming the fact that the lower environmental impacts of the Z medium are solely related to the higher yield. Compared to the Z medium, the total environmental impact of the cyanobacteria production in the liquid digestate is 3 % (pilot scale) and 5 % (industrial scale) lower, while impacts are 7 % and 10 % lower in the aquaculture water.

Reason for the higher total environmental impact of the Z medium is primarily the sodium nitrate input, which is absent in both wastewater media. The slightly higher impacts of the liquid digestate compared to the aquaculture water can be explained by the direct emissions of NH_3 that occur during cyanobacteria cultivation in the liquid digestate, along with a slightly lower electricity consumption.

The scenario thus shows that if yields of the same magnitude as in the Z medium should be achieved with the wastewater media, cyanobacteria cultivation in aquaculture water would be the advisable choice of medium, regarding the total environmental impact.

[2] Marina Mariotto, questionnaire, 19 November 2021, Institute of Natural Resource Sciences, ZHAW

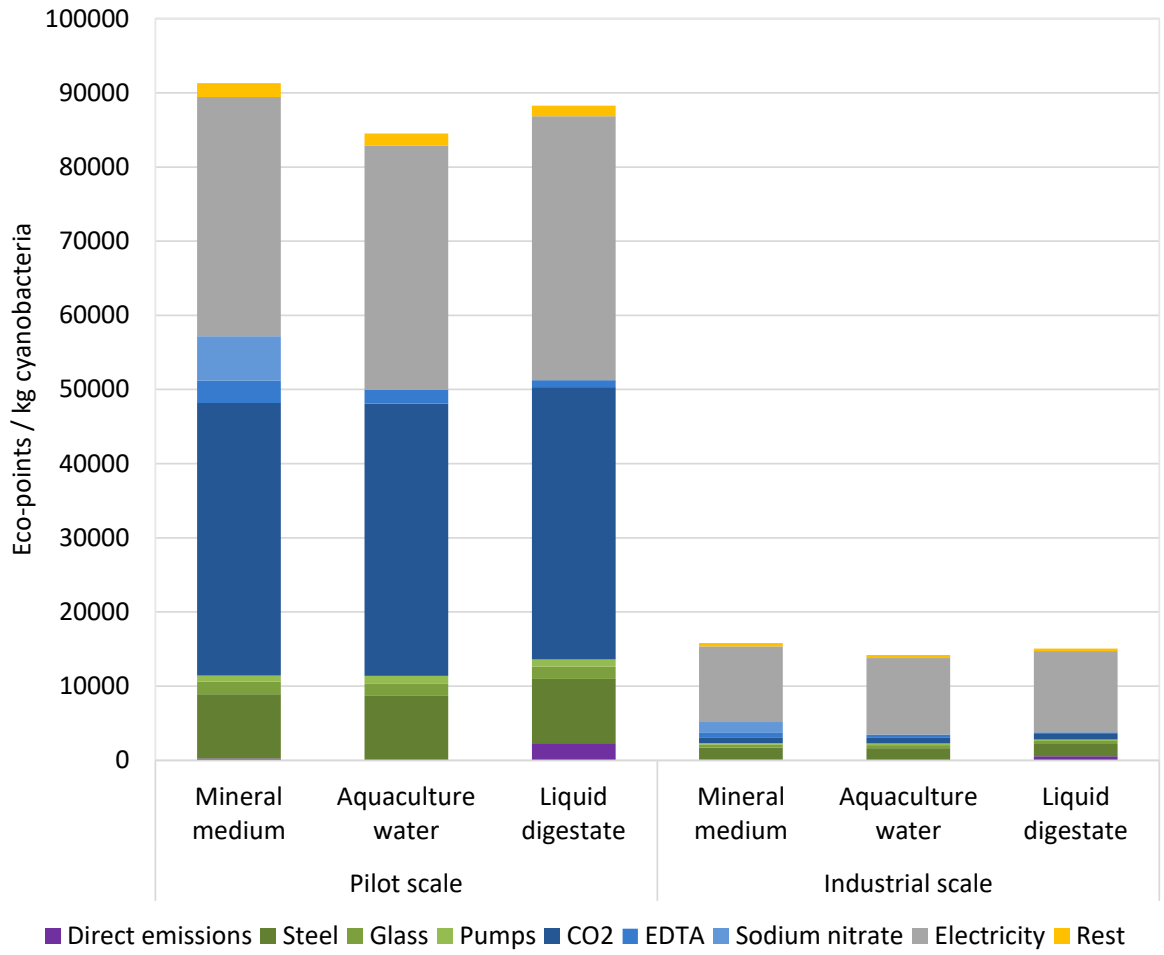

Figure 6 Total environmental impact according to ecological scarcity 2021 (Frischknecht et al., 2021) per kilogram of cyanobacteria, with a harmonised yield between the three cultivation media. Results are shown for the pilot and industrial scale. Impacts are divided into direct emissions (purple), infrastructure of the plant (green shades), operation of the plant (blue shades) and rest / remaining processes (yellow).

