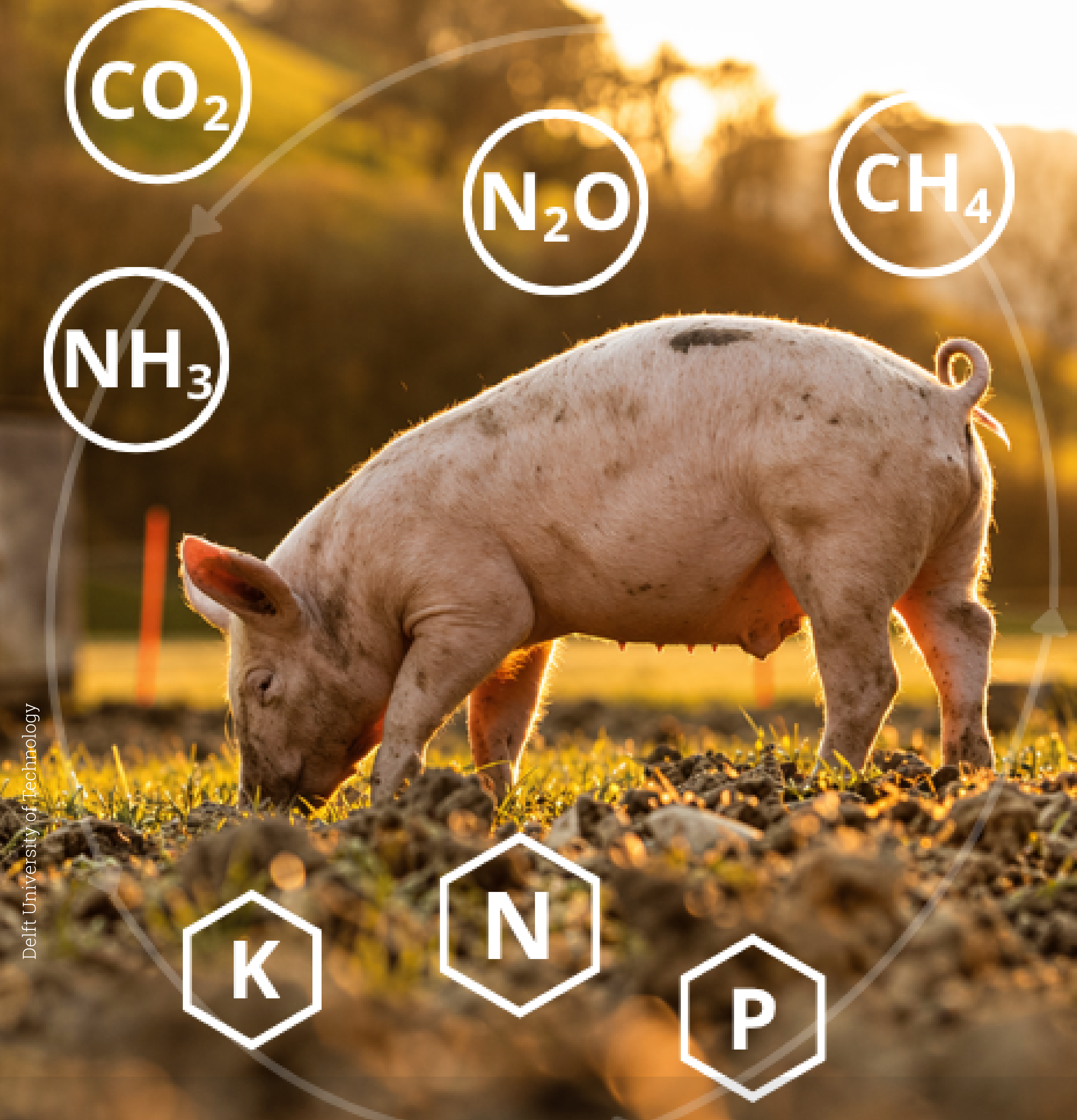


Delft University of Technology

Optimization of swine manure treatment based on environmental and economic parameters to produce a valuable end product

Fleur Ieneke Alida Sneller

Optimization of swine manure treatment based on environmental and economic parameters to produce a valuable end product

by

Fleur Ieneke Alida Sneller

Instructor:	Rebeca Gonzalez Cabaleiro
Teaching Assistant:	Christoff Odendaal
Thesis committee	Rebeca Gonzalez Cabaleiro
	Lotte Asveld
	Yuemei Lin
Project Duration:	September, 2025 - April, 2026
Faculty:	Faculty of Applied Sciences, Delft

TU Delft

Preface

The challenges we face today are, in many ways, linked to how we use materials and energy. From a sustainability perspective, biotechnology has always inspired me, as it shows how microorganisms can make industrial processes more environmentally friendly. By making use of the natural digestion processes of organisms, biology can be applied in controlled systems to achieve desired outcomes, simply by letting the organisms grow and do their work. My interest in this study was sparked by the extensive use of materials on this planet and, more specifically, the potential of materials that are currently considered waste. As fossil fuels are depleting and fertilizers continue to have large environmental impacts, especially visible in the nitrogen crisis in the Netherlands, this topic felt both relevant and urgent. At first glance, these biological processes seem simple and efficient. However, the obvious question remains: why has this manure problem not already been solved? The answer, which also reflects the biggest challenge of this thesis, is that the system is extremely complex. The substrate that we use encounters large variation in nutrient contents and is affected by multiple conditions, making it hard to estimate the constant values. Exploring these possibilities often meant working with situation-specific data and building a deeper understanding of the process, in order to determine which values were appropriate for each specific part of my process. Since this subject was completely new to me, becoming familiar with it was a major part of my thesis. This is also reflected in the extensive literature review included in this work. As the goal to fully solve the problem is rather impossible in a masters thesis, the identification of the factors that have the greatest impact was focused on.

For helping me navigate in the manure treatment world, I would like to thank prof. Sara Gonzalez Garcia and Arsal Tehseen from the University of Santiago for their guidance. They helped me approach manure management from a life cycle assessment perspective, and I am very grateful for their feedback, insights, and for welcoming me in Spain.

I would also like to express my gratitude to dr. ir. Rebeca Gonzalez Cabaleiro and dr. Christoff Odendaal for their support throughout the whole thesis process. Our discussions, your questions, and your guidance were essential in reaching the results I am proud of today. Without both your help, the struggle of navigation though these uncertainties would have been much harder. Parallel to my thesis, I have always found it difficult to see the purpose of my Master's in Life Science and Technology, as it focuses heavily on fundamental research and less on its application. This thesis has helped me understand the value of what I have learned during my studies and has made it clearer to me what I want to pursue in the future.

I want to thank everyone who supported me during these eight months. It has been a challenge for me, given my perfectionism, to navigate through all uncertainties of this complex substrate called manure. The support I received helped me stay focused on what I could contribute, rather than on everything that could still be improved.

At last, this work was made possible through my Bachelor's and Master's studies at the Technical University of Delft, funding provided by FAST funding of the TU Delft, the support of the European Union through the Nutritive Project, and the collaboration with the University of Santiago.

I sincerely hope that this final result reflects the effort I have put into it, and that you enjoy reading it.

Fleur Ieneke Alida Sneller
Delft, July 2026

Abstract

Europe produces approximately 1.4 billion tonnes of manure annually [101]. Of total greenhouse gas (GHG) emissions in Europe, manure management alone contributes around 17% [41]. In this study, liquid swine manure from intensive production systems in Spain is used as the basis for evaluating treatment strategies.

A decentralized, farm-scale system treating 8,500 tonnes of manure per year is evaluated. Two treatment pathways are considered: (i) a biogas production route using anaerobic digestion (AD) with on-site combined heat and power (CHP), followed by solid–liquid separation, composting, and microalgae cultivation for nutrient recovery from the liquid fraction; and (ii) a bioplastic production route via ammonia stripping, followed by acidogenesis for volatile fatty acid (VFA) production, which are subsequently used for microbial polyhydroxyalkanoate (PHA) accumulation, with chloroform–hypochlorite extraction, and downstream composting and algae cultivation of residual streams.

The major nutrient flows, carbon (C), nitrogen (N), phosphorus (P), and potassium (K), are tracked throughout the system to identify which treatment performs best in terms of nutrient recovery, environmental impact reduction, and economic feasibility. A stoichiometric macroscopic mass-balance model was constructed based on an elemental substrate composition derived from Spanish field data ($C_1H_{2.6}O_{0.75}N_{0.054}$, d.o.r =4.9 e^-/Cmol). Environmental impacts were assessed using life cycle assessment (LCA) in SimaPro with the ReCiPe 2016 Midpoint (Hierarchist) method, applying a system expansion approach within a cradle-to-gate system boundary. This allows for a direct comparison of the environmental performance of both processes by substituting for all products. The economic feasibility of both scenarios was evaluated through a techno-economic analysis (TEA), including capital expenditures (CAPEX) and operating expenditures (OPEX). With a discount rate of 3.24%, the net present value (NPV) is calculated. Under base-case yields (30% biodegradability in AD and 54% acidogenesis efficiency), the biogas route produces 2.2 kg of biogas, 25 kg of compost, and 7.2 kg of algae per tonne of manure. The bioplastic route yields 2.9 kg of PHA, 8.9 kg of ammonium sulfate, 36 kg of compost, and 1.2 kg of algae. Nutrient retention favors the bioplastic route for nitrogen (38% vs. 85%), while phosphorus is largely conserved in both systems (≥93%). Based on environmental impact, the biogas route performs better across multiple impact categories, including global warming potential (GWP) (23 vs. 120 kg CO_2-eq per tonne manure), fossil resource scarcity (FPMF), and freshwater eutrophication. However, marine eutrophication is 23% lower in the bioplastic scenario and 22% for terrestrial acidification. Neither scenario is economically viable at this scale, as break-even requires production volumes 18–31 times higher at maximal yields. Only the biogas route achieves a positive gross profit when improvements in yield, product prices, and EU subsidies are considered. In conlclusion, valorization of swine manure is technically (close to) feasible and environmentally beneficial for biogas, and technically feasible but environmentally and economically premature for bioplastics.

Contents

Nomenclature

Abbreviations

Abbreviation	Full term
AD	Anaerobic digestion
ADF	Aerobic dynamic feeding
BMP	Biochemical methane potential
BMP_{th}	Theoretical biochemical methane potential
BOD_5	Biochemical oxygen demand after 5 days
C/N	Carbon to nitrogen ratio
CAPEX	Capital expenditure
CEPCI	Chemical engineering plant cost index
CHP	Combined heat power
COD	Chemical oxygen demand
CSTR	Continuously stirred tank reactor
DFC	Direct fixed capital
DM	Dry matter
DO	Dissolved oxygen
DoR	Degree of reduction
DSP	Downstream processing
FAN	Free ammonia content
FE	Freshwater eutrophication
FISF	Installation scaling factor
FPMF	Fine particulate matter formation
Gdo	Guarantees of origin (Guarantía de origen)
GPM	Gas permeable membrane
GWP	Global warming potential
HHV	Higher heating value
HRT	Hydraulic retention time
IPCC	Intergovernmental panel on climate change
LCA	Life cycle assessment
LCI	Life cycle inventory
LCIA	Life cycle impact assessment
LHV	Lower heating value
ME	Marine eutrophication
MMC	Mixed microbial culture
MO	Organic matter (Materia orgánica)
MW	Molecular weight
NMVOC	Non methane volatile organic compounds
NPV	Net present value
ODM	Organic dry matter
OFMSW	Organic fraction municipal solid waste
OPEX	Operational expenditure
PEC	Purchased equipment cost
PET	Polyethylene terephthalate
PHA	Polyhydroxyalkanoate
PHB	Polyhydroxybutyrate
PHV	Polyhydroxyvalerate
PHBV	Poly(3 hydroxybutyrate co 3 hydroxyvalerate)

Abbreviation	Full term
PDMS	Polydimethylsiloxane
S/L	Solid liquid separation
SBR	Sequencing batch reactor
SLS	Solid liquid separation
SRT	Solids retention time
TA	Terrestrial acidification
TAN	Total ammonium content
TKN	Total kjeldahl nitrogen
TN	Total nitrogen
TS	Total solids
TSP	Total suspended particles
VFA	Volatile fatty acid
VS	Volatile solids

Symbols

Symbol	Definition	Unit
C	Capital cost	[EUR]
CF	Cash flow	[EUR/yr]
D	Depreciation	[EUR/yr]
E_{raw}	Annual raw energy	[kWh/yr]
fs	Safety factor	[dimensionless]
ir	Interest rate	[dimensionless]
k_La	Mass transfer coefficient	[1/h]
L_a	Lang factor	[dimensionless]
m	Mass	[kg]
n	Number of moles	[mol]
n_{yr}	Useful life	[yr]
P_e	Electrical capacity	[kWe]
Q	Volumetric flow rate	[m^3/h]
T_g	Glass transition temperature	[$^{\circ}$C]
V	Volume	[m^3]
x	Volumetric or molar fraction	[dimensionless]
Y	Yield	[mol/mol]
γ	Degree of reduction	[e^-/mol]
η	Efficiency	[%]
η_e	Electrical efficiency	[%]
λ_{cat}	Coupling factor	[dimensionless]
ν	Stoichiometric coefficient	[dimensionless]
ρ	Density	[kg/m^3]

Declaration of generative artificial intelligence use

During this study, AI-based language tools (e.g., ChatGPT by OpenAI and Gemini 3 Flash) were used to improve grammar, clarity and structure of written text. The tool was not used for generating original scientific content, data analysis, or drawing conclusions, but it was used for fixing error in Python. All outputs were critically reviewed and verified by the author. The author maintains full responsibility and accountability for the final content.

Cover image

Cover image: Swine farming nutrient cycle. Generated using ChatGPT (OpenAI, 2026; DALL-E 3), based on an image from [114] and further modified by the author using Canva.

1 Introduction

Livestock farming constitutes a major component of the European agricultural sector, accounting for approximately 40% of its total activity and generating an estimated € 219 billion in annual product value in 2024 in Europe [44, 150]. Besides that, this sector is also associated with significant environmental challenges, largely due to the production of more than 1.4 billion tonnes of manure each year in Europe [101]. Improper manure management contributes substantially to greenhouse gas (GHG) emissions, nutrient losses, and environmental pollution.

In Europe, intensive livestock production is a major source of agricultural GHG emissions, representing 10.8% of total EU emissions [42]. Methane (CH_4) emissions from enteric fermentation and nitrous oxide (N_2O) emissions from agriculture account for the largest fraction, but solely manure management contributes around 17% of the total in itself[41]. Beyond climate impacts, manure mismanagement leads to the release of nutrients and contaminants into the environment. In 2018, agricultural systems emitted approximately 60 million tonnes of nitrogen, of which 35% was released as gaseous emissions (e.g., N_2, N_2O, NO_X, NH_3) and 58% leached into surface and groundwater [95]. Agriculture is responsible for about 93% of total ammonia NH_3 emissions in the EU, contributing to fine particulate matter formation ($PM_{2.5}$), soil acidification, eutrophication, and biodiversity loss [36, 59]. Additionally, the production of synthetic fertilizers contributes approximately 1.75% to total EU emissions, highlighting the potential benefits of recovering nutrients from manure as biofertilizers [64].

Among different livestock types, swine manure presents particular challenges due to its high water content, low carbon-to-nitrogen ratio, and high ammonia concentration. These characteristics make it prone to ammonia emissions, hence causing environmental harm when applied directly on land. As a result, effective treatment strategies are essential to improve nutrient recovery and reduce emissions associated with swine manure management.

High productivity regions are of specific interest, as less intensive systems are generally able to recycle manure within their own land without creating surpluses or environmental risks [136]. For this reason, the geographical focus of this study is Spain, where swine manure production consists of 30% of total manure production of the country and 12% of the European union. Spain contains some of the highest pig densities in Europe, leading to significant manure surpluses that exceed the nutrient absorption capacity of local agricultural land. This imbalance makes direct land application increasingly unviable and contributes to severe soil and water pollution [113]. Swine manure in Spain is mostly obtained in liquid form, leading to a focus of this study on liquid swine manure [98].