5.3.2 Avoided emissions and wastewater treatment

Cyanobacteria cultivation in aquaculture water has the beneficial side-effect that the aquaculture water is not discharged into natural waters or that wastewater treatment can be avoided. To provide context on how relevant this beneficial side effect is, an additional analysis is carried out regarding the associated avoided environmental impacts. One scenario reflects an avoided nitrate emission in the case of discharging the aquaculture water into natural waters, along with a second scenario considering additional emissions next to nitrate. The third scenario reflects an avoided wastewater treatment in the case of treating the aquaculture water in a wastewater treatment plant.

Results are indicated for the industrial scale scenario and regarding the total environmental impact, as shown in Figure 7. The avoided nitrate emission, in case of the direct discharge of low-polluted wastewater, results in an avoided environmental impact of 510 eco-points per kg of cyanobacteria. This value represents 1.2 % of the overall environmental impact of cyanobacteria production in the aquaculture water on industrial scale. The avoided environmental impact of discharging higher-polluted fish wastewater amounts to 1'600 eco-points, representing 3.8 % of the total environmental impact. Finally, regarding the avoided wastewater treatment, total environmental impact is reduced by 1'400 eco-points or 3.3 % of the overall environmental impact.

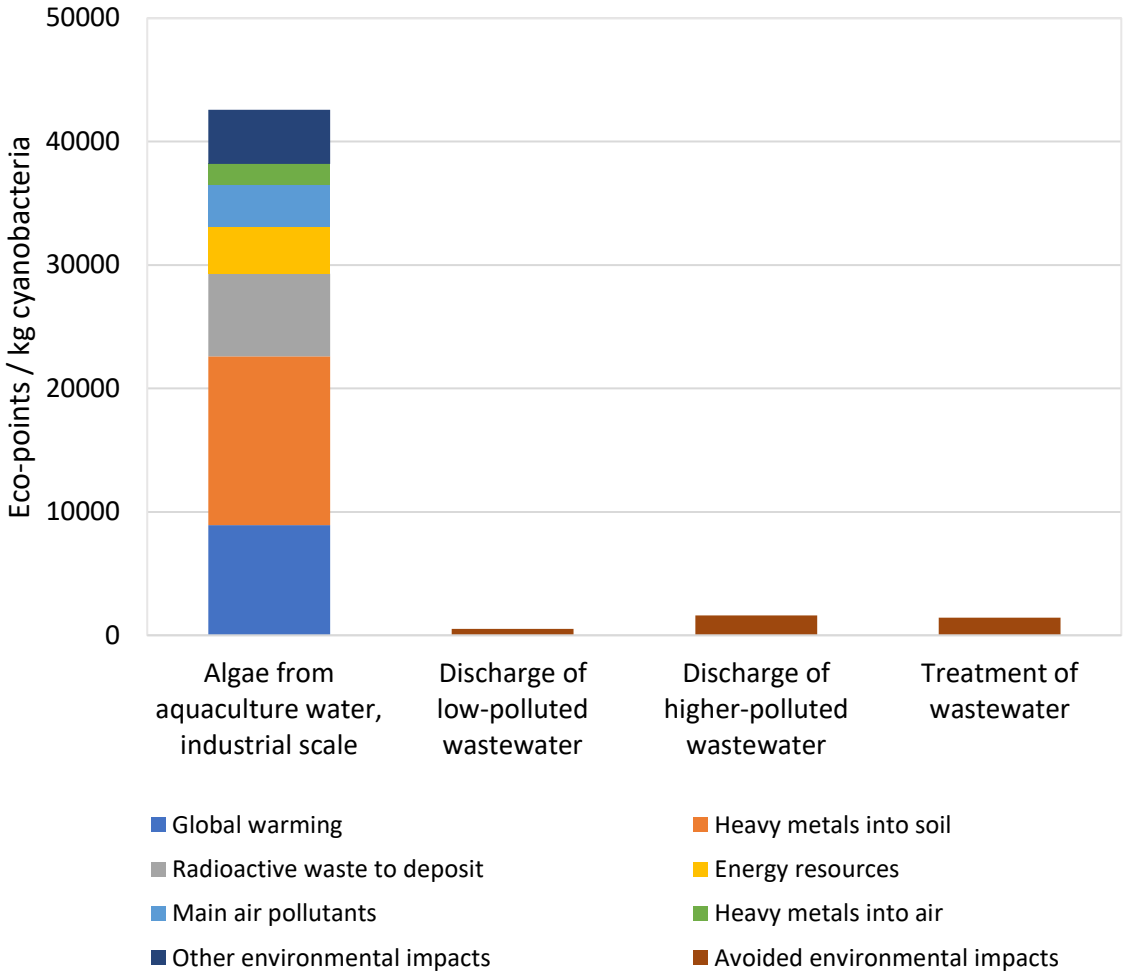

Figure 7 **Total environmental impact according to ecological scarcity 2021 (Frischknecht et al., 2021) per kilogram of cyanobacteria cultivated in aquaculture wastewater on industrial scale. Scenarios shown are the baseline scenario (industrial scale), direct discharge of low- and higher-polluted wastewater and treatment of wastewater. Impacts are divided into the dominating environmental impacts and avoided environmental impacts (brown).**

Through treatment of the aquaculture water on the PBR, total environmental impact can thus be reduced by 1.2-3.8 %, considering that the aquaculture water would otherwise be directly discharged into natural waters or treated in a wastewater treatment plant.

5.4 Conclusion & Recommendations

The results of the present study on the environmental impacts of PHB production from cyanobacteria identified two major environmental hotspots for the pilot-scale production. Primary environmental hotspot for a majority of impact categories analysed was found to be the CO_2 input, due to the energy consumption and direct emissions during its production. A second environmental hotspot for a number of impact categories was found to be the electricity consumption of the pumps and centrifugation. Finally, the production of the steel used for the scaffolding of the PBR showed a relevant contribution to a majority of impact categories analysed. Cyanobacteria