In this study, liquid swine manure from intensive production systems in Spain is used as the basis for evaluating treatment strategies. All major nutrient flows, being carbon (C) , nitrogen (N), phosphorus (P), and potassium (K), are tracked throughout the system to assess nutrient recovery efficiency. Two treatment pathways are investigated: conventional anaerobic digestion for biogas production and an alternative process promoting volatile fatty acid (VFA) accumulation for bioplastic (PHA) production. Life Cycle Assessment (LCA) and Techno-Economic Assessment (TEA), including sensitivity analysis on key operational parameters, is used to answer the question: *What treatment strategy for swine* *manure performs best in terms of nutrient recovery, environmental impact reduction, and economic feasibility?* . This is decomposed into (i) which process steps dominate the results, (ii) which of the two scenarios is preferable overall and at last (iii) how yield improvements in the processes affect the overall system performance.

2 Literature review

To properly motivate the process design of this study, a literature review is used to evaluate literature on the treatment of swine manure management. First, characteristics of swine manure are evaluated, secondly applied manure management systems, leading to two treatment options evaluated in complete process requirements.

2.1. Content of manure

Manure of swine is a mixture of faeces (approximately 40%), urine (approximately 60%), water used for cleaning processes, and residual feed. The faecal fraction contains undigested and partially digested organic matter, including fibers, cellulose, and microbial biomass, while urine consists of dissolved organic and inorganic compounds such as nitrogen species, vitamins, and hormones. Due to the addition of water, slurry typically has a high moisture content and is classified as liquid manure when the dry matter content is below 15%, with typical values between 1–8% [100, 76].

The physicochemical composition of pig slurry is highly variable and depends on factors such as animal type, feeding strategy, and management practices. Key parameters include pH, total solids, chemical oxygen demand, and nutrient content (N, P, K). The fate of the nutrient in the manure can determine whether it can be used as fertilizer in a proper way, as the nutrients are widely used in applications on the land to improve crop growth [127, 21].

Nitrogen is present in both organic and inorganic forms, with organic nitrogen referring to nitrogen that is chemically bound within organic compounds, like proteins. A large fraction (up to 75%) of swine manure is however inorganic nitrogen, mainly ammonium, which is readily available but also prone to losses during storage. Over time, the nitrogen content can decrease significantly due to volatilization and microbial processes[100, 87, 107].

Phosphorus in manure is predominantly present in inorganic form, accounting for approximately 74–87% of the total phosphorus content. For swine manure in Spain, phosphorus is primarily found in the suspended or solid fraction of the manure[87, 107].

Potassium is predominantly found in the liquid fraction. The potassium in the manure primarily comes from the swine urine. Around 90% of the total potassium contained in slurry can be diluted in water, thus after solid-liquid separation treatments, the vast majority of the potassium is recovered in this liquid fraction. Diluted potassium is a highly available nutrient that is easily absorbed by plants when applied as a liquid fertilizer [58, 2, 100].

Furthermore, swine manure contains a range of micro-nutrients, including trace elements such as calcium (Ca), magnesium (Mg), sulfur (S), and iron (Fe), which are related to elevated microbial activity and plant growth. However, swine slurry may also contain heavy metals, such as copper (Cu), zinc (Zn), cadmium (Cd), and lead (Pb), originating from feed additives and farm management practices. heavy metals can impose environmental risks when exposed to the soil [76, 87, 100]

As swine manure originates from the intestinal digestion of the swine, it also contains microorganisms, including bacteria, viruses, and pathogens. These organisms can persist after manure management and thus end up in the soil after land application[87, 100].

The water content of manure is measured by drying the manure at $105°C$ to evaporate water, the remaining is described as dry matter (DM) or total solids (TS)[85]. The mass loss determines the water fraction in the manure. According to [107], the average total solids (TS) content of swine manure in Spain ranges between 1–5%, indicating that the majority of the manure consists of water. Based on previous definition, this is classified as liquid slurry.

Reported density values for pig slurry show some variability. Moral et al. [107] reports a density range of 1006–1022 g/L (SD: 8–14), while Kowalski, Makara, and Fijorek [87] indicates a broader range of 900–1100 g/L, with separate measurements of 886–889 g/L for slurry. In addition, [101] reports a density of 1040 g/L for swine manure in Spain. Indicated by this variability, all reported values are relatively close to the density of water (1000 g/L). Because of this, it is assumed reasonable to approximate the density of swine manure as 1000 g/L. This assumption simplifies calculations while remaining consistent with literature values and does not introduce significant error within the scope of this study.

In Table 2.1, the molecular structures in swine manure are shown for an average from a source of the IPCC in 2000 for a picture of what structures are found typically in swine manure [80].

Table 2.1: Average composition of swine manure based on literature values (in % of Total Solids. Carbohydrates are calculated here as the sum of Cellulose, Hemicellulose, and Lignin. The remaining mass ($\approx 6\%$) likely consists of soluble carbohydrates and other minor compounds not explicitly categorized in the source data.) [80]

Component	**Average Value (% TS)**
Lipids	9.0
Proteins	21.9
Carbohydrates (Structural)	**42.5**
Cellulose	16.6
Hemicellulose	19.0
Lignin	6.9
Inorganic residue (Ash)	20.6
Sum of analyzed components	*94.0*

Cellulose, hemicellulose, and lignin are components of plant-derived material present in swine feed that are not fully degraded during enteric digestion in the animal. Cellulose and hemicellulose are biodegradable polysaccharides, whereas lignin is a complex and recalcitrant polymer that is highly resistant to degradation during (anaerobic) digestion[76, 94]

As is shown in Table 2.1, the largest fraction is carbohydrates. Apart from that, 20% of the total solids is inorganic residue. Around 70-75% of the pig manure is based of lipids, proteins, carbohydrates, being the from biologically degradable particles.

2.1.1. Inorganic content

The inorganic content, also described as ash, is an inactive fraction of the manure. This fraction is measured by heating the dry matter to $550°C$. The residue left represents the inorganic ash content [85]. Ash is primarily composed of inorganic material and is particularly rich in phosphorus and potassium, which makes it valuable for recycling as a fertilizer. Because elements like copper and zinc are often added to livestock diets to promote growth and health, these metals are highly concentrated in the manure and will remain also in the inorganic ash fraction [132]

Incineration of manure can lead to drastically reduction of the volume and mass of the waste by 70% to 90%, leaving only the incombustible ash behind. This can be done under very high temperatures (often between 850 °C and 1200 °C) and the ash can be used as mineral fertilizer, but due to the high temperatures, this is not considered as optional treatment for a sustainable method. [132].

2.1.2. Biodegradable fraction

The biodegradable part is the fraction of the manure that can be used for possible products using biodegradation. Biogas, fertilizer are possible products from treatment of this biodegradable fraction[94] The structure of the biodegradable fraction can be explained by the molecular structures as done in Table 2.1, but it can also be described in elemental composition[84].

To characterize the elemental structure of the organic substrate, three standard measurements are routinely conducted, with which the elemental composition, the amount of carbon, hydrogen,oxygen and nitrogen that is inside the manure is estimated. The analytical measurements needed for this are first explained[85]

Chemical Oxygen Demand (COD) The Chemical Oxygen Demand (COD) is measured in $g_{O_2} \cdot kg^{-1}$ and represents the amount of oxygen equivalents required to oxidize all organic carbon to carbon dioxide [84]. The COD is experimentally determined through dichromate oxidation in acidic conditions [84]. The quantity of dichromate reduced during the reaction is measured spectrophotometrically to quantify the oxygen demand[85].

Organic Dry Matter (ODM) Organic Dry Matter (ODM) or Volatile Solids (VS) is measured in $g_{ODM} \cdot kg^{-1}$ or $g_{VS} \cdot kg^{-1}$. This fraction is the difference between the total and inorganic dry matter and contains all mass that is possibly degraded. It is measured as the mass that is converted to gas between 105-550 $°C$[85].

Organic Nitrogen (N_{org}) Organic nitrogen (N_{org}) represents the nitrogen integrated into the organic biomass structure and is measured in $g_N \cdot kg^{-1}$. The mass content is derived from the Kjeldahl Nitrogen (N_{kj}) measurement, which involves the following steps[84]:

1. Destruction: The sample is digested in sulfuric acid (H_2SO_4) to convert organic nitrogen into ammonium (NH_4^+).
2. Stripping: The solution is treated with sodium hydroxide (NaOH) to convert ammonium to ammonia gas (NH_3), which is then stripped from the solution.
3. Titration: The stripped ammonia is captured in acid and titrated to determine the total N_{kj}.

Since N_{kj} includes both organic nitrogen and free ammonium, the organic nitrogen is calculated by subtracting the initial ammonium concentration [85].

The elemental composition of the substrate allows for the estimation of the overall stoichiometry biological treatment processes. By defining the organic substrate as $C_cH_hO_oN_n$ (normalizing on carbon where $c = 1$), using a system of equations 2.1, 2.2 and 2.3[84].

$$h = \frac{308 \cdot COD + 704 \cdot N_{org}}{49 \cdot COD - 64 \cdot N_{org} + 112 \cdot ODM} \tag{2.1}$$

$$o = \frac{224 \cdot ODM - 56 \cdot COD - 304 \cdot N_{org}}{49 \cdot COD - 64 \cdot N_{org} + 112 \cdot ODM} \tag{2.2}$$

$$n = \frac{352 \cdot N_{org}}{49 \cdot COD - 64 \cdot N_{org} + 112 \cdot ODM} \tag{2.3}$$

Where the units for input variables are: COD ($g_{O_2} \cdot kg^{-1}$), ODM ($g_{ODM} \cdot kg^{-1}$), and N_{org} ($g_N \cdot kg^{-1}$).

With these equations, the elemental composition of manure can be calculated.

Ammonium content

As explained, apart from the organic nitrogen, ammonium is dissolved in the swine slurry. The total ammonium content (TAN) describes both NH_4^+ and NH_3 form, whereas the Free ammonia content (FAN) specifically describes the NH_3 fraction. As NH_3 is the toxic un-ionized form of ammonia, the fraction can be causing problems in manure treatment. When pH or temperature increases, the equilibrium of NH_4^+ and NH_3 shifts towards the formation of NH_3. The concentration can be calculated using equation 2.4 [98]

$$FAN = TAN \cdot \left(1 + \frac{10^{-pH}}{10^{-(0.09018+\frac{2729.92}{273.15+T})}}\right)^{-1} \quad (2.4)$$

The FAN and TAN are defined as concentrations in mg/L or kg/m^3, the temperature is in degrees Celsius ($^\circ$C). In literature, for all nitrogen estimations, masses are often defined in mass of nitrogen (kg N), instead of total mass. The relation is also visualized in Appendix B over temperature and pH.

2.1.3. Location, seasonal and feeding dependence

Manure characteristics show inherent variability of the composition, over all characteristics of swine manure. This is caused by variability in location, seasonal conditions, and feeding practices. Location-dependent differences arise from variations in diet composition, climate, animal management, and dilution with water, which influence nutrient concentrations such as nitrogen, phosphorus, and potassium, as well as physicochemical properties including pH and dry matter content. A comparison between manure characteristics from Poland, Denmark, Spain, Canada, and Russia shows clear variability in these properties, highlighting the site-specific nature of manure composition, as reported by Marszałek, Kowalski, and Makara [100] and shown in Table 2.2[100, 129].

Table 2.2: Averaged physicochemical parameters of swine slurry per country.

Parameter	**Netherlands**	**Poland**	**Denmark**	**Spain**	**Canada**	**Russia**
pH	–	7.70	7.09	7.62	7.48	7.05
TS [%]	9.7	3.05	–	1.29	4.05	1.25
COD [g O_2/L]	–	55.3	70.0	24.1	–	14.0
N–NH_4^+ [g/L]	4.8	3.4	4.8	1.8	4.6	0.47
P_{tot} [g/L]	2.1	0.26	1.6	0.76	1.3	0.22
N_{tot} [g/L]	7.4	1.4	5.6	–	–	0.76
Source	[129]	[100]	[100]	[100]	[100]	[100]

In relation to the location, climate and seasonal fluctuations influence the biological and chemical processes during storage and treatment. Higher temperatures enhance microbial activity, leading to increased ammonia volatilization, faster organic carbon degradation, and accelerated conversion of ammonium to nitrate, while lower temperatures slow these processes [162, 76].

Furthermore, feeding practices strongly affect manure composition, as dietary protein, fiber, and mineral content determine nutrient intake and excretion pathways. For example, reducing dietary protein decreases total nitrogen excretion and ammonia emissions, while increasing fiber content influences dry matter and nutrient partitioning [162]

To eliminate these uncertainties as much as possible, constraints for manure characteristics are applied. Manure data used in this study is limited to Spain, ensuring consistency in climate conditions and averaged dietary conditions. Data from measurements conducted throughout the year are used, providing a representative and stable dataset. Variations in feeding are left outside the scope of this study and are assumed to be incorporated in the measured manure data from Spain. As the sulfur content of manure is highly dependent on feed composition, since it originates primarily from dietary inputs, sulfur is excluded from this study [162]. Heavy metals and micro-nutrients are left out of the scope for the same reasons.

For the estimation of emissions, temperature effects are accounted for by using climate zones as defined by the IPCC [78].

2.1.4. Swine manure characteristics obtained from Spain

Measurements of swine manure in Spain are presented in Table 2.3. When available, the number of samples over which the values are averaged is indicated (as Q in m^3/yr). The climate zone of the region where the manure was collected is also specified. The pH of the manure is reported when available.

The TS and VS (Volatile Solids or Organic Dry Matter) are expressed in g/L of manure, assuming a density of 1000 g/L in all recalculations. TKN and NH_4-N are both expressed in g N/L, where NH_4-N denotes the ammonium concentration and TKN the Total Kjeldahl Nitrogen. TP refers to the total phosphate concentration, K to the potassium concentration, and the C/N ratio describes the distribution of carbon and nitrogen in the manure.

As described previously, COD, ODM, and N_{org} are calculated and also presented in Table 2.3. Based on this information, the elemental composition is determined.

The maximum methane yield can then be calculated with the elemental composition, which will be explained in the section 2.5, but already visualized to show fluctuation over data.

Table 2.3: Physicochemical properties of pig manure from various sources. TN = TKN + N_{org}. Q is stated to show the amount of manure the values are the average from. COD and TCOD are used as the same, only if TCOD was not mentioned and no other mentions of SCOD values, the COD value was seen as total. Conversion factor of VS is assumed to be 1.4 g O2/ g VS [80]

Location Year IPCC climate zone Type	Castilla y León, ES 2016 Warm Temperate Dry Swine manure	Castilla y León, ES 2016 Warm Temperate Dry LF Swine	Segovia, ES 2014 Warm Temperate Dry Liquid swine	Vega Baja, ES 2015 Warm Temperate Dry Pig slurries	Average Spain
Q [m^3/yr]	42000	–	–	23800	–
pH [-]	–	–	7.2 ± 0.2	7.54 ± 0.34	7.37 ± 0.27
TS [g/L]	27 ± 17.8	22.5	18.3 ± 6.6	31.0 ± 41.3	24.7 ± 21.9
VS [g/L]	18 ± 14.1	19.5 ± 11.5	13.9 ± 5.5	39.7 ± 27.5	22.8 ± 14.6
COD [g/L]	31 ± 20	26.7 ± 12.8	17.2 ± 8.2	56.4 ± 39.0	32.8 ± 20.0
TKN [gN/L]	3.5 ± 0.9	2.1 ± 0.5	1.424 ± 0.316	3.42 ± 1.75	2.6 ± 0.9
NH_4-N [gN/L]	2.8 ± 0.7	1.6 ± 0.4	1.022 ± 0.233	2.73 ± 1.51	2.0 ± 0.7
TP [g/L]	0.4 ± 0.2	0.7 ± 0.4	0.384 ± 0.145	1.07 ± 1.64	0.6 ± 0.6
K [g/L]	–	–	–	3.46 ± 2.01	3.5 ± 2.0
C/N ratio (calculated)	14.16	19.66	16.78	–	16.9
Additional parameters					
COD [gO_2/kg]	31 ± 20	26.7 ± 12.8	17.2 ± 8.2	56.4 ± 39.0	32.8
ODM [g/kg]	18 ± 14.1	19.5 ± 11.5	13.9 ± 5.5	39.7 ± 27.5	22.8
N_{org} [gN/kg]	0.7 ± 0.2	0.5 ± 0.1	0.402 ± 0.083	0.69 ± 0.24	0.6
Elemental composition (calculated)					
C	1.000	1.000	1.000	1.000	1.00
H	2.877	2.478	2.351	2.491	2.6
O	0.597	0.786	0.854	0.771	0.75
N	0.071	0.051	0.060	0.034	0.054
Maximal methane yield(calculated)					
MW [g/mol]	25.45	27.80	28.88	27.34	27.4
BMP_{th} [$CH_4\ m^3/kgVS$]	0.646	0.514	0.464	0.533	0.541
Source	[47]	[47]	[125]	[107]	

The values from Table 2.3 are as expected in Spain, with the TS is around 2.47 %. By Table 2.1, a VS of 80 % is expected, which is in Table 2.3 somewhat higher, 90%. The elemental composition is $C_1H_{2.6}O_{0.75}N_{0.054}$, being 44% for carbon, 9.4% for hydrogen, 44% for oxygen and 2.8%. The degree of reduction is 4.9 e^-/Cmol. This is more reduced than pure carbohydrates and typical biomass, but less reduced than methane.

2.2. Manure treatment methods

As explained, liquid swine manure consists of multiple nutrients, that can be valorized if treated in the correct way. In this section some treatment methods are discussed, ranging form no to limited intervention to extensive .

2.2.1. No treatment

Conventional management of swine manure typically involves minimal to no actual treatment before the manure is utilized as an organic fertilizer. In these scenarios, raw manure is simply collected, stored for a required period (often up to 6 months) to allow for some stabilization of the organic matter, and then directly applied to agricultural soils. Storage can happen in open lagoons or closed storage tanks. The primary difference is that closed storage prevents direct contact between the liquid manure and the air[4]. Applying a tight, rigid cover to a slurry store can reduce ammonia and odour emissions by 80% to 90% compared to an uncovered lagoon[89]. Open lagoons collect rainwater, which dilutes the nutrient concentration of the slurry. Closed impermeable covers completely prevent rain and snow from entering the tank. This reduces the overall volume of the stored slurry. But, unlike open lagoons, closed tanks trap explosive (methane) and toxic (hydrogen sulfide) gases. Therefore, closed systems must be carefully designed with gas vents to safely release or capture these gases to prevent dangerous accumulation[132, 4, 76].

2.2.2. Solid liquid separations

Solid/liquid separation is frequently implemented as a primary pretreatment step in manure management to improve handling and facilitate targeted nutrient recovery [58, 132]. Using mechanical devices such as decanter centrifuges, screw presses, and sieves, raw manure is partitioned into two distinct streams [58, 2]. The resulting solid fraction retains the bulk of the phosphorus and organic matter, yielding a stackable product that is easier and more cost-effective to transport to nutrient-deficient regions [58, 132]. Conversely, the liquid fraction retains the majority of the nitrogen and potassium, which can be used as soil conditioner for crop growth, but the largely diluted nutrients could be transported better if a concentration step would be added [58, 132, 100]

2.2.3. Anaerobic digestion (AD)

Anaerobic digestion is widely recognized as a core biotechnology for organic waste stabilization and bioenergy generation [72, 35]. In this process, a consortium of microorganisms decomposes organic matter in an oxygen-free environment to produce biogas, a mixture primarily composed of methane and carbon dioxide [72, 132]. The capture and utilization of this biogas for heat or electricity significantly offsets fossil fuel consumption and mitigates the methane emissions that would otherwise occur during conventional open-lagoon storage [132, 101]. Furthermore, the residual digestate is hygienized, stabilized, and contains mineralized nutrients that serve as a high-quality organic fertilizer [72, 101]. However, AD of nitrogen-rich substrates (like swine or poultry manure) must be carefully managed, as the accumulation of free ammonia can inhibit methanogenic bacteria and reduce process efficiency [139].

2.2.4. Composting

Composting is a controlled aerobic decomposition process, utilized to treat solid raw manure, the solid fraction obtained from SLS or digestate out of an AD [132]. Conducted in windrows, static piles, or enclosed vessels, composting relies on microbial activity to break down complex carbon compounds [112, 132]. The exothermic nature of this biological degradation drives the pile temperatures into the thermophilic range (50–70 °C), effectively destroying pathogens and weed seeds [132]. Additionally, the heat evaporates significant amounts of moisture, substantially reducing the total volume and mass of the waste [132]. The final product is a friable, humified, and odor-reduced compost that improves soil structure, though care must be taken to balance the carbon-to-nitrogen (C/N) ratio and aeration to prevent volatile nitrogen losses and greenhouse gas emissions [132, 104].

2.2.5. Ammonia stripping

Ammonia stripping is a physicochemical process used as a pre- or post-treatment to remove and recover nitrogen from liquid streams such as manure or anaerobic digestate [132, 62]. The process relies on shifting the equilibrium between ammonium (NH_4^+) and ammonia (NH_3) towards the gaseous form by increasing the pH (typically to 9–10 using alkali addition) and temperature [132]. In a stripping unit, the liquid is introduced at the top of a vertical packed column, while a stripping gas, usually air or steam, is injected from the bottom. The gas flows counter-currently to the liquid, enhancing mass transfer and promoting the volatilization of ammonia from the liquid phase into the gas phase. The packing material increases the contact surface area, improving the efficiency of ammonia removal. The treated liquid, with reduced ammonia concentration, is collected at the bottom of the column. The ammonia-rich gas stream is subsequently directed to a scrubber or absorber, where it reacts with an acidic solution (e.g., sulfuric or nitric acid). This results in the formation of stable ammonium salts such as ammonium sulfate or ammonium nitrate, which can be used as concentrated liquid fertilizers [132, 154]. In the case of steam stripping, ammonia may alternatively be recovered via condensation as an aqueous solution. Although ammonia stripping is highly effective in recovering nitrogen and reducing ammonia toxicity for downstream biological processes, it is associated with relatively high energy and chemical consumption [132].

2.2.6. Membrane filtration

Membrane technologies offer advanced separation and nutrient recovery capabilities for liquid manure fractions. Pressure-driven systems, such as microfiltration, ultrafiltration, and reverse osmosis, can physically separate suspended solids, large macromolecules, and dissolved ions, ultimately yielding a concentrated nutrient retentate and purified water suitable for discharge or reuse [58, 2]. More recently, gas-permeable membrane (GPM) technology has emerged as a promising alternative for nitrogen recovery. In GPM systems, a microporous hydrophobic membrane is submerged in the wastewater; volatile ammonia gas diffuses through the membrane pores and is directly captured by an acidic trapping solution circulating on the inside [52]. Compared to traditional stripping, GPM systems operate at lower pressures and can avoid the heavy use of alkali chemicals, though membrane fouling remains a persistent operational challenge across all membrane-based applications [58, 52].

2.2.7. Treatment method choice

For the purpose of nutrient recovery of liquid swine manure in a usable end-product, the anaerobic digestion of swine manure is identified as the most promising treatment, given the capture of a usable end product, hygienization and stabilization of the solid outflow and with possible use of the nutrients as fertilizer. The technology is widely applied on manure management and studied. Gas of methane is in this way used, instead of emitted to the environment. To investigate the treatment method further, the anaerobic digestion is further explained.

2.2.8. The process of anaerobic digestion

The anaerobic digestion process relies on four distinct functional groups of microbes that act successively through four main stages, that is visualized in Figure 2.1 [72, 111].

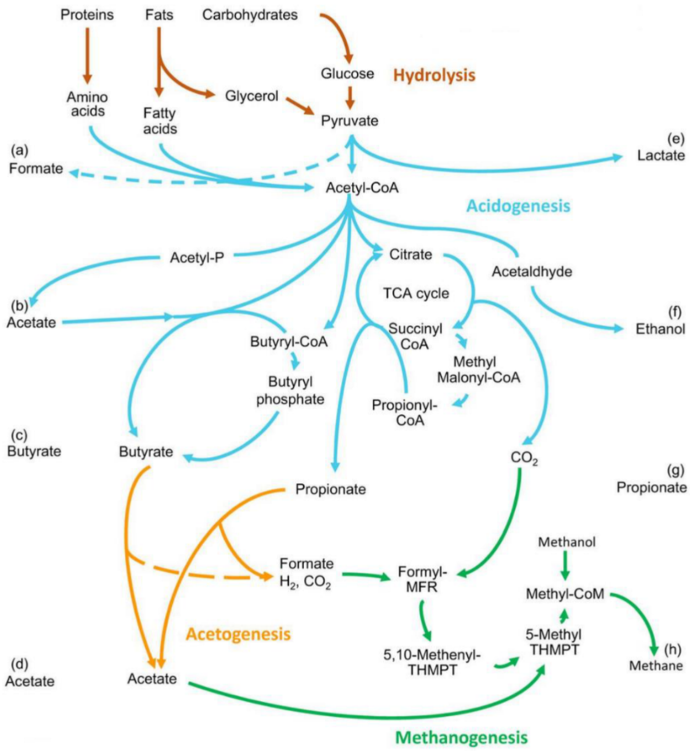


Figure 2.1: Anaerobic digestion metabolic pathways and key intermediates [111]

The steps shown in Figure 2.1 are:

- Hydrolysis: In this initial stage, hydrolytic bacteria secrete extracellular enzymes to break down complex organic materials, such as carbohydrates, proteins, and fats—into simpler monomers and oligomers like sugars, amino acids, and lipids[84, 85]
- Acidogenesis: Next, acidogenic microorganisms ferment these simple monomers into short-chain Volatile Fatty Acids (VFAs), acetic acid, and other organic molecules. During this phase, significant amounts of carbon dioxide (CO2) and hydrogen (H2) are also produced [111] Genera such as *Clostridium*, *Proteiniphilium*, and *Saccharofermentans* play key roles in the hydrolysis and acidogenesis[84, 57]
- Acetogenesis: Acetogenic bacteria, such as *Syntrophomonas* and *Petrimonas*, then step in to further break down the organic molecules from the previous stage, converting them into acetate, CO2, and H2. These specific compounds serve as the direct precursors for the final methane-producing step[139, 84].
- Methanogenesis: Finally, methanogenic archaea finalize the transformation. This stage involves two distinct groups: slow-growing acetoclastic methanogens, like *Methanothrix*, that consume acetate to create methane, and a second group of methanogens, hydrogenotrophic methanogens such as *Methanospirillum*, that consume hydrogen and carbon dioxide to produce methane[72]

Ultimately, this four-step process converts the organic substrates into biogas, which is typically composed of approximately 70% methane (CH4) and 30% carbon dioxide (CO2), leaving behind a stabilized residual digestate[72, 111, 84].

In the acidogenesis, Volatile fatty acids (VFAs) are produced which are short-chain carboxylic acids, including acetic, propionic, butyric, isobutyric, valeric, isovaleric, and caproic acids [118, 57, 132]. A large propotion of the global VFA market is currently supplied by petrochemical synthesis, leading to a major focus on sustainably producing VFAs by fermenting renewable organic waste streams[145, 8].

VFA's can be used as precursors for bioplastics, when they serve as the primary carbon source for the biological synthesis of polyhydroxyalkanoates (PHAs), a family of fully biodegradable and biocompatible bioplastics [115, 57]. VFAs extracted from fermented waste are also applied in wastewater treatment plants, used as a highly efficient external carbon source to drive denitrification (nitrogen removal) and enhanced biological phosphorus removal [157, 8]. Additionally, it can be utilized to generate various renewable energy carriers, including biogas (methane), biohydrogen, and bioelectricity via microbial fuel cells [6, 8].

Individual VFAs are lucrative commercial commodities, which can be obtained after separation and purification. Acetic acid is used heavily in polymers, paints, adhesives, and food preservatives . Propionic acid is essential for manufacturing vitamin E, herbicides, and animal feed supplements. Butyric acid is used as an animal feed antibiotic alternative, a food flavoring agent, and is being researched for anticancer properties. Caproic acid serves as a raw material for lubricants, antimicrobials, and pharmaceuticals[115, 8]

As purification of VFA' mixtures require additional energy, biofuel applications do not differ much from biogas production through AD and waste water applications not produce a valuable end-product apart from clean water, the production of bio-plastics is considered a potentially useful end-product for swine manure treatment. This is also funded but the fact that VFA mixtures are considered the preferred substrates over individual VFA's for mixed microbial culture (MMC) PHA production[151].

Because of this, two treatment methods are further analysed in for treatment of swine manure in this study. First, the production of biogas with anaerobic digestion, with additional treatment of digestate and secondly the production of bio-plastic, using shortened anaerobic digestion for VFA production, followed by PHA production and required down stream processing. But first, the size of the plant is decided, to specify the treatment methods for this.

2.3. Plant size

The selection of the plant size is based on regulatory , environmental or practical considerations and economic, related to pig farming and manure processing in Spain.

First, regulations play an important role in determining the minimum relevant farm size. According to Spanish legislation on integrated pollution prevention, farms with more than 2000 fattening pigs are subject to stricter environmental controls due to their significant emissions to air, soil, and water [131]. This threshold ensures that manure treatment technologies are most relevant for medium to large-scale farms. This was also explained in the Introduction, on low-productivity farms with fewer than 2000 pigs, manure is does not have to be actively managed by the farmer, as it can be applied directly on the land [76]. Thus, farms larger than 2000 farms were only considered for this study.

But, as manure and its resulting digestate consist mostly of water, long-distance transport of large quantities is highly uneconomical [35, 124] Additionally, hauling the liquid manure and compost to and from centralized plants generates significant environmental burdens, including greenhouse gas emissions and NO_x emissions from heavy trucks [32, 136]. Decentralized on-farm plants minimize the distance between the waste generation and the treatment facility, effectively mitigating these transport-related impacts.

Apart form importing the manure, centralized plants process waste from dozens of farms, concentrating enormous volumes of nutrient-rich digestate in a single location. If the agricultural land surrounding the centralized plant is too small to safely absorb this fertilizer, it leads to a local nutrient oversupply, forcing the facility to transport the surplus digestate out of the region at a high logistical cost. Keeping treatment at individual farm level avoids this extreme concentration of nutrients. Decentralized treatment also allow for better control over the composition and flow of the manure, which is particularly important for sensitive biological processes such as shortened anaerobic digestion [72, 150, 124]

From an economic perspective, mechanical treatment is more preferred for very large farm, whereas for smaller farms, biological upgrading is the recommended and most cost-effective option, according to Sánchez-Martín et al. [131]. This makes biological treatment like anaerobic digestion suitable for medium-sized farms[131].

Regarding geographical context, pig production in Spain is highly concentrated. The main regions are Catalonia, Aragon, and Castilla y León, which together account for approximately 65% of national production [137]. Catalonia is characterized by farms with approximately 32,000 pigs per farm, making membrane separation treatment more optimal[137]. The region Castilla y León has a large number of farms with significant production capacity, with farm size ranging between 1000 and 20,000 pigs per farm [137, 131, 128].

Based on these considerations, a farm with approximately 3400 pigs located in Castilla y León was selected for this study, somewhat larger than the minimal of 2000 pigs, but with considerable amounts of manure. By Martínez-Hernando et al. [101] a similar capacity for manure treatment is chosen (3450 pigs for one farm).

The manure production is estimated using a standard value of 2.5 m^3 per pig per year, resulting in a total annual manure production of approximately 85,000 m^3/year [131]. This scale is large enough to justify the implementation of anaerobic digestion, while still being representative of a single-farm system with controlled and consistent feedstock input.