cultivation using wastewater streams consistently showed higher environmental impact compared to the cultivation in the Z medium, due to the considerably lower biomass yields and areal productivity rates. Results of the sensitivity analysis showed that if equal yields between the cultivation media could be achieved, the most environmentally friendly option would change from Z medium to aquaculture water. Regarding the aquaculture water, environmental impacts could additionally further be reduced by up to 4 %, when considering avoided environmental impacts of wastewater disposal.

Extrapolating the data from the pilot plant to an industrial-scale production scenario and reducing the CO_2 input, showed a reduction potential of 40-42 % of the total environmental impact, across all three cultivation media. An increase in the productivity rate and therefore a higher yield showed an even higher reduction of the total environmental impact of 74 % in all media, compared to cyanobacteria production on the pilot plant. Combining the decreased CO_2 input with an increased productivity rate yields a reduction of the total environmental impact of 83 % for the cyanobacteria production in all three media. This scaled scenario with adjustment of the CO_2 input and productivity rates reveals the electricity consumption as the distinct, primary hotspot of the total environmental impact, as well as the majority of environmental impact categories analysed.

The present study thus shows, that from an environmental perspective, PHB production from cyanobacteria using waste streams would have to achieve an increase of productivity rates by a factor 3, if it were to compete with production using a Z medium. Regardless of the medium used, focus for reducing the environmental impact of PHB production from cyanobacteria should lie in reducing the CO_2 input, while aiming to maximise the areal productivity rates of the cyanobacteria cultivation in a

first step. In a second step, in order to compete with PHB produced from other feedstock as well as petroleum-based polymers on an industrial scale, electricity consumption of the cyanobacteria production needs to be reduced by about 50-60 %.

Certain parameters of PHB production from cyanobacteria thus require considerable adjustment and environmental improvement, in order to compete with the bio-based as well as petroleum-based alternatives on an industrial scale. Along with this insight it is important to understand that this technology is yet in its beginning stages. With its further development, production processes can be expected to continuously grow more efficient, while focusing on the improvement possibilities highlighted in this study. Overcoming these challenges can move this technology into the centre of biopolymers, which will continue to be an increasingly urgent need in the wake of advancing depletion of fossil resources, food competition and climate change.