2.4. Environmental effects of manure treatment

The environmental effects of manure treatment methods can be quantified using Life Cycle Assessment (LCA). LCA is a standardized environmental management technique (regulated by ISO 14040/44 standards) used to evaluate the potential environmental impacts and aspects of a product, process, or service throughout its entire lifespan. LCA consists of 4 phases, in which first the scope of the study is determined and the boundary limits, after which all materials, energy, emission going in and out of the system are quantified. Using characterization factors, the mass flows can be translated to equivalents per category. This is at last evaluated with literature to identify the meaning [45]. A visual overview of an example LCA method is shown in Appendix C

2.4.1. Impact categories

In LCA studies, Global warming potential(GWP), terrestrial acidification (TA), and freshwater eutrophication (FE) are the most important effects in literature for swine manure treatment [45]. Regarding the ammonia gas emissions possible in swine manure, fine particulate matter formation (FPMF) is also discussed in this study, next to the effect of phosphorus emission to water environments, leading to marine eutrophication (ME). These effects are in LCA methods called impact categories.

Global warming potential

This metric evaluates a system's contribution to global warming through the emission of greenhouse gases. The primary greenhouse gases addressed are CO_2, CH_4, and N_2O. In contexts such as manure treatment, these gaseous emissions must be calculated and aggregated into a common unit of carbon dioxide equivalents (kgCO_2-eq). As storage and treatment of manure influence the emission of greenhouse gasses, the global warming potential is considered in this report.

Terrestrial acidification

This category quantifies the potential for atmospheric depositions of inorganic compounds, such as NO_x, NH_3, and SO_2, to induce a decrease in soil pH. This change in acidity, commonly referred to as acid deposition or acid rain, poses a risk to the viability of terrestrial organisms and soil microbial communities. The characteristic unit for this impact is often expressed in sulfur dioxide equivalents (kgSO_2-eq). As the agricultural sector is a large contributor of the emissions of NH_3, this impact category is selected for this study.

Eutrophication

This effect is induced by the discharge of excess nutrients, specifically nitrogen and phosphorus, into freshwater and marine water bodies. The subsequent elevation in nutrient concentrations triggers the uncontrolled proliferation of micro-algae. The eventual decomposition of this massive biomass consumes dissolved oxygen, resulting decline in species diversity and mortality of aquatic fauna. This impact is typically measured in phosphate equivalents in freshwater (kgP-eq) and nitrogen equivalents in marine water(kgN-eq.). Regarding the large effect of fertilizers on eutrophication, this effect is accounted for in the evaluation of processes in this report.

Fine particulate matter formation

This effect is caused by the emission of fine particulate matter, consisting of very small airborne particles (PM_{10} and $PM_{2.5}$) released by activities such as traffic, industry, shipping, and agriculture. Due to their small size, these particles can penetrate deep into the respiratory system, where they induce inflammation and contribute to respiratory and cardiovascular diseases, ultimately leading to increased morbidity and reduced life expectancy. The impact is evaluated through population exposure to PM concentrations (µg/m^3). Because even low concentrations are associated with adverse effects, fine particulate matter is considered a major contributor to the health burden from air pollution in manure treatment [63]. By Nordahl et al. [112] the effect of ammonia (NH_3) emissions to air pollution is emphasized. It is shown that ammonia is a major driver of air quality-related health damages, especially in agricultural and composting systems, where it can dominate total social costs. This is why this impact category is included in this study.

These environmental effects are taken into account in designing the swine manure treatment method.

2.5. Biogas production

In this section the complete process of swine manure to biogas is discussed, with application of methods to prevent negative environmental effects and produce useful products instead. As biogas consist mainly of methane and carbon dioxide, of which methane can be used for energy generation, the potential methane production from manure is first discussed.

Potential methane production in anaerobic digestion

In anaerobic digestions of manure, the theoretical methane yield and actual produced methane in biogas differ.

The theoretical Biochemical Methane Potential (BMP) is the total maximum of methane that could yield from the substrate. In practice, this never happens, as never a 100 % efficiency is reached. So this value estimates the upper bound. The theoretical BMP can be estimated using the Buswell formula. This formula balances the chemical composition of the substrate ($C_aH_bO_c$) with only water to predict a reaction with only products of CO_2 and CH_4, which is shown in equation 2.5 [133]

$$C_aH_bO_c + \left(a - \frac{b}{4} - \frac{c}{2}\right) H_2O \rightarrow \left(\frac{a}{2} + \frac{b}{8} - \frac{c}{4}\right) CH_4 + \left(\frac{a}{2} - \frac{b}{8} + \frac{c}{4}\right) CO_2 \tag{2.5}$$

With equation 2.5, the theoretical methane potential (BMP_{th}) in normal liters of methane per volatile solids ($NL\ CH_4 \cdot VS^{-1}$) can be calculated. This is shown in equation 2.6 [133]

$$BMP_{th} = \frac{V_m \times moles_{CH4}/mol_{substrate}}{MW_{substrate}} = \frac{V_m \times \left(\frac{a}{2} + \frac{b}{8} - \frac{c}{4} - \frac{3d}{8}\right)}{12.017a + 1.0079b + 15.999c + 14.0067d} \tag{2.6}$$

By applying the elemental composition of the Spanish swine manure of Table 2.3 $C_1H_{2.6}O_{0.75}N_{0.054}$, the BMP_{th} for these averages can be calculated, being 541 L CH_4 kg^{-1}.

The theoretical maximum methane producing capacity (BMP) for swine manure is also described by the IPCC (Table 10.16), as approximately 450 L CH_4 kg^{-1} VS for Western Europe.

Given that estimation in Table 2.3 assume all volatile solids to be biodegradable, the theoretical maximal yield can be adjusted with leaving out the lignin in the manure, using the molecular structures of swine manure in Table 2.1.

The fractions in the volatile solids of the main biodegradable fractions are shown in in Table 2.1 and elemental structures are considered $\mathrm{C_6H_{10}O_5}$ for cellulose, $\mathrm{C_5H_{10}O_5}$ for hemicellulose, proteins with $\mathrm{C_5H_7O_2N}$, and lipids $\mathrm{C_{57}H_{104}O_6}$ [98]. Lignin is excluded from the stoichiometric calculations, as it is a highly recalcitrant polymer that is degraded very slowly or not at all under anaerobic digestion conditions [143].

By averaging the elements in the molecular structures with the fractions of occurrence in swine manure, a general approximation of the biodegradable fraction can be made. For this, relative mass fractions of the biodegradable components are normalized with respect to the total volatile solids content, including lignin. Thus lignin is included to express the methane yield per unit mass of total volatile solids, but not considered as a fraction that can be converted to CH_4 and CO_2. After this normalization and expressed per 1 carbon, an empirical molecular formula is derived as $C_1H_{1.77}O_{0.38}N_{0.027}$

Using the corresponding molar mass and Buswell stoichiometry, the maximum theoretical methane potential of the biodegradable fraction is calculated. The resulting theoretical yield is 495 L CH_4 kg^{-1}.

This value represents a better estimation of the upper limit of methane production based on complete conversion of the biodegradable organic matter under ideal anaerobic digestion conditions and is used in the stoichiometric models in this study.

Actual methane yield

The actual methane yield is influenced by multiple substrate- and process-related factors and is generally lower than the theoretical methane potential.

Substrate composition strongly affects the actual methane yield. High ammonium concentrations may cause ammonia inhibition, reducing methane production efficiency. Process conditions further determine the extent to which BMP is achieved. Temperature, pH, inoculum-to-substrate ratio, hydraulic retention time, and mixing intensity affect microbial activity and degradation efficiency.

To approximate the anaerobic digestion of the manure better for the methane yield, a factor to describe the efficiency of the degradation of the substrate, the biodegradability, is used.

Biodegradability determines the extent to which the organic matter in a substrate can be decomposed by anaerobic microorganisms[133]. The biodegradability percentage is normally experimentally determined by comparing the actual methane produced during a Biochemical Methane Potential (BMP) assay to the theoretical maximum potential derived from stoichiometric equations (such as the Buswell formula). It is calculated according to the following equation 2.7 [133]

$$Biodegradability\ (\%) = \frac{Observed\ methane\ yield\ (CH_4\ L/kg\ VS)}{Theoretical\ methane\ yield\ (CH_4\ L/kg\ VS)} \times 100 \tag{2.7}$$

This parameter is essential for assessing the efficiency of the anaerobic digestion process, as it reveals how much of the theoretical energy potential can essentially be recovered in a real-world system [133].

Reported biodegradability values for swine manure vary considerably, ranging from approximately 70% under ideal conditions [80] to 40–52% in full-scale mono-digestion systems in the Netherlands [82].

2.5.1. Reactor parameters

The anaerobic digestion of manure at a large scale is typically performed using continuous flow reactors. The choice of reactor configuration significantly impacts the economic viability and efficiency of the industrial process.

The efficiency of anaerobic digestion is heavily influenced by operational parameters, particularly temperature and pH. The optimum temperature for mesophilic digestion typically lies between 35-40 °C. Deviations from this range, such as sub-mesophilic or thermophilic conditions, often result in reduced methane yields [31]. While thermophilic conditions can increase reaction rates, they also heighten the risk of free ammonia (NH_3) inhibition. At higher temperatures, the chemical equilibrium shifts, increasing the concentration of toxic free ammonia gas, as is shown in equation 2.4, which induces microbial stress in methanogens.

Elsgaard, Olsen, and Petersen [34] investigated the temperature response of methane production from liquid pig manure and reported a strong dependency within the range of 5–37 °C. The study shows that methane production increases by nearly a factor of three for every 10 °C rise in temperature.

The total solids (TS) concentration of the feedstock is an important operational parameter in anaerobic digestion, as it directly influences process stability, potential methane yield and reactor operation. Based on total solids (TS) concentration, anaerobic digestion processes are commonly classified as wet digestion ($<$10% TS), semi-dry digestion (10–20% TS), and dry digestion ($>$20% TS) [31]. As the swine manure considered in this study has a TS content of approximately 2%, wet digestion can be applied directly without the need for pre-heating. Dry digestion systems offer advantages such as smaller reactor volumes, since a higher fraction of volatile solids (VS) is present, which can reduce energy demand for heating and slurry pumping [31]. However, substrates with high solid contents are more difficult to handle, pump, and mix, leading to potential mass transfer limitations and local accumulation of inhibitory compounds [31]. As a result, dry digestion processes are more prone to operational instability, including ammonia toxicity and volatile fatty acid accumulation. Furthermore, dry anaerobic digestion systems are generally more sensitive to temperature variations, which can negatively impact process performance [31].

Wet digestion is applied in a centralized anaerobic digestion plant in Castilla y León treating exclusively pig manure from 46 surrounding farms operates with an influent TS concentration of 27.0 ± 17.8 g/L (approximately 2.7%) [47]. Also, a proposed full-scale centralized system in Galicia evaluating pig manure mono-digestion measured the raw manure TS concentration at approximately 3.0% and laboratory-scale evaluations of pig manure from an experimental farm in Segovia also utilized a raw manure TS concentration of 2.7%[128, 51]. Lymperatou et al. [98] did not include additional water, operating at a TS content of 3.81%. Given that the average TS content of Spanish pig manure is 2.47%, wet digestion is applied with no extra needed dilution.

2.5.2. Co-digestion

Co-digestion refers to the practice of mixing swine manure with one or more complementary organic substrates (co-substrates) in an anaerobic digester, rather than treating manure alone. This approach is widely applied to overcome the limitations of mono-digestion, as swine manure typically has a high water content, low organic matter content, and a low carbon-to-nitrogen (C/N) ratio. As a result, mono-digestion often leads to relatively low biogas yields and increased susceptibility to ammonia inhibition due to the high nitrogen content[51, 128, 50]

The addition of co-substrates can improve nutrient balance, enhance biogas and energy yields, and result in a higher-quality digestate. Furthermore, co-digestion can improve the overall economic feasibility of anaerobic digestion by increasing process efficiency and energy recovery.

However, co-digestion also introduces logistical and operational challenges. The addition of external substrates increases the total volume of digestate, requiring more land for application and potentially leading to higher transportation and handling costs. Moreover, the variability in co-substrate availability and composition introduces additional uncertainties and fluctuations in process performance.

As the objective of this study is to evaluate treatment options specifically for swine manure, and to limit uncertainties, only mono-digestion is considered[84, 132].

2.5.3. Study cases of real life digesters

To estimate the methane yield of an actual anaerobic digestion, real-time industrial plants are studied in Spain.

First, the LIFE Smart Agromobility project is a farm-scale demonstration plant located in the province of Soria, Spain, designed to produce vehicular biomethane from pig slurry through anaerobic digestion combined with an innovative biological upgrading system based on microalgae. The anaerobic digestion unit consists of a low-cost flexible horizontal bag-type digester, with a total volume of approximately 150 m^3. The digester operates under mesophilic conditions. The feedstock consists exclusively of pig slurry originating from a farm housing approximately 3,450 pigs. Due to technical and operational constraints, only about 29% of the total manure produced on-site is directed to the digester, corresponding to an average inflow of approximately 4,680 kg of manure per day.

A average methane content of 70.5 vol.%, with observed seasonal variation ranging from 62.8 vol.% in winter to 75.1 vol.% in late summer.

The digestate produced is predominantly used as organic fertilizer, while a small fraction (approximately 3.2%) is diverted to support microalgae growth in the upgrading system. The micro-algae pond is used to subtract the CO_2 form the biogas for upgrading to biomethane of 90 vol.%,.

All operational data reported for the LIFE Smart Agromobility plant are based on daily averages derived from continuous operation over a full year. [101, 150]

Girón-Rojas et al. [50] presents a mono-digestion system as farm-scale anaerobic digestion plant located in Extremadura, Spain, designed to treat pig slurry exclusively throughout the year. The system was developed through process upscaling based on laboratory data and aims to evaluate the technical, environmental, and economic feasibility of decentralized energy production on livestock farms. The AD unit is dimensioned specifically for the treatment of pig slurry generated on-site, without the inclusion of co-substrates. Farm 2 consists of approximately 167 pig units, producing around 18 t of slurry per day. This corresponds to an annual slurry input of approximately 6,654 t. The digester volume is estimated at 881 m^3.

The digestion process operates under mesophilic conditions at approximately 37 °C, with continuous mixing to maintain homogeneous conditions within the reactor. The organic loading rate is approximately 1.4 kg VS/m^3. A yield of approximately 237 L/kg VS and a methane content of around 57% is estimated.

The produced biogas is utilized in a combined heat and power (CHP) unit with an electrical efficiency of approximately 30% and a thermal efficiency of 55%. A significant portion of this energy is used on-site to meet the farm's operational needs, the energy surplus is sold.

Table 2.4: Comparison of anaerobic digestion for biogas and digestate production. *Climate zones determination with [78]. [a]: yield calculations done in assumption of 2.3 % VS of total mass.

Specification	**LIFE Smart Pilot** [101]	**Farm-scale Mono-AD** [50]
Location of plant	Soria, Spain	Extremadura, Spain
Climate zone*	Warm temperate, dry	Warm temperate, dry
AD reactor volume [m^3]	150	881
Reactor type	Flexible bag-type digester	Regular digester
Reactor temperature [$^\circ$C]	Mesophilic	Mesophilic (37 °C)
Hydraulic retention time (HRT) [days]	30–32	6–9
Methane yield [L CH_4/kg VS]	112 [a] (70.52%)	135 (57%)
Annual manure treatment [$\times 10^6$ kg/yr]	5.39	6.65
Electricity demand [kWh/day]	728.5	294.6
Operating time [days/year]	365	365
Sources	[101, 116, 131]	[50]

Based on the available studies, an estimation of a realistic methane yield for swine manure under industrial conditions in Spain can be made. Reported yields of 112 and 135 L CH_4 kg^{-1} VS allow for the estimation of the effective biodegradability of the substrate. The observed methane yield in a low-cost bag digester was approximately five times lower than the expected value, primarily due to temperature variability, resulting in an estimated biodegradability of around 22% when compared to the theoretical maximum methane yield of 495 L CH_4 kg^{-1} VS of this study [150].

However, Martínez-Hernando et al. [101] considers only a fraction of the total manure directed to the digester, and the reported values rely on assumptions for recalculation to kg VS units (2.3% VS), the yield reported by Girón-Rojas et al. [50] is considered more representative for this study. This study reports a methane yield of 135 L CH_4 kg^{-1} VS, corresponding to a biodegradability of approximately 27%.

2.5.4. Down stream processing

The upgrading to bio-methane done by Martínez-Hernando et al. [101] seems like a good initiative, but to keep the system simple, this is not considered in the design for this study. The micro-algae growth on the liquid fraction of the digested manure does seem like a good option, to use the nutrients in the water and clean it simultaneously. The direct combustion of the biogas of Girón-Rojas et al. [50] seems like a optimal way to generate both electricity and heat for the total plant, thus this is applied.

In conclusion, an AD reactor is followed by an CHP for biogas combustion. For the digestate, in literature, solid liquid separation is largely applied to partition the P and N fractions. To process the solid fraction of the digestate properly, composting is chosen as the most simple application for nutrient recovery and sensitization. As the liquid fraction of the digestate still contains a lot of nitrogen and potassium, micro-algae cultivation is proposed to use the nutrients efficiently and reduce nutrient exposure to the environment.

Combined Heat Production (CHP)

In a combined heat and power (CHP) system, biogas is first transformed into mechanical energy, which drives a generator to produce electricity. At the same time, the produced heat is recovered and can be used for plant operations.

The energy content of biogas depends mainly on its methane concentration. Two measures are commonly used to characterize fuel energy content: the higher heating value (HHV) and the lower heating value (LHV). The HHV represents the total heat released during complete combustion, including the latent heat recovered from water vapor condensation, whereas the LHV excludes this latent heat and is therefore more relevant for practical energy applications. Since methane is the primary combustible component, the LHV of biogas is approximately proportional to its methane content and can be estimated with equation 2.8, where x_{CH_4} is the volumetric methane fraction.

$$\text{LHV}_{\text{biogas}} \approx x_{\mathrm{CH}_4} \times 9.94\,\text{kWh/m}^3, \tag{2.8}$$

Normal biogas has a composition of approximately 50-70 % methane, 30-50% CO_2, leading to a LHV between 4.7 and 7.0 kWh/m^3. Trace elements can be water vapor and hydrogen sulfide, of which the latter is very toxic and should always be eliminated from the gas before use, but is left outside of the scope given the constraints of this study. [20, 74].

During the storage and combustion of biogas leaking usually occurs. The range of leaking applied by the IPPC is stated between 1-10 %[76].

Digestate storage

The remaining liquid flowing out of the anaerobic digestion is called the digestate. The digestate contains all dissolved components, like ammonium, inorganic ash residues, the manure that is not digested, the biomass cells of the micro organisms growth in the digester and water.

In most treatment processes, the digestate is first stored for a period of time. During storage, as part of the digestate is still active, methane can still be produced. Next to this, ammonium gas is also emitted [32, 89].

Solid-liquid separation

To use of the nutrients properly, the digestate needs to be processed further. For this, solid-liquid separation is largely applied as goal to partition the phosphor and the nitrogen [32].

Multiple solid-liquid separation units are applied in industry. For example, a centrifuge, natural settling process and a decanter are applied in manure treatment [58, 32]. Pig slurry typically contains a high fraction of fine particles and a relatively low to moderate dry matter (DM) content. Approximately 65-70% of the dry matter in pig slurry is present in particles smaller than 25 μm, which limits separation efficiency by gravity-based methods [58]. Furthermore, most ammonium and potassium are dissolved in the liquid phase, while phosphorus is mainly associated with small particles [58]. To separate these small particles from the liquid, a decanter is the most efficient technique for removing dry matter and phosphorus in swine manure [58].

A decanter centrifuge is a technology used for the separation of solids and liquids in digestate that partitions the material into a solid fiber fraction and a liquid fraction. It is frequently utilized in manure digestion or industrial waste treatment plants and can process large volumes of biomass, with documented capacities reaching 13.72 m^3 per hour [2].

General separation of the fractions are described and compared to real-life decanter separation efficiencies of a biogas production plant, in Table 2.5.

Table 2.5: Partitioning factors for separation of digestate by decanter centrifuges. Comparison between general sepration values of decanters, the measurements of a industrial application and specific separation values of swine and cow manure. SF: solid fraction. LF: liquid fraction.

Specification	General separtion [2]		Industrial application [15]		Manure separation [58]		Average	
	SF (%)	LF (%)	SF (%)	LF (%)	SF (%)	LF (%)	SF (%)	LF (%)
Total mass	8	92	12	88	14	86	11	89
TS	86	14	47	53	61	39	65	35
VS	65	35	50	48	-	-	58	42
Water	6	94	9.1	91	8	93	8	92
TN	25	75	20	81	28	72	24	76
TAN	8	92	16	83	16	84	13	86
TP	78	22	63	32	71	29	71	28
K	7	93	13	91	-	-	10	92
C	70	30	-	-	-	-	70	30

According to Al Seadi et al. [2], ash separation in digestate is 53 % of the mass to the liquid fraction, 47% in the solid.

Composting

Composting of the solid fraction of digestate is widely applied [132].

Emissions and nutrient losses during composting occur due to the coexistence of aerobic and anaerobic zones. Limited aeration promotes methane (CH_4) and nitrous oxide (N_2O) formation, while excessive aeration enhances ammonia (NH_3) volatilization. Nitrogen losses through leaching and runoff may also occur, particularly in less controlled systems [76]. Overall, carbon is mainly emitted as CO_2, while nitrogen losses can be substantial.

Emission profiles are influenced by both feedstock characteristics and process conditions. AD as a pre-treatment reduces volatile solids, limiting degradable organic matter and generally lowering N_2O emissions compared to raw manure. In contrast, CH_4 emissions are primarily governed by process conditions such as aeration and moisture content, with the effect of AD remaining uncertain. NH_3 emissions tend to be lower for digestate due to prior stabilization, although results vary [112, 132].

Overall, while AD pre-treatment can reduce emissions, particularly N_2O, the final emission profile is strongly dependent on composting conditions such as aeration, moisture content, and C/N ratio [112].

Micro-algae cultivation

After anaerobic digestion and solid liquid separation, the liquid fraction of digestate contains high concentrations of ammonium, phosphate, and soluble organic matter [2, 32]. This nutrient-rich stream can be reused as a growth medium for micro-algae in cultivation ponds or photo-bioreactors, providing an effective method for nutrient recovery and wastewater treatment[156, 93, 61].

Microalgae utilize sunlight to absorb carbon dioxide and inorganic nutrients, while producing oxygen that supports aerobic bacteria in degrading organic matter [25, 156]. CO_2 is often supplied through gas injection [25, 123]. Ammonia concentration is a critical operational parameter. Excessive ammonium levels may inhibit algal growth and reduce biomass productivity [156]. Therefore, dilution, filtration, or partial treatment of the digestate is often applied prior to cultivation. At last, temperature also strongly affects performance, with higher productivity in warm conditions [116].

Unicellular green algae from the Chlorellaceae family, particularly *Chlorella vulgaris*, are commonly used due to their robustness and adaptability to outdoor conditions. These species often dominate naturally in open raceway systems, maintaining relative abundances above 50% across seasons [116]. Apart form this, Chlorella species are mixotrophic, allowing growth on both inorganic carbon (CO_2) and organic carbon from wastewater, which enhances treatment efficiency [25, 156].Chen et al. [19] investigated the nutrient removal efficiency of *Chlorella* species cultivated on the liquid fraction of digested swine manure under conditions comparable to this study. Reported removal efficiencies for *Chlorella vulgaris* are 84% for nitrogen and 91% for phosphorus. Additionally, ammonium removal efficiencies of 91% is reported under the same conditions. As no mention of nutrient uptake efficiency is done by this source, the 86% SCOD removal efficiency of Li et al. [93] is used, which was observed in Chlorella grown on the liquid fraction of chicken manure. The harvesting of *Chlorella* requires sedimentation or centrifugation [156].

With an ultimate analysis, the elemental composition of *Chlorella vulgaris* is measured to be $C_1H_{1.9}O_{0.49}N_{0.13}$ [130]. The compositional properties of *Chlorella* 1067 are derived from Li et al. [93] and shown in Table 2.6.

Table 2.6: Characteristics of *Chlorella* 1067 obtained form Li et al. [93]

Parameters	*Chlorella* 1067
TS, %	97.23 ± 0.14
VS, %TS	82.91 ± 0.07
TKN, mg·g^{-1} TS	75.74 ± 5.43
TN, mg·g^{-1} TS	85.11 ± 3.52
TP, mg·g^{-1} TS	18.75 ± 2.78
N-[NH_4^+, NH_3], g·L^{-1}	2.67 ± 0.03
Protein, %TS	47.34 ± 0.55
Lipids, %TS	6.76 ± 0.44
Carbohydrate*, %TS	29.02 ± 0.52
C/N	5.97 ± 0.15
N/P	4.53 ± 0.78

*Values calculated by difference.

The algae product of the cultivation pond has been investigated as a strategy to enhance methane production and process efficiency. The inclusion of algae biomass can offer synergistic benefits under specific conditions[156].

However, batch experiments with swine manure in Spain were studied, with increasing algae fractions (0 % and 14 % VS). The study found comparable or slightly reduced methane yields (246 and 238 L CH_4 kg^{-1} VS), indicating no positive synergistic effect from algae addition in these cases. Thus, no addition of algae in the anaerobic digestion is considered in this study [51].

2.6. End products

After anaerobic digestion, solid-liquid separation, composting and micro-algae cultivation, the products of the biogas production are compost, algae, biogas and water.

2.6.1. Compost

Within the European Union, fertilizing products are regulated under Regulation (EU) 2019/1009, which defines quality, safety, and nutrient requirements for products placed on the European market. Regarding the products that can be obtained from this study, organic soil conditioners and bio-fertilizers requirements are described.

Organic soil conditioners, also referred to as organic soil improver, are primarily intended to enhance soil structure and organic matter content. According to Regulation (EU) 2019/1009, these products must meet strict contaminant and hygiene limits. Apart from restrictions on heavy metals, the main requirements are: at least 20% organic matter on a dry mass basis and a minimum organic carbon content of 7.5% by mass. Apart form this, Organic soil conditioners must be free of hazardous pathogens.

Liquid organic fertilizers, often referred to as bio-fertilizers, are regulated based on their declared nutrient content. Such products must contain at least one primary nutrient, namely nitrogen (N), phosphorus (expressed as P_2O_5), or potassium (expressed as K_2O). When only one nutrient is declared, minimum concentrations of 2% N, 1% P_2O_5, or 2% K_2O by mass are required.

In cases where more than one nutrient is declared, lower individual thresholds apply, with minimum contents of 1% N, 0.5% P_2O_5, and 1% K_2O. However, the combined concentration of declared nutrients must be at least 3.5% by mass. In addition to nutrient requirements, liquid organic fertilizers must contain a minimum organic carbon content of 5% by mass.

When the compost is compared to the effect mineral fertilizers have, the nutrient efficiency of the application should be taken into account. Due to lower nutrient availability in compost, is can be assumed to have a use efficiency of 30% for both nitrogen and phosphorus [136]. Fertilizer substitution in LCA should be based on average national consumption patterns. This way, the fertilizer that is normally used, which is directly related to the needs of the soil, is taken into account.

2.6.2. Processed water

In Spain, wastewater discharge to surface waters is regulated under Order AAA/2056/2014, which establishes the legal framework for discharge authorization and monitoring. However, this regulation does not define fixed, universal concentration limits for pollutants such as nitrogen, phosphorus, or chemical oxygen demand (COD). Instead, discharge limits are determined on a case-by-case basis by the competent river basin authorities, taking into account the characteristics of the receiving water body, local hydrological plans, and applicable European directives.

In practice, reference values aligned with European standards are commonly applied. Typical discharge concentrations are approximately 125 mg L^{-1} for COD, 15 mg L^{-1} for total nitrogen, and 2 mg L^{-1} for total phosphorus. These values are indicative and may be subject to stricter requirements, particularly in environmentally sensitive areas[11, 105]

Therefore, while general benchmark values can be used for comparison and modeling purposes, the actual permissible discharge concentrations in Spain depend on site-specific regulatory conditions.

Potassium (K) is generally not subject to specific regulatory discharge limits in environmental policy, as it is not considered a primary driver of eutrophication compared to nitrogen and phosphorus.

2.6.3. Algae product

The produced algae can be used as an slow-release organic fertilizer that can directly replace energy-intensive synthetic fertilizers like urea [26], or as a protein sources in diets for livestock, poultry, and aquaculture [159, 134]. Algae should be further processed before it can be used as food-grade nutrition supplement [26].

To use algae instead of mineral fertilizers, the microalgal biomass requires accounting for the plant-available fraction of nutrients. Although microalgae such as *Chlorella vulgaris* contain substantial nitrogen and phosphorus, not all nutrients are immediately available for plant uptake. For nitrogen, microalgal biomass is considered an effective fertilizer, with a nutrient use efficiency of approximately 50% [21]. This relatively high availability is attributed to the mineralization of organic nitrogen into plant-available forms in the soil. In contrast, phosphorus availability is lower. A significant fraction is stored in stable forms, such as polyphosphate granules or metal-bound complexes, which require microbial degradation before becoming available. As a result, microalgae functions as a slow-release phosphorus fertilizer. Phosphorus fractionation studies indicate that only about 30% of the total amount of phosphorus is directly available, while the remainder is present in moderately available or stable forms [21]. These fractions are released gradually through soil processes, leading to delayed availability but improved long-term nutrient supply compared to conventional fertilizers.

The restrictions or guidelines for content in the products are summarized in Table 2.7.

Table 2.7: Summary of product requirements

Product Type	Parameter	Requirement
Soil Conditioner	Organic matter (dry mass basis)	$\geq$ 20 %
	Organic carbon	$\geq$ 7.5 % (by mass)
Biofertilizer	*If stated in total*	
	Nitrogen (N), Phosphorus (P_2O_5) or Potassium (K_2O)	$\geq$ 2 %
	If stated per nutrient	
	Nitrogen (N)	$\geq$ 1 %
	Phosphorus (P_2O_5)	$\geq$ 0.5 %
	Potassium (K_2O)	$\geq$ 1 %
	Combined declared nutrients	$\geq$ 3.5 %
	Organic carbon	$\geq$ 5 %
Processed Water (Effluent)	Phosphorus	$\leq$ 2 mg P/L
	Nitrogen	$\leq$ 15 mg/L
	COD	$\leq$ 125 mg/L
	BOD (after 5 days)	$\leq$ 25 mg/L
	Potassium (K)	No restriction

2.7. Bioplastic production

For the second treatment pathway, a process is proposed to produce polyhydroxyalkanoates (PHA) from swine manure. To identify a suitable process configuration, a literature review is conducted on volatile fatty acid (VFA) production from manure, PHA synthesis from VFA's, and the required downstream processing steps.

2.7.1. VFA production with acidogenesis

As explained before, anaerobic digestion naturally produces VFA's, by interrupting the anaerobic digestion pathway prior to methanogenesis in order to accumulate VFA's.

This process is commonly referred to as short-term anaerobic digestion or acidogenic fermentation, in which the process is limited to the hydrolysis and acidogenesis stages [84]. In contrast to full anaerobic digestion, the subsequent steps of acetogenesis and methanogenesis are suppressed. This "interruption" can be achieved through various operational strategies, including pH control, hydraulic retention time (HRT) reduction, and microbial community engineering, all of which aim to inhibit methanogenic activity while promoting acidogenic pathways [57].

The carbohydrates, proteins, lipids in swine manure are first hydrolysed into simpler compounds such as sugars, amino acids, and long-chain fatty acids. Subsequently, during acidogenesis, these intermediates are converted into short-chain VFA's, including acetate, propionate, butyrate, and valerate.

The composition of the substrate influences the resulting VFA profile, which in turn affects the yield and composition of the produced PHA.

Given the general composition of swine manure, the hydrolysis step can be approximated with equation 2.9.

$$\begin{aligned} \mathrm{CH_{2.6}O_{0.75}N_{0.054}} + aH_2O \longrightarrow & 57\%\,\mathrm{C_6H_{12}O_6} \quad \text{(Glucose)} \\ & 41\%\,\mathrm{C_5H_7O_2N_1} \quad \text{(Amino acid)} \\ & 2.2\%\,\mathrm{C_{57}H_{104}O_6} \quad \text{(Lipid)} \end{aligned} \tag{2.9}$$

Following hydrolysis, the acidogenesis step occurs. This stage proceeds through multiple metabolic pathways carried out by diverse microbial communities. Several representative acidogenic pathways are presented in Table 2.8.