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APPENDIX I: INVENTORY PILOT PLANT

Table 7 depicts all inputs and outputs calculated for the modelling of the cyanobacteria production on the pilot plant. All values refer to 0.9 kg of cyanobacteria (DW), due to a 10 % loss of biomass during centrifugation. Steel, glass, copper and synthetic rubber represent the construction materials of the pilot plant. Sodium hydroxide, phosphoric acid, non-ionic surfactant, hydrogen peroxide and acetic acid are the chemicals used for the disinfection of the plant. Further indicated is the number of pumps needed for the operation of the plant. For the Z medium, solely the reactor pump is needed. For cyanobacteria cultivation in aquaculture water and liquid digestate, a nutrient pump is additionally required. Pump power is 50 W (aquaculture water) and 30 W (liquid digestate), and calculation is according to the reactor pump. The remaining inputs in the materials section describe the nutrients added to the medium. Amounts of nutrients are listed in Table 6. Electricity low voltage reflects the total electricity demand of the bioreactor, including pumping system, heating rods, which are used for the disinfection of the plant, and the centrifuge. Emissions include, as described in chapter 3, NH_3, NO_3^- and NH_4^+ emissions from N losses of the nutrient input. As N_2 emissions to the air are negligible they were not modelled. Waste disposal of input materials solely includes synthetic rubber. All other materials are assumed to be recycled.

Table 6 Nutrients added to / contained in cultivation medium. Amounts are indicated in kg per year.

Nutrient	Unit	Z medium	Aquaculture water	Liquid digestate
N in medium	kg/a	-	0.44	1.25
$NaNO_3$	kg/a	14.20	-	-
$Ca(NO_3)_2$	kg/a	1.25	0.75	0.37
K_2HPO_4	kg/a	0.94	0.57	0.28
$MgSO_4$	kg/a	0.37	0.22	0.11
$NaCO_3$	kg/a	0.64	0.38	0.19
Micronutrient solution	kg/a	6.08	3.65	1.83
FeEDTA	kg/a	2.43	1.46	0.73

Table 7 Inventory of the cyanobacteria production on the pilot plant. In green, the resulting processes are shown and in yellow all in- and outputs from and to the technosphere as well as emissions. Values of the in- and outputs for 0.9 kg of cyanobacteria are shown in blue. All data, unless stated otherwise, refer to the questionnaire by Marina Mariotto (received on 19.11.21).