	Pathway showns	*Source*
Propionibacterium freudenreichii 1.5 Glucose $\longrightarrow$ Acetate$^-$ + 2 Propionate$^-$ + CO_2 + 3 H^+ + H_2O	1 Acetate: $1CO_2$ 1 Propionate : $0CO_2$	[140]
Pyrococcus furiosus Glucose + 2 H_2O $\longrightarrow$ 2 Acetate$^-$ + 2 H^+ + 2 CO_2 + 4 H_2	1 Acetate: $1CO_2$: 2 H_2	[140]
Clostridium pasteurianum 1.5 Glucose + H_2O $\longrightarrow$ Acetate$^-$ + Butyrate$^-$ + 3 CO_2 + 4 H_2 + 2 H^+	1 Acetate : $1CO_2$: 1 H_2 1 Butyrate : 2 CO_2: 2 H_2	[140]
Clostridium propionicum 3 Alanine + 2 H_2O $\longrightarrow$ Acetate$^-$ + 2 Propionate$^-$ + CO_2 + 3 NH_4^+	1 Acetate: $1CO_2$ 1 Propionate: $0CO_2$	[140]
Clostridium luticellarii 2 Methanol + Propionate $\longrightarrow$ Valerate + 2 H_2O	1 Valerate: $0CO_2$	[27]

Table 2.8: Stoichiometric comparison of fermentation pathways of acidogenesis

The specific composition of these acids dictates the properties of the final plastic [57]. Even chain acids like acetic acid promote the synthesis of polyhydroxybutyrate, which is highly crystalline and rigid [57]. Odd chain acids like propionic acid facilitate the production of polyhydroxyvalerate, which provides flexibility and elasticity to the plastic [57].

Feasibility of swine manure as substrate

Huang et al. [62] demonstrated the feasibility of short-term dry anaerobic digestion for VFA production from swine manure. Their study showed that thermophilic operation at 55 °C and a total solids content of 20% enabled rapid acidification within 8 days. Under optimal conditions, total VFA yields reached up to 55.3 mg-COD/g-VS, with acetic acid as the dominant product. Temperature strongly influences microbial activity, metabolic pathways, and product distribution during short anaerobic digestion. Acidogenic fermentation can be operated under psychrophilic (<25 °C), mesophilic (30–40 °C), or thermophilic (50–60 °C) conditions, each resulting in distinct VFA production characteristics [62]. According to Hidalgo et al. [57], mesophilic and thermophilic regimes are generally preferred for targeted VFA production due to their higher reaction rates and improved substrate conversion efficiencies.

In continuous stirred tank reactors (CSTRs), hydraulic retention time (HRT) plays a decisive role in determining both VFA concentration and composition. The study presented by Wang et al. [157] explicitly demonstrates that HRT strongly influences the balance between acidogenesis and methanogenesis. Short HRTs suppress the growth of slow-growing methanogens, thereby enhancing VFA accumulation. However, excessively short HRTs may reduce hydrolysis efficiency and overall substrate conversion. Conversely, longer HRTs improve substrate degradation but increase the risk of VFA consumption by methanogens.

The findings of Wang et al. [157] therefore indicate the existence of an optimal HRT window in CSTR operation around 1.5 days of HRT and 6065 mg-COD/L OLR. Acetate is typically reported as the dominant acid, followed by propionate and butyrate, although the distribution can shift depending on reactor conditions [157]. High organic loading rates and excessive VFA accumulation were shown to inhibit microbial activity and compromise process stability[157]

The pH of the fermentation medium is one of the most important parameters governing volatile fatty acid (VFA) production during acidogenic fermentation. Hidalgo et al. [57] report that optimal acidogenesis generally occurs at moderately acidic conditions, with an ideal pH range between 4.5 and 6.0. However, if specifically swine manure is studied, it is shown by Sun et al. [142], Wang et al. [157], and Huang et al. [62] that elevating the pH to a value of 10 enhances volatile fatty acid (VFA) production from swine manure as substrate solubilization and microbial activity are improved. [62].

Apart from pH adjustment, the NH_4-N concentration must also be controlled, as it should not exceed 1.2 g/L, as high ammonia concentrations, and in particular elevated NH_3 concentrations, can inhibit the microorganisms involved in fermentation. [157]. In the manure data used in this study, the ammonia concentration is approximately 2 g N/L. In the experimental study by Wang et al. [157] a NH_4-N concentration of approximately 0.224 g/L was reported, which is nearly an order of magnitude lower.

Considering these sources were all in lab-scale, an industrial or pilot-scale measurements are more reliable for this study. A pilot or industrial scale study on the production of PHA from swine manure has not been found, but for the treatment of dairy manure Guho et al. [54] executed a pilot-scale operation. The study is done in the United States on dairy manure obtained form the region. The study is done with the least amount of control in the reactors, so no control on the pH, temperature or OLR is executed. A HRT of 4 days is applied, compared to the 1.5 days of Wang et al. [157]. A much higher organic loading rate is measured but the VFA production is lower. The temperature and pH are uncontrolled. The VFA profile of the fermentation is reported to be similar to that obtained from the acidogenesis of swine manure, only the amount of acetate is much higher from the dairy manure. Given the use of dairy manure in the pilot-study, this study is not in line with the swine manure treatment. Thus, no pilot-scale but only lab-scale data is used.

2.7.2. Ammonia stripping as pretreatment

Because of the observed increased yield in pH 10 and need for ammonia reduction, a pretreatment step of ammonia stripping is implemented before the acidogenesis. The efficiency of ammonia stripping is primarily governed by pH and temperature. Typically, the pH is adjusted to values between 9 and 10 to favor the formation of gaseous ammonia, while higher temperatures enhance the stripping process. Under optimal conditions, ammonia removal efficiencies of up to 95% can be achieved[132]

The practical implementation of pH control is described by Rodrigues et al. [127], who compared different alkaline agents for manure pretreatment and sanitization, including NaOH, KOH, and $Ca(OH)_2$, on swine manure with 3.9 % TS in Portugal. Both NaOH and KOH were found to be effective in increasing the pH to the desired alkaline range, while also improving substrate solubilization and hygienization. Although NaOH is economically attractive due to its lower market price, its use results in sodium accumulation in the digestate. Excess sodium may contribute to soil salinization after land application, negatively affecting soil structure and crop productivity.

In contrast, potassium hydroxide (KOH) does not pose this disadvantage. Potassium is an essential macronutrient for plants and can contribute positively to soil fertility when digestate is applied as fertilizer [127].

2.7.3. S/L separation

After the fermentation, the effluent consists of VFA products, water and most-likely un-reacted pig manure and other solids. To extract the VFA product, solid-liquid separation process can be used [54, 30]. In the pilot-scale PHA production on dairy manure by Guho et al. [54], filtration is performed using a steel mesh screen with a pore size of 0.5 mm. Subsequently, a centrifuge is applied to separate the liquid and solid fractions. Finally, a second filtration step with a pore size of 10 μm is used to further remove fine suspended solids.

2.7.4. Storage

After solid-liquid separation, the liquid fraction containing the produced VFA's is stored under ambient conditions in the pilot-study of Guho et al. [54]. A storage time of approximately 3 to 5 days was applied, as the VFA production reactor was too small to ensure continuous downstream operation in this operation.

Although the scale of the reactor can be adjusted, inclusion of a storage step remains recommended. The OLR applied by Guho et al. [54] are low, thus small volumes need to be applied to obtain this. As a result, only a limited amount of VFA-rich substrate can be supplied at a time. A storage unit is therefore necessary to buffer the VFA liquid and control the following steps precisely.

2.7.5. PHA production from VFA

In the three-stage PHA production system described by Guho et al. [54], enrichment and accumulation reactors are used. The enrichment reactor is designed to impose feast–famine selection pressure to obtain a PHA-storing microbial bacteria, whereas the accumulation reactor is designed to maximize PHA content through sustained substrate availability in a fed-batch regime [54].

Regarding the sustainability goals and economically feasibility of this study, only mixed-cultures are evaluated, as pure microbial cultures are expensive and require strict sterilization[163].

Enrichment phase

The volatile fatty acids produced from the swine manure can be use to enrich a mixed microbial culture. The purpose of the enrichment reactor is to selectively cultivate a mixed microbial consortium (MMC) capable of rapid feast–famine PHA synthesis [54] . Mixed microbial cultures derived from activated sludge are used by multiple studies [151]. This sludge can be placed in a sequencing batch reactor and undergoes a domestication phase using a feast and famine strategy [163, 109, 30] During the famine state, the microorganisms convert excess carbon into PHA for storage, while in the feast state, the bacteria that can synthesize PHA survive and multiply [163]. This cycle successfully enriches the sludge with bacteria capable of producing the plastic [145, 163, 54, 22, 109, 30] .

By Coats, Watson, and Brinkman [22], studied the aeration needed for the enrichment phase in a sequencing batch reactor (SBR) operated under aerobic dynamic feeding (ADF) conditions using VFArich fermented dairy manure as substrate. Four aeration regimes were evaluated ($k_L a$ = 4, 8, 12, and 20 h^{-1}). Although the dissolved oxygen (DO) profile did not show a sharp famine transition at low $k_L a$, feast–famine metabolic behavior was clearly maintained. This indicates that strict high-DO conditions are not required for enrichment [22].

An important advantage of VFA-rich effluents is their favorable carbon-to-nitrogen ratio. Ammonium concentrations of approximately 0.5–1.0 g/L allow the effluents to serve as the sole nutrient source during PHA accumulation, facilitating nitrogen limitation while maintaining high carbon availability. This condition enhances polymer storage and suppresses biomass growth [115].

The conditions applied in the pilot-scale study are summed in Table 2.9.

Table 2.9: PHA enrichment phase operator parameters of pilot-scale dairy manure PHA production plant[54]

Parameter	Value
Reactor type	680 L baffled conical-bottom tank
Inoculum	Activated sludge from a water reclamation facility
Cycle length	24 h
Aeration rate	280 to 350 L/min
OLR	0.52 g COD VFA L^{-1} d^{-1}
SRT	of 4 days
Active biomass (C_X)	2.5 $\pm$ 0.2 g/L
Observed yield	0.8 g COD-PHBV/g COD-VFA

Accumulation reactor

In MMC-based PHA production systems, the accumulation reactor is designed to maximize intracellular polymer storage in an already enriched microbial culture and is typically implemented as the third process step [158, 109, 54].

The accumulation reactor is generally operated as a batch or fed-batch system. In pilot-scale dairy manure systems, a fed-batch configuration is used in which enriched biomass is supplied with repeated pulses of VFA-rich fermenter liquor [54]. Similar strategies are applied in Organic Fraction of Municipal Solid Waste (OFMSW) based systems and laboratory studies, where pulse-wise substrate addition is often controlled based on dissolved oxygen (DO) levels [30, 109]. DO signals are commonly used for control: oxygen decreases during substrate uptake and increases sharply upon depletion, triggering subsequent feeding [54, 109]. This ensures continuous feast conditions, promoting PHA accumulation rather than microbial growth or selection [30]. Dissolved oxygen is typically maintained above 1–2 mg L^{-1} to avoid oxygen limitation, enabling rapid conversion of VFA's into intracellular PHA [54, 109]. Maintaining a neutral pH between 7.0 and 8.0 is considered optimal [145].

Typical yields are around 0.5 $gCOD_{PHA}/gCOD_{VFA}$, with PHA contents reaching 60–70% under optimal conditions, while pilot-scale systems achieve 40–50% of cell dry weight [55, 30, 158]. However, performance strongly depends on substrate composition. High concentrations of non-VFA organics or nutrients (e.g., nitrogen and phosphorus) can promote biomass growth instead of PHA storage [109]. Therefore, centrifugation or filtration are often applied to obtain a clarified, VFA-rich feed stream [54, 30]. The operation parameters and yield of Guho et al. [54] are shown in Table 2.10.

Table 2.10: PHA enrichment phase operator parameters of pilot-scale dairy manure PHA production plant[54]

Parameter	Value
Reactor type	450 L baffled fed-batch tank
Feeding strategy	3 to 7 pulses of fermenter liquor
Production duration	5.0 to 8.5 h
Observed yield	0.58 to 0.87 g COD-PHBV/g COD-VFA
Final PHBV fraction	0.29 to 0.40 g/g volatile solids

2.7.6. PHA extraction from the biomass

After intracellular synthesis, PHA must be extracted and purified to obtain a usable product, requiring separation of the polymer from residual biomass [57]. Downstream processing (DSP) is a major cost contributor and often relies on toxic and energy-intensive methods, making the development of sustainable alternatives essential for economic competitiveness [145, 99].

Various extraction strategies, including chemical, mechanical, enzymatic, and supercritical methods, offer trade-offs between simplicity, scalability, environmental impact, and polymer quality [99]. Among these, mild alkaline extraction (e.g., NaOH) has emerged as a promising alternative that preserves polymer properties while reducing solvent use, although further optimization is required for industrial application [49].

For manure-based PHA production, DSP is less extensively described. Wu et al. [158] report full-scale recovery routes involving chemicals such as H_2SO_4, NaOH, and SDS, but without specifying approaches. In contrast, Guho et al. [54] describe a detailed process including freezing, lyophilization, acetone washing, filtration, and drying, followed by chloroform extraction and multi-step purification using petroleum ether and methanol. This method achieves high purity (94–96%) but moderate recovery (65–70%) and relies on multiple solvents.

A simpler approach is reported by Wu et al. [158], where freeze-dried biomass undergoes sodium hypochlorite digestion, chloroform extraction, and centrifugation, with solvent recovery reaching approximately 85%. This method achieves 99% purity and 85% recovery. A similar sequential digestion and solvent extraction strategy is described by Mudliar et al. [110], involving biomass concentration via centrifugation, chemical lysis using hypochlorite (NaOCl), and subsequent chloroform extraction, filtration, and solvent evaporation.

In these processes, chloroform is typically recovered and recycled (85–90% efficiency) due to its high PHA solubility and relatively straightforward recovery [158, 110]. While the detailed manure-based process is considered representative, its complexity and solvent use limit sustainability. Therefore, simpler sequential digestion and extraction methods are preferred, although further development of greener alternatives remains necessary. The process is summarized in Table 2.11

Table 2.11: Downstream processing of sequential digestion and solvent extraction method [158, 110]

Step	Description
Biomass recovery	Separation of biomass from fermentation broth using a centrifuge
Cell disruption	Treatment with sodium hypochlorite (NaClO) to lyse cells and release intracellular PHA
Solvent extraction	Addition of chloroform to dissolve PHA from the lysed biomass
Phase separation	Centrifugation to separate organic (chloroform + PHA) and aqueous phases
Solvent recovery	Chloroform recovery system applied (approximately 85–90% recovery efficiency)
Polymer precipitation	Precipitation of PHA from the chloroform phase after solvent regeneration
Purification	Removal of residual impurities through washing and separation steps
Final processing	Drying of purified PHA to obtain the final polymer product

2.7.7. Ammonia concentrations

As discussed, for the acidogenesis, PHA production and micro-algae cultivation, the ammonia concentration should be monitored as too high values lead to inhibition. The ranges are summarized in Table A.2 in Appendix A.

2.7.8. End products

The end products of the bioplastic production process are algae, compost and PHA powder. The same regulations and application efficiencies apply as in the biogas production for the compost and algae. The quality of the PHA is determined by the PHA extraction method, which is a purity of 99% which can be sold in this form as powder.

To compare PHA to conventional plastic, the functionality should be evaluated first. To determine an appropriate reference product, the material properties and potential applications of the produced PHA are considered. Li et al. [92] states that the glass transition temperature (T_g) of a suitable packaging polymer should be low enough to prevent degradation during freezing, while also being high enough to remain stable at elevated temperatures (>120 °C). In addition, Guho et al. [54] reports that the purest PHA sample has a T_g of -6 °C and first and second melting peaks at 130.9 and 153.5 °C, respectively. These properties indicate that the material is suitable for applications such as food packaging. Conventional plastics that it could replace, can be polyethylene terephthalate (PET), the equivalent substitution ratio between PHA and conventional plastic is described by Del Oso et al. [30], who assume that 1 kg of PHA replaces 0.72 kg of PET. This is largely based on density [5, 14].

By [142], the end-of-life of PHA is also investigated, in which study bioplastics are subjected to anaerobic digestion, in which PHA is biologically degraded into simpler compounds. If carefully managed, the co-digestion of PHA with swine manure can be highly effective ”upcycling” strategy to convert plastic waste back into valuable platform chemicals like VFA’s or biogas.

3 Materials and method

In this section, the data used and method for calculations is explained for both digital twins of manure treatment for biogas and bio-plastic production. The calculations in both models are done per 1000 kg manure. The design choices are made based on the information in the Literature review (section 2).

3.1. System definition

The average of swine manure data from Spain in Table 2.3 is taken as input data in this study, shown in Table 3.2. [47] [125, 107].

Table 3.1: Data used in this study

Parameter	
Density [kg/L]	1.00
pH	7.4
TS [g/L]	24.7 ± 21.9
VS [g/L]	22.8 ± 14.6
Total Nitrogen (TN) [g N /L]	2.6 ± 0.9
Ammonium (NH_4-N, TAN) [g N /L]	2.0 ± 0.7
Organic N (TON) [g N/L]	0.6 ± 1.1
Phosphate (TP) [g/L]	0.6 ± 0.6
Potassium (K) [g/L]	3.5 ± 2.0
COD [gO_2/ kg]	32.8
Elemental composition	$C_1H_{2.6}O_{0.75}N_{0.054}$

The following assumptions were considered:

1) Sulfur content is not considered because this varies given the feed intake and this is outside of the scope. S_2O emissions caused by biogas combustion are considered .Considerations for micro-nutrient contents and heavy metals are left out the scope for the same reasons.
2) The density of manure, digestate, and the liquid fraction after centrifugation is assumed to be 1000 kg m^{-3} for all relevant streams.
3) A volumetric gas constant of 24 L mol^{-1} at 20°C and 1 atm is used. The density of methane is assumed to be 0.714 kg m^{-3}, the density of CO_2 is 1.98 kg m^{-3} and density of H_2 is 0.081 kg/m3 based on biological standard pressures and temperatures.
4) The volatile solids are assumed to be equal to the biodegradable fraction, a biodegradability factor is applied to estimate the actual degraded fraction.
5) For literature values used in this study, swine and pigs are considered as the same [76]. If difference in breeding stage is stated in the reference, finishing swine data is used in this study. For data use of literature, parameters are exclusively taken from liquid swine manure, with a TS

fraction between 1-5 %, preferably from research in Spain. If no statement is done in a reference source, the density of 1 kg/L is assumed for liquid swine manure.

To answer the research question, a process model for both scenarios is build in Python, executing all calculations noted in this Method section. The same manure data is used for both models.

- **Biogas production**: Anaerobic digestion to biogas with composting of the solid fraction of the digestate and micro-algae cultivation on the liquid fraction of the digestate.
- **Bioplastic production**: Pretreatment of ammonia stripping, acidogenesis of manure and PHA accumulation on the produced VFA's for bio-plastic production. Rest stream treatment with composting and micro-algae cultivation similar to biogas scenario.

Both scenarios are based on a farm-scale system processing 8,500 tons of swine manure per year, corresponding to a volumetric inflow of 1.07 $m^3\ h^{-1}$, located in Spain. No transport was considered for the manure input. Calculations in the models are done per ton manure treated.

3.1.1. Biogas production model

The biogas production model is based on stoichiometric modeling of the anaerobic digestion. All emissions are implemented in the model for conservation of mass and elements. The total mass and the water, TS, VS, TAN, TN, P, K, C and ash contents are calculated per step.

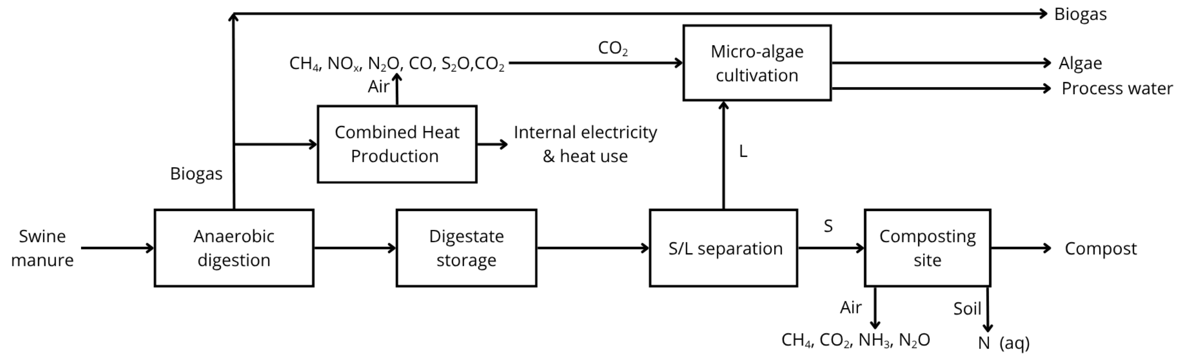


Figure 3.1: Block flow diagram of biogas production scenario.

Table 3.2: Data used in biogas production process

Parameter	Value
AD reactor	Continuous
Operator temperature [Celsius]	37
TS in reactor [%]	original 2.5
Biodegradability	30 - 70

To evaluate possible higher of yields, related to optimal conditions, the range of the biodegradability is stated. The 30% discussed in the Literature review is taken as a base value. For anaerobic digestion, the IPCC reports that the theoretical maximum biodegradability of swine manure is approximately 70% [80]. This implies that only 70% of the theoretical methane potential can be achieved in practice.

Anaerobic digestion

The core of the digestion process is driven by a complex microbial community, with methanogens performing the final step of methane production. To model this biological process, a macroscopic mass balance approach is applied. Although this method introduces some uncertainty, it provides a robust approximation of the stoichiometry of the system. The elemental composition of the substrate determines the stoichiometry of the electron donor and acceptor reactions. For swine manure with an elemental composition of $CH_{2.6}O_{0.75}N_{0.054}$, the degree of reduction (γ_S) is calculated as 4.9 e^-/C-mol. Based on this composition, the following half reactions are established:

The Donor half-reaction (R_d) describes the oxidation of the organic substrate into carbon dioxide:

$$\mathrm{CH_{2.6}O_{0.75}N_{0.054} + 1.3\,H_2O \longrightarrow CO_2 + 0.054\,NH_3 + 4.9\,H^+ + 4.9\,e^-} \tag{3.1}$$

The Acceptor Half-reaction (R_a) describes the reduction of carbon sources to methane:

$$\mathrm{CH_{2.6}O_{0.75}N_{0.054} + 3.1\,H^+ + 3.1\,e^- \longrightarrow CH_4 + 0.054\,NH_3 + 0.75\,H_2O} \tag{3.2}$$

By coupling the donor and acceptor half reactions to balance the electrons, the overall catabolic reaction (Cat) is derived. This reaction represents the breakdown of the substrate purely for energy generation:

$$\mathrm{CH_{2.6}O_{0.75}N_{0.054} + 0.016\,H_2O \longrightarrow 0.62\,CH_4 + 0.38\,CO_2 + 0.054\,NH_3} \tag{3.3}$$

Simultaneously, a portion of the substrate is assimilated into biomass. The anabolic reaction (An) describes the synthesis of biomass ($CH_{1.8}O_{0.5}N_{0.2}$). Since the degree of reduction of the substrate

($\gamma_S = 4.9$) is higher than that of biomass ($\gamma_B = 4.20$), the synthesis process releases electrons, which are typically disposed of by a formation of methane (mixotrophic growth):

$$1.2\,\mathrm{CH_{2.6}O_{0.75}N_{0.054}} + 0.13\,\mathrm{NH_3} \longrightarrow \mathrm{CH_{1.8}O_{0.5}N_{0.2}} + 0.24\,\mathrm{CH_4} + 0.43\,\mathrm{H_2O} \tag{3.4}$$

The total metabolism (Met) is the sum of the catabolic and anabolic reactions. The ratio between them depends on the energy required for growth. This relationship is defined by the coupling factor λ_{cat}, which represents the moles of catabolic reaction required per mole of anabolic reaction (eq. 3.5). [84].

$$Met = \lambda_{cat} \cdot Cat + An \tag{3.5}$$

The value of λ_{cat} can be determined using the observed methane yield ($Y^{obs}_{CH4/PM}$) and the stoichiometric coefficients derived above, following the thermodynamic state analysis method described by [86]. The theoretical methane yield of 495.1 L CH_4 / $kgVS$ is taken, recalculated to -0.551 mol CH4 / mol VS.

$$\lambda_{cat} = \frac{Y^{An}_{CH4} - Y^{Met}_{CH4/PM} \cdot Y^{An}_{S2}}{Y^{Met}_{CH4/PM} \cdot Y^{Cat}_{PM} - Y^{Cat}_{CH4}} \tag{3.6}$$

Combining the determined λ_{cat}, with the catabolic and anabolic reaction, the overall metabolic reaction for the system can be established.

$$10\,\mathrm{CH_{2.6}O_{0.75}N_{0.054}} \rightarrow 1.00\,\mathrm{CH_{1.8}O_{0.5}N_{0.2}} + 5.7\,\mathrm{CH_4} + 3.4\,\mathrm{CO_2} + 0.34\,\mathrm{NH_3} + 0.29\,\mathrm{H_2O} \tag{3.7}$$

The biodegradability represents the fraction of volatile solids (VS) in the pig manure that reacts according to the defined metabolic pathway. This results in a portion of the pig manure being converted into metabolic products, while a remaining fraction stays unreacted. A biodegradability of 30% is used, an approximation based on reported values by [50].

The unreacted manure, together with the biomass formed during the metabolic process, is considered as digestate after digestion. Based on the mass contributions of these components, an average composition of the digestate is determined.

The emissions from the anaerobic digestion (AD) process are estimated based on emission factors reported by the IPCC. The applied emission factors and their descriptions are summarized in Table 3.3.

Table 3.3: Emission factors applied for anaerobic digestion of swine manure.

Gas	Emission factor	IPCC description
CH_4	1.0%	of methane produced
N_2O	0.0	EF_3 [kg N_2O / kg N]
NH_3	4.65%	$FRAC_{Gas}$ (swine), NH_3:NO_x = 93:7
NO_x	0.35%	$FRAC_{Gas}$ (swine), NH_3:NO_x = 93:7
Leaching N	0.0%	$FRAC_{Leach}$ (swine)
N_2	0.0	3 $\times EF_3$ [kg N_2O / kg N]

Although the IPCC reports an emission factor of 0.0006 for N_2O, this value is primarily associated with emissions during manure storage rather than during the digestion process itself. Since storage emissions are treated separately in this study, N_2O emissions from the AD reactor are assumed to be negligible and are therefore set to zero.

The partitioning of gaseous nitrogen emissions into NH_3 and NO_x is based on a ratio of 93:7, as reported by [152], which follows IPCC guidelines. This ratio is applied to distribute total gaseous nitrogen emissions between NH_3 and NO_x.

As a high-quality anaerobic digester is assumed, no nutrient leaching is considered to occur during the digestion process. Nitrogen gas (N_2) emissions do not contribute to impact categories in life cycle assessment. For mass balance purposes, the formation of N_2 can are related to N_2O emissions by the IPCC. As, no N_2O emissions are assumed to occur, N_2 emissions are not estimated.

For the fractions and emission factors of nitrogen emissions, the following equation 3.8 is used.

$$Emission = FRAC_{gas} * TNinflow \tag{3.8}$$

CHP

Given the metabolic reaction, the composition of the biogas can be determined with equations 3.9 & 3.10 [84].

$$x_{CH_4} = \frac{n_{CH_4}}{n_{CH_4} + n_{CO_2}} \tag{3.9}$$

$$x_{CO_2} = \frac{n_{CO_2}}{n_{CH_4} + n_{CO_2}} \tag{3.10}$$

By IPCC [76] a leaking factor of 1.0 % of the methane is stated, which is representative for a high quality AD reactor, with exclusion of storage emissions[76].

In the CHP, electricity is partly used to meet internal energy demands, while surplus power may be exported. Duan et al. [32] assumes in the combustion of biogas from swine manure an electrical and thermal efficiencies of 39% and 46%, respectively, resulting in a total energy efficiency exceeding 80% [32].

The emission factors for the combustion of raw biogas in internal combustion engines, used for combined heat and electricity production, are derived from Duan et al. [32] and are presented in Table 3.4.

Table 3.4: Emission factors for biogas combustion in internal combustion engines (per kg CH_4) [32].

Off-gas	Emission factor (g/kg CH_4)
SO_2	0.03
NO_x	4.42
NMVOC	0.16
CH_4 (biogenic)	0.08
CO	1.21
CO_2 (biogenic)	4.47 $\times 10^3$
N_2O	0.08
NH_3	0.00
TSP	0.01
PM_{10}	0.01
$PM_{2.5}$	0.01

The emission factor of NH_3, TSP, PM_{10} and $PM_{2.5}$ are however too low to have significant emissions in the system studied. Non-methane volatile organic compounds (NMVOC) emission contribute to impact categories that are not discussed in this report, so these emissions are not calculated.

Digestate storage

For the digestate storage, open storage is assumed , leading to only emissions to air and leaching of liquid to soil. Because the composition of digestate differs from that of raw manure, different emission factors must be applied. The variation in emissions between raw and digested manure is quantified based on measurements conducted at European cattle and pig farms by Kupper et al. [89]. These results are presented in Table 3.5, together with the resulting emission factors for digestate derived for European conditions.

Table 3.5: Emission factors for solid storage of pig manure in its raw state and after anaerobic digestion. A positive sign (+) indicates an increase in emissions, while a negative sign (–) indicates a decrease.

Gas	Emission factor	IPCC description for solid storage	Effect of digestion	Result
N_2O	0.01	EF_3 [kg N_2O/kg N]	+363%	0.0363 kg N_2O/kg N
CH_4	4%	MCF (warm temperate climate)	–99%	MCF = 0.04%
NH_3	45%	$FRAC_{Gas}$(swine) (NH_3 & NO_x)	–45%	0%

Nitrogen leaching is assumed to be equal to the default IPCC value for storage, i.e. 2% of the initial TN content of the manure, as the leaching factor is based on management practices, like how often the piles are aerated or other causes.

N_2 emissions are estimated based on N_2O emissions. According to the IPCC, the amount of N_2 emitted is assumed to be three times the amount of N_2O emitted. [76].

Solid-liquid separation

The average separation values presented in Table 2.5 for a decanter are used to model the solid-liquid separation. No additional processes, such as emissions, are considered. The electricity demand of the decanter is assumed to be 4 kWh/m^3 [32]. The liquid fraction is directed to the micro-algae pond, while the solid fraction is sent to the composting site.

Composting

In this study, composting is modeled as an enclosed windrow system with periodic turning to ensure adequate aeration. Data of composing of co-digested manure in Germany is used reported by Rehl and Müller [124]. Mixing is performed using a diesel-powered machine with a processing capacity of 600 m^3 h^{-1} and a fuel consumption of 10 L h^{-1}. As no continuous turning has to take place, the relation of Duan et al. [32] is used resulting in 2.8 L diesel per ton input.

The composting process is assumed to result in a loss of 50% of the initial carbon content and 80% of the initial water content. For gas emissions, 8.12% methane, 41.88% carbon dioxide, 20 % NH4-N ammonia and 1.4 % N_2O is assumed. The assumptions of the IPCC are taken for N_2 emissions, resulting in 4.2 % of TN content, and nitrogen leaching to the soil of 4% for passive composting of swine manure [76].