	Name	Unit	Cyanobacteria production, Z medium	Cyanobacteria production, aquaculture water	Cyanobacteria production, liquid digestate	Remarks
	Location		CH	CH	CH	
	Infrastructure Process		no	no	no	
	Unit		kg	kg	kg	
Production	Cyanobacteria production, harvested from ZHAW pilot plant, Z medium | Biomat	kg	0.9			cyanobacteria production in a Z medium, harvested from the ZHAW pilot plant, 0.9 kg as reference due to 10% biomass loss during centrifugation
	Cyanobacteria production, harvested from ZHAW pilot plant, aquaculture water | Biomat	kg		0.9		cyanobacteria production with aquaculture water, harvested from the ZHAW pilot plant, 0.9 kg as reference due to 10% biomass loss during centrifugation
	Cyanobacteria production, harvested from ZHAW pilot plant, liquid digestate | Biomat	kg			0.9	cyanobacteria production with liquid digestate, harvested from the ZHAW pilot plant, 0.9 kg as reference due to 10% biomass loss during centrifugation
Materials	Steel, chromium steel 18/8 {GLO}| market for | Cut-off, U	kg	5.04E-01	1.51E+00	1.51E+00	Amount of steel needed for the production of 1kg of cyanobacteria; 180kg + 50kg (total amount of steel) / 456kg or 152kg (total production volume over lifetime of the reactor)
	flat glass, uncoated {RER}| market for flat glass, uncoated | Cut-off, U	kg	9.86E-01	2.96E+00	2.96E+00	Amount of glass needed for the production of 1kg of cyanobacteria; 450kg (total amount of glass) / 456kg or 152kg (total production volume over lifetime of the reactor)
	Copper {GLO}| market for | Cut-off, U	kg	1.10E-03	3.29E-03	3.29E-03	Amount of copper needed for the production of 1kg of cyanobacteria; 0.5kg (total amount of copper) / 456kg or 152kg (total production volume over lifetime of the reactor)
	Wire drawing, copper {RER}| processing | Cut-off, U	kg	1.10E-03	3.29E-03	3.29E-03	Amount of copper processed to wires
	Synthetic rubber {GLO}| market for | Cut-off, U	kg	5.48E-02	1.64E-01	1.64E-01	Amount of silicone needed for the production of 1kg of cyanobacteria; 5kg (total amount of silicone) / 91.3kg or 30.4kg (total production volume over lifetime of the silicone)
	Neutralising agent, sodium hydroxide-equivalent {GLO}| market for | Cut-off, U	kg	4.32E-02	1.29E-01	1.29E-01	Amount of sodium hydroxide used for desinfection, during the production of 1kg cyanobacteria; 2.25kg (amount of Pasteurreiniger used/a) * 0.35 (share of sodium hydroxide in Pasteurreiniger) / 18.25kg or 6.08kg (cyanobacteria production volume/a)
	phosphoric acid, fertiliser grade, without water, in 70% solution state {RER}| phosphoric acid production, dihydrate process | Cut-off, U	kg	2.47E-02	7.40E-02	7.40E-02	Amount of phosphoric acid used for desinfection; 2.25kg (amount of Halacid P used/a) * 0.2 (share in Halacid P) / 18.25kg or 6.08kg
	Non-ionic surfactant {GLO}| market for non-ionic surfactant | Cut-off, U	kg	1.85E-02	5.55E-02	5.55E-02	Amount of non-ionic surfactant used for desinfection; 2.25kg (amount of Halacid P used/a) * 0.15 (share in Halacid P) / 18.25kg or 6.08kg
	Hydrogen peroxide, without water, in 50% solution state {RER}| market for hydrogen peroxide, without water, in 50% solution state | Cut-off, U	kg	3.70E-02	1.11E-01	1.11E-01	Amount of hydrogen peroxide used for desinfection; 2.25kg (amount of Halades PE used/a) * 0.3 (share in Halades PE) / 18.25kg or 6.08kg
	Acetic acid, without water, in 98% solution state {GLO}| market for | Cut-off, U	kg	2.47E-02	7.40E-02	7.40E-02	Amount of acetic acid used for desinfection; 2.25kg (amount of Halades PE used/a) * 0.2 (share in Halades PE) / 18.25kg or 6.08kg
	Carbon dioxide, liquid {RER}| market for | Cut-off, U	kg	2.50E+01	7.50E+01	7.50E+01	Amount of CO2 used for production of 1kg cyanobacteria; 456.25kg (amount of CO2/a) / 18.25kg or 6.08kg
	Pump, 40W {CH}| production | Cut-off, U	p	1.71E-02	6.16E-02	5.75E-02	Reactor pump: 250W/40 (number of 40W pumps needed across lifetime) / 365kg or 121.6kg (total production volume over lifetime of pump)
	Sodium nitrate {GLO}| market for | Cut-off, U	kg	7.78E-01	-	-	Amount of Sodium nitrate needed for production of 1kg cyanobacteria; 14.2kg (amount of NaNO3/a) / 18.25kg
	calcium nitrate {RER}| market for calcium nitrate | Cut-off, U	kg	6.83E-02	1.23E-01	6.15E-02	Amount of calcium nitrate used for production of 1kg cyanobacteria; amount of Ca(NO3)2 / cyanobacteria production volume/a
	Potassium hydroxide {GLO}| market for | Cut-off, U	kg	5.17E-02	9.30E-02	4.65E-02	Amount of potassium hydroxide used for production of 1kg cyanobacteria; amount of K2HPO4 / cyanobacteria production volume/a
	Magnesium sulfate {GLO}| market for | Cut-off, U	kg	2.03E-02	3.66E-02	1.83E-02	Amount of magnesium sulfate used for production of 1kg cyanobacteria; amount of MgSO4 / cyanobacteria production volume/a
	Soda ash, dense {GLO}| market for | Cut-off, U	kg	3.50E-02	6.30E-02	3.15E-02	Amount of sodium carbonate used for production of 1kg cyanobacteria; amount of NaCO3 / cyanobacteria production volume/a
	EDTA, ethylenediaminetetraacetic acid {GLO}| market for | Cut-off, U	kg	3.33E-01	6.00E-01	3.00E-01	Amount of FeEDTA used for production of 1kg cyanobacteria; amount of FeEDTA / cyanobacteria production volume/a
	Boric acid, anhydrous, powder {GLO}| market for | Cut-off, U	kg	6.67E-04	1.20E-03	6.00E-04	Amount of micronutrientsolution for production of 1kg cyanobacteria; amount of solution per yea * 0.005 (share of boric acid) / cyanobacteria production volume/a
	Manganese sulfate {GLO}| market for | Cut-off, U	kg	2.67E-04	4.80E-04	2.40E-04	Amount of micronutrientsolution for production of 1kg cyanobacteria; amount of solution per yea * 0.002 (share of magnese sulfate) / cyanobacteria production volume/a
Electricity / Heat / Fuels	Electricity, low voltage {CH}| market for | Cut-off, U	kWh	8.14E+01	2.49E+02	2.69E+02	Electricity demand of reactor pump for production of 1kg cyanobacteria; 1277.5 kWh (energy demand/a) / 18.25kg or 6.08kg
Emissions	Ammonia	kg	-	-	1.29E-01	Amount of NH3 emissions for production of 1kg cyanobacteria; NH3 emissions per year / cyanobacteria production volume/a
	Nitrate	kg	2.97E-02	1.35E-02	-	Amount of NO3 emissions for production of 1kg cyanobacteria; NO3 emissions per year / cyanobacteria production volume/a
	Ammonium, ion	kg	-	-	2.10E-02	Amount of NH4 emissions for production of 1kg cyanobacteria; NH4 emissions per year / cyanobacteria production volume/a
Waste	Synthetic rubber {GLO}| market for | Cut-off, U	kg	5.48E-02	1.64E-01	1.64E-01	Disposal of silicone

APPENDIX II: INVENTORY INDUSTRIAL SCALE

Table 8 depicts all inputs and outputs calculated for the modelling of the cyanobacteria production for the upscaling of the pilot plant to industrial scale. The first three columns refer to a 1:1 upscaling of the pilot plant, considering the same areal productivity rate. The last three columns depict the cyanobacteria production considering an increased areal productivity rate (yield increase). All values refer to 0.9 kg of cyanobacteria (DW), due to a 10 % loss of biomass during centrifugation.

Values regarding the 1:1 upscaling of the pilot plant refer to a surface of the PBR of 9863 m^2. Yearly production volumes on a plant of this surface, considering an areal productivity rate equal to that of the pilot plant, reaches 10 tonnes for the Z medium and 3.34 tonnes for aquaculture water and liquid digestate. All values were consequently calculated based on an areal upsize factor, calculated by the size increase from the pilot plant (18 m^2) to the upscaled bioreactor (9863 m^2). For an unmodified yield, this results in an upscale factor of 547.95. All input and output parameters of the cyanobacteria production on the pilot plant were accordingly multiplied with this upscale factor and divided by the adjusted yearly production volume. Values for all input and output parameters therefore remain unchanged, with the exception of steel. In the pilot plant scenario, the centrifuge is in operation for 10 hours across 14 days. In case of an upscaling, operation hours of the centrifuge would be increased to 24 h/d for means of efficiency and saving of resources. This thus results in a smaller number of required centrifuges and reduces the amount of steel needed.

When adjusting the areal productivity rate to those indicated in Table 4, the annual production volume of cyanobacteria increases to 42 tonnes in the Z medium, and to 14 tonnes in the other two media. For the adjusted yield, the input and output parameters were therefore divided by 42 tonnes instead of 10 tonnes and 14 tonnes instead of 3.34 tonnes (see Table 8, “Remarks” column for detailed calculations). For the parameter CO_2 input, 3 different values were calculated, with the first one being the adjusted yield calculation as described above. The second value reflects the CO_2 input considering an increase of the CO_2 uptake rate by the cyanobacteria biomass from 6 % (pilot plant scenario) to 70 % (Z medium) and 23.34 % (aquaculture water and liquid digestate). The third value reflects the CO_2 input considering both the yield increase and the increase of the CO_2 uptake rate combined.

Table 8 Inventory of the cyanobacteria production for the upscaling of the pilot plant to industrial scale. All values refer to an increased areal productivity rate. All data, unless stated otherwise, refer to the questionnaire by Marina Mariotto (received on 19.11.21).

	Name	Unit	Cyanobacteria production, upscale, Z medium	Cyanobacteria production, upscale, aquaculture water	Cyanobacteria production, upscale, liquid digestate	Z medium, increased yield	Aquaculture water, increased yield	Liquid digestate, increased yield	Remarks
	Location		CH	CH	CH	CH	CH	CH	
	Infrastructure Process		no	no	no	no	no	no	
	Unit		kg	kg	kg	kg	kg	kg	
Production	Cyanobacteria production, upscale, Z medium | Biomat	kg	0.9						Upscaled cyanobacteria production in a Z medium, harvested from the ZHAW pilot plant, 0.9 kg as reference due to 10% biomass loss during centrifugation
	Cyanobacteria production, upscale, aquaculture water | Biomat	kg		0.9					Upscaled cyanobacteria production with aquaculture water, harvested from the ZHAW pilot plant, 0.9 kg as reference due to 10% biomass loss during centrifugation
	Cyanobacteria production, upscale, liquid digestate | Biomat	kg			0.9				Upscaled cyanobacteria production with liquid digestate, harvested from the ZHAW pilot plant, 0.9 kg as reference due to 10% biomass loss during centrifugation
	Cyanobacteria production, upscale, increased yield, Z medium | Biomat	kg				0.9			Upscaled cyanobacteria production in a Z medium with increased yield, harvested from the ZHAW pilot plant, 0.9 kg as reference due to 10% biomass loss during centrifugation
	Cyanobacteria production, upscale, increased yield, aquaculture water | Biomat	kg					0.9		Upscaled cyanobacteria production with aquaculture water with increased yield, harvested from the ZHAW pilot plant, 0.9 kg as reference due to 10% biomass loss during centrifugation
	Cyanobacteria production, upscale, increased yield, liquid digestate | Biomat	kg						0.9	Upscaled cyanobacteria production with liquid digestate with increased yield, harvested from the ZHAW pilot plant, 0.9 kg as reference due to 10% biomass loss during centrifugation
Materials	Steel, chromium steel 18/8, hot rolled {RER}| production | Cut-off, U	kg	3.97E-01	1.19E+00	1.19E+00	9.59E-02	2.88E-01	2.88E-01	Amount of steel for pilot plant * upscale factor + amount of steel for centrifuge / total production volume over lifetime of the reactor
	flat glass, uncoated {RER}| market for flat glass, uncoated | Cut-off, U	kg	9.86E-01	2.96E+00	2.96E+00	2.35E-01	7.05E-01	7.05E-01	Amount of glass needed; 450kg (total amount of glass) * upscale factor / total production volume over lifetime of the reactor
	Copper {GLO}| market for | Cut-off, U	kg	1.10E-03	3.29E-03	3.29E-03	2.61E-04	7.83E-04	7.83E-04	Amount of copper needed; 0.5kg (total amount of copper) * upscale factor / total production volume over lifetime of reactor
	Wire drawing, copper {RER}| processing | Cut-off, U	kg	1.10E-03	3.29E-03	3.29E-03	2.61E-04	7.83E-04	7.83E-04	Amount of copper processed to wires
	Synthetic rubber {GLO}| market for | Cut-off, U	kg	5.48E-02	1.64E-01	1.64E-01	1.30E-02	3.91E-02	3.91E-02	Amount of silicone needed; 5kg (total amount of silicone) * upscale factor / 50000kg (total production volume over lifetime of silicone)
	Neutralising agent, sodium hydroxide-equivalent {GLO}| market for | Cut-off, U	kg	4.32E-02	1.29E-01	1.29E-01	1.03E-02	3.08E-02	3.08E-02	Amount of sodium hydroxide used for desinfection, in pilot plant; 2.25kg (amount of Pasteurreiniger used/a) * 0.35 (share of sodium hydroxide in Pasteurreiniger) * upscale factor / 42t or 14t (cyanobacteria production volume/a)
	phosphoric acid, fertiliser grade, without water, in 70% solution state {RER}| phosphoric acid production, dihydrate process | Cut-off, U	kg	2.47E-02	7.40E-02	7.40E-02	5.87E-03	1.76E-02	1.76E-02	Amount of phosphoric acid used for desinfection in pilot plant; 2.25kg (amount of Halacid P used/a) * 0.2 (share in Halacid P) * upscale factor / 42t or 14t
	Non-ionic surfactant {GLO}| market for non-ionic surfactant | Cut-off, U	kg	1.85E-02	5.55E-02	5.55E-02	4.40E-03	1.32E-02	1.32E-02	Amount of non-ionic surfactant used for desinfection; 2.25kg (amount of Halacid P used/a) * 0.15 (share in Halacid P) * upscale factor / 42t or 14t
	Hydrogen peroxide, without water, in 50% solution state {RER}| market for hydrogen peroxide, without water, in 50% solution state | Cut-off, U	kg	3.70E-02	1.11E-01	1.11E-01	8.81E-03	2.64E-02	2.64E-02	Amount of hydrogen peroxide used for desinfection; 2.25kg (amount of Halades PE used/a) * 0.3 (share in Halades PE) * upscale factor / 42t or 14t
	Acetic acid, without water, in 98% solution state {GLO}| market for | Cut-off, U	kg	2.47E-02	7.40E-02	7.40E-02	5.87E-03	1.76E-02	1.76E-02	Amount of acetic acid used for desinfection; 2.25kg (amount of Halades PE used/a) * 0.2 (share in Halades PE) * upscale factor / 42t or 14t
	Carbon dioxide, liquid {RER}| market for | Cut-off, U	kg	2.50E+01	7.50E+01	7.50E+01	5.95E+00	1.79E+01	1.79E+01	Amount of CO2 used; 456.25kg (amount of CO2/a) * upscale factor / 42t or 14t
	Carbon dioxide, liquid {RER}| market for | Cut-off, U	kg	-	-	-	2.09E+00	6.28E+00	6.28E+00	Amount of CO2 used; 98.3kg (amount of CO2/d) * operativity of reactor / 42t or 14t
	Carbon dioxide, liquid {RER}| market for | Cut-off, U	kg	-	-	-	4.99E-01	1.50E+00	1.50E+00	Amount of CO2 used; 456.25kg (amount of CO2/a) * upscale factor * CO2 factor / 42t or 14t
	Pump, 40W {CH}| production | Cut-off, U	p	1.71E-02	6.16E-02	5.75E-02	4.08E-03	1.47E-02	1.37E-02	Reactor pump: 250W/40 (number of 40W pumps needed across lifetime) * upscale factor / 200000kg (total production volume over lifetime of pump)
	Sodium nitrate {GLO}| market for | Cut-off, U	kg	7.78E-01	-	-	1.85E-01	-	-	Amount of Sodium nitrate added as nutrient for pilot plant; 14.2kg (amount of NaNO3/a) * upscale factor / 42t or 14t
	calcium nitrate {RER}| market for calcium nitrate | Cut-off, U	kg	6.83E-02	1.23E-01	6.15E-02	1.63E-02	2.93E-02	1.46E-02	Amount of calcium nitrate added as nutrient for pilot plant; amount of Ca(NO3)2 * upscale factor / 42t or 14t
	Potassium hydroxide {GLO}| market for | Cut-off, U	kg	5.17E-02	9.30E-02	4.65E-02	1.23E-02	2.21E-02	1.11E-02	Amount of potassium hydroxide added as nutrient for production of 1kg algae; amount of K2HPO4 * upscale factor / 42t or 14t
	Magnesium sulfate {GLO}| market for | Cut-off, U	kg	2.03E-02	3.66E-02	1.83E-02	4.84E-03	8.72E-03	4.36E-03	Amount of magnesium sulfate added as nutrient for pilot plant; amount of MgSO4 * upscale factor / 42t or 14t
	Soda ash, dense {GLO}| market for | Cut-off, U	kg	3.50E-02	6.30E-02	3.15E-02	8.33E-03	1.50E-02	7.50E-03	Amount of sodium carbonate added as nutrient for pilot plant; amount of NaCO3 * upscale / 42t or 14t
	EDTA, ethylenediaminetetraacetic acid {GLO}| market for | Cut-off, U	kg	3.33E-01	6.00E-01	3.00E-01	7.94E-02	1.43E-01	7.14E-02	Amount of FeEDTA used for pilot plant; amount of FeEDTA * upscale factor / 42t or 14t
	Boric acid, anhydrous, powder {GLO}| market for | Cut-off, U	kg	6.67E-04	1.20E-03	6.00E-04	1.59E-04	2.86E-04	1.43E-04	Amount of micronutrientsolution for pilot plant; amount of solution per year * 0.005 (share of boric acid) * upscale factor / 42t or 14t
	Manganese sulfate {GLO}| market for | Cut-off, U	kg	2.67E-04	4.80E-04	2.40E-04	6.35E-05	1.14E-04	5.71E-05	Amount of micronutrientsolution for pilot plant; amount of solution per year * 0.002 (share of magnese sulfate) * upscale factor / 42t or 14t
Electricity / Heat / Fuels	Electricity, low voltage {CH}| market for | Cut-off, U	kWh	8.14E+01	2.49E+02	2.69E+02	2.57E+01	8.95E+01	8.30E+01	Electricity demand of reactor pump for production of 1kg algae; 1077.5 MWh (energy demand/a) / 42t or 14t
Emissions	Ammonia	kg	-	-	1.29E-01	-	-	3.08E-02	Amount of NH3 emissions for production of 1kg cyanobacteria; NH3 emissions per year / cyanobacteria production volume/a
	Nitrate	kg	2.97E-02	1.35E-02	-	7.08E-03	3.22E-03	-	Amount of NO3 emissions for production of 1kg algae; NO3 emissions per year / algae production volume/a
	Ammonium, ion	kg	-	-	2.10E-02	-	-	5.01E-03	Amount of NH4 emissions for production of 1kg algae; NH4 emissions per year / algae production volume/a
Waste	Synthetic rubber {GLO}| market for | Cut-off, U	kg	5.48E-02	1.64E-01	1.64E-01	1.30E-02	3.91E-02	3.91E-02	Disposal of silicone