Unlike nitrogen, which is highly reactive and easily lost during composting through ammonia volatilization or nitrate leaching, the assumption is that phosphorus loss during the active composting process is negligible or rare [28]. During passive composting, phosphorus leaching could be observed due to water leaching or poor management. But, Hollas et al. [59] and Shin, Kim, and Moon [136]measure zero emissions of phosphorus to the environment. This, the assumption is done that no phosphorus leaching is used in this process step. Potassium and ash also are assumed to be completely conserved.

The NO_x emissions were assumed to be zero, as life cycle assessments of Hollas et al. [59] and Hollas et al. [60] give no mention of these emissions next to de sources discussed. The total nitrogen loss is determined as the sum of losses via NH_3, N_2O, N_2, and leaching.

Table 3.6: Composting site dataRehl and Müller [124]

Item	Value	Unit
Water loss	80 %	of initial water content
C loss	50 %	of initial C content
NH_3 emission	20 %	of initial NH_4 content
N_2O emission	1.4 %	of initial TN content
N_2 emissions	4.2 %	of initial TN content
CH_4 emission	8.12%	of initial C content
CO_2 emissions	41.9 %	of VS content
$N_{leaching}$	4%	of initial TN content

The difference of the used emission factors in this study between raw manure and digestated swine manure in Brazil are shown in Appendix A.

Micro-algal cultivation

Micro-algal growth is modelled as a photosynthetic conversion process in which inorganic carbon (CO_2), volatile solids and ammonium are converted into algal biomass. Since the cultivation takes place in an aqueous environment, dissolved inorganic carbon may be present partly as CO_2 and partly as HCO_3^-, depending on the pH. For simplicity, the growth reaction is written here using CO_2 as carbon source. Ammonium is assumed to be the nitrogen source, which is consistent with the composition of the liquid fraction after anaerobic digestion. When ammonium is assimilated into biomass, protons are formed [155].

For the estimation of the mass balance, the elemental composition of a micro-algae species of Chlorella estimated by Saidu et al. [130] is used. As the sulfur content is very small (0.82% S in elemental composition) and sulfur was not taken into account before, this is neglected.

The digestate composition after the anaerobic digestion reactor is used as an approximation for the volatile solids supplied to the cultivation system. This composition is taken as $C_1H_{2.54}O_{0.73}N_{0.018}$, which is close to the reported composition of pig manure.

Palomar et al. [116] report a relation of 1.83 g CO_2 per g of algal biomass formed during photosynthesis. This ratio can be used to estimate the stoichiometric coefficients in the growth reaction, shown in equation 3.11.

With this information, the production of algae is visualized in equation 3.11.

$$\begin{aligned} -0.019\, C_1H_{2.56}O_{0.74}N_{0.060} \; - 0.98\, \mathrm{CO_2} \; - 0.13\, \mathrm{NH_4^+} \; - 0.73\, \mathrm{H_2O} \\ +0.13\, \mathrm{H^+} \; + 1\, C_1H_{1.8}O_{0.5}N_{0.2} \text{ (algae)} \; + 1.1\, \mathrm{O_2} \end{aligned} \tag{3.11}$$

Ammonium limitation and water addition

As reported by Duan et al. [32] and Zhou et al. [164], elevated ammonia concentrations can inhibit micro-algal growth. Therefore, the cultivation process is constrained by a maximum allowable total ammonium nitrogen (TAN) concentration in the liquid phase. In this study, a target concentration of 3.3 g NH_4-N per L is applied.

$$m_{water,add} = \frac{m_{NH_4-N}}{c_{TAN}} - m_{liquid} \tag{3.12}$$

The liquid fraction obtained after the decanter centrifuge contains a relatively high TAN concentration. To meet the required threshold, dilution with water is necessary.

Specifically, freshwater addition is limited to 5% of the total dilution water demand, while the remaining water is supplied through internal recycling.

Limiting nutrient

In the stream entering the cultivation pond, total nitrogen (TN), ammonium (NH_4^+), and phosphorus (P) are the primary nutrients that may limit micro-algal growth. Carbon in the water can also be a limiting factor, but given the ability to assimilate CO_2 of the algae, carbon is not considered limited. To identify the limiting nutrient, the elemental composition of the algae is first considered.

Using the values from Table 2.6, the biomass composition of *Chlorella* is determined to be 28.4% C, 8.26% N, and 1.82% P of the TS. From this, the N:P ratio in the algae can be derived. This results in an approximate ratio of 4.5:1 (N:P), meaning that 4.5 units of nitrogen are required per unit of phosphorus for algal growth. [55]

With the relation of N and P in the inflowing liquid fraction of digested swine manure, the limiting nutrient can be determined, which can be calculated as shown in equation 3.13.

$$N : P\ inflow = \frac{[TN_{inflow}]}{[P_{inflow}]} : 1 \tag{3.13}$$

Algae biomass production
Based on the literary review, removal efficiencies of 84% for nitrogen, 91% for phosphorus, and 91% for ammonium are assumed for Chlorella in this study. It should be noted that these values are derived from laboratory-scale experiments and may overestimate performance at larger scales.

The removal efficiencies are used to estimate nutrient uptake by the algae and, consequently, the theoretical biomass production. By Guillen et al. [55] a harvesting efficiency of 85.5% for micro-algal cultivation systems is estimated. With the limited nutrient as X, the amount of algae produced can be estimated with equation 3.14.

$$Algae\ produced = \frac{X\ uptake\ efficiency\ [\%] * X\ available\ [kg]}{X\ fraction\ in\ algae} * Harvesting\ efficiency[\%] \quad (3.14)$$

It should be noted that both the assumed substrate composition and the applied removal efficiencies introduce uncertainty into this estimation. Nevertheless, this approach provides a approximation of biomass production under the given conditions.

For the volatile solids in the pond a removal rate of 86 % is assumed, observed by Li et al. [93].

No emissions were measured at the pond in a similar process of micro-algae cultivation, in which also the liquid fraction of swine manure (from the same region in Spain) is used for micro-algae cultivation in an open pond. Only only the consumption of CO_2 and even H_2S were described from the measurements done on the pilot plant in 2023. The design of the pond was open. [101]

The water leaving the pond after cultivation remains contaminated; therefore, the concentrations of nitrogen (N), phosphorus (P), and chemical oxygen demand (COD) in the effluent are quantified and included in the analysis.

3.1.2. Bioplastic production
In the bioplastic production treatment, as is explained in the literary review, a pretreatment step of ammonia stripping needs to be applied, after which acidogenesis is taking place for VFA production. The VFA containing liquid is centrifuged and stored, after which it is used for PHA enrichment and accumulation. The PHA rich biomass is extracted using a chloroform and sodium hypochlorite. Solid biological waste is composted and nutrient rich water is led into a micro-algae pond for nutrient retention in algae. The process is shown in Figure 3.2.

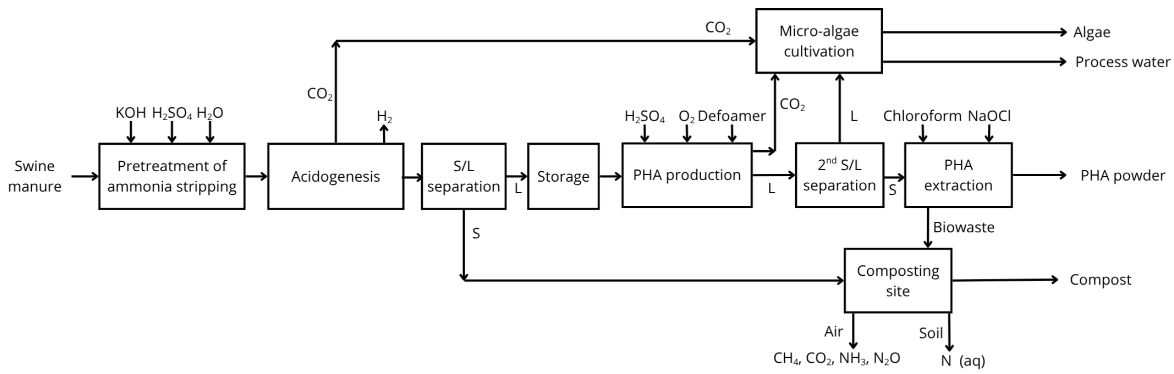


Figure 3.2: Block flow diagram of bio-plastic production scenario.

The key parameters used are shown in Table 3.7. The efficiency of the theoretically possible acidogenesis degree is later explained.

Table 3.7: Data used in this study

Parameter	**Value**	Source
Acidogenesis reactor	Continuous	[157]
TS in reactor [%]	2.2	[157]
Acidogenesis degree [% gCOD VFA/ g COD in]	0.383	[157]
Efficiency acidogenesis range [%]	54 - 99	section 3.1.2
OLR enrichment	0.52 g COD VFA L^{-1} d^{-1}	[54]
PHBV yield [mol_{PHA}/mol_{VFA}]	0.1859	[54]
Recovery yield PHA extraction [%]	85	[158]

Pretreatment

Pretreatment of the manure is modelled through pH adjustments combined with ammonia stripping. Potassium hydroxide (KOH) is used as the alkaline agent, despite the slightly higher material cost, given the improvement of the agronomic value of the final digestate product. Based on experimental measurements with swine manure from Portugal with similar characteristics as the studies swine manure in this study, a dosage of 11 g KOH per kg manure is applied to achieve a pH of 10 [127].

The equilibrium relationship between NH_4^+ and NH_3, as described in eq. 2.4, is used to calculate the fraction of free ammonia (NH_3) at pH 10. Under operating temperature of 20 °C, an ammonia removal efficiency of 95% can be assumed [132].

To ensure consistency with the operating conditions reported by [157], the total solids (TS) content is adjusted to 2.2%. Since KOH is applied in solid form, additional water is assumed to be added to the system to achieve the desired TS content and to ensure adequate mixing.

Zero leaking of gasses is assumed in this study.

VFA production

For the production of VFA's from swine manure, the conditions from Wang et al. [157] are used, as this report researches a continuous system for VFA production from specific swine manure. It should be noted that the manure from this study is not from the studied region, but the TS and TAN are considered key and considerably the same as this study characteristics. The study applies pH adjustment to 10 , but no pH control is executed during the acidogenesis.

Given the complexity of acidogenesis and the unknown efficiencies of the individual microbial pathways involved, the observed VFA composition of Wang et al. [157] is used, rather than the individual metabolic pathway conversions. This mass distribution was 34.8 % acetate, 22.2 % propionate , 25.8 % butyrate and 17.3 % valerate. This is translated to a mol distribution, shown in equation 3.15. The distribution will vary in real-time reactors.

$$VFA_{avg} = 0.43 * C_2H_3O_2^- + 0.22 * C_3H_5O_2^- + 0.22 * C_4H_7O_2^- + 0.13 * C_5H_9O_2^- = C_{3.05}H_{5.1}O_2^- \quad (3.15)$$

The acidogenesis process is modeled using a theoretical carbon conservation approach, to determine the yield coefficient Y_{VFA}, defined as the moles of volatile fatty acids produced per mole of carbon present in the pig manure. This derivation assumes that carbon is conserved and distributed exclusively between the produced organic acids and the stoichiometric CO_2 byproduct.

The average number of carbon atoms per mole of VFA in the product mixture is calculated to be 3.05, as shown in Equation 3.15.

The carbon content of each VFA (per mole) and the associated CO_2 production are determined based on the metabolic pathways described in Table 2.8. For each mole of VFA produced, a corresponding amount of CO_2 is released according to these reactions. This relationship can be used to estimate the amount of carbon present in the produced CO_2 per unit of VFA formed, as expressed in equation 3.16.

$$C_{CO_2} = Y_{VFA} \times (f_{Ac} \times 1_{[1CO2:1Ac]} + f_{Pr} \times 0_{[0CO2:1Pr]} + f_{But} \times 2_{[2CO2:1But]} + f_{Val} \times 0_{[0CO2:1Val]}) \quad (3.16)$$

With the mole fractions produced, the relation in equation 3.17 is formed.

$$C_{CO_2} = Y_{VFA} \times (0.43 \times 1 + 0.22 \times 2) = Y_{VFA} \times 0.87 \tag{3.17}$$

For 1 Cmol swine manure, the total carbon output should be equal, resulting in the yields, as shown in equation 3.17, 3.19 and 3.20.

$$1 = C_{vfa} + C_{CO_2} = Y_{VFA} \times (3.05 + 0.87) = Y_{VFA} \times 3.92 \tag{3.18}$$

$$Y_{VFA} = \frac{1}{3.92} \approx 0.255 \text{ mol VFA / Cmol VS} \tag{3.19}$$

$$Y_{CO2} = 0.255 * 0.87 = 0.222 \text{ mol CO2 / Cmol VS} \tag{3.20}$$

The production of ammonia is determined by the nitrogen content of the substrate, and with the nitrogen and carbon balances fixed, the elemental mass balance for hydrogen (H), and oxygen (O) are closed.

The total theoretical metabolic reaction of the VFA production of the swine manure is shown in equation 3.21.

$$\begin{aligned}
\mathrm{CH_{2.6}O_{0.75}N_{0.054}} + 0.20\,\mathrm{H_2O} \longrightarrow& \\
& 0.64\,\mathrm{H_2} + 0.22\mathrm{CO_2} + 0.054\,\mathrm{NH_3} + 0.26\,\mathrm{H^+} \\
& + 0.11\,\mathrm{C_2H_3O_2}^- \quad \text{(Acetate)} \\
& + 0.057\,\mathrm{C_3H_5O_2}^- \quad \text{(Propionate)} \\
& + 0.055\,\mathrm{C_4H_7O_2}^- \quad \text{(Butyrate)} \\
& + 0.032\,\mathrm{C_5H_9O_2}^- \quad \text{(Valerate)}
\end{aligned} \tag{3.21}$$

Regarding the reaction in 3.21, a high hydrogen production is observed. This is primarily due to the absence of alternative electron sinks in the stoichiometric representation, while in practice multiple parallel pathways occur that consume reducing equivalents. The actual H_2 production is expected to be significantly lower. This is supported by experimental data from a pilot-scale study on volatile fatty acid (VFA) production from food waste, where a hydrogen concentration of approximately 1 vol% was measured [158]. In an other study, 4.8 mL H_2/g TS is observed on cattle manure digestion, leading to an even smaller H_2 fraction [160].

Based on the value of the pilot-scale study, a value of 1 vol% is assumed for hydrogen production in this study. The rest of the reaction is considered the same, as the electron sinks are still available, but possible products from this are neglected in this study. For example, biomass formation is likely happening during the process, which has been neglected in this reaction. Cao et al. [17] noted that when treating swine manure, operating under strictly anaerobic conditions limits microbial growth, diverting only a very small fraction of 0.9% of the soluble hydrolysate toward biomass generation. Because of this, the biomass production is neglected in this study, but products like this are expected to lead to electron sinks.

Finally, the theoretical values are adjusted to better reflect practical conditions by applying the measured acidification degree. This parameter represents the overall efficiency of the acidogenesis step and was reported by the CSTR study on swine manure of Wang et al. [157] to be 38.3% mg COD VFA/L over mg COD_{in}/L. As such, it reflects the combined efficiency of both the hydrolysis and acidogenesis stages within the reactor. By applying the conversions factors and molecular weights of the swine manure and VFA mixture, a yield of 0.138 mol VFA per mol VS of swine manure is found. Given the theoretical maximum carbon conversion (Y_{VFA}), the efficiency of the reaction can be calculated, as is shown in equation 3.22.

$$Efficiency = \frac{Y_{obs}[mol_{VFA}/mol_{VS}]}{Y^{th}_{VFA}[mol_{VFA}/mol_{VS}]} = \frac{0.138\ mol_{VFA}/mol_{VS}}{0.255mol_{VFA}/mol_{VS}} = 54\% \quad (3.22)$$

This means that of the theoretical possible conversion, 54% is reached with the acidogenesis degree of Wang et al. [157].Of 54% efficiency, the remaining substrates stays un-reacted. For sensitivity analysis, a maximum of 99% is applied, as 100 % conversion is hardly ever observed and 99% the approached maximum. Only CO_2 and H_2 are considered as gas emissions [158].

S/L separation

For the solid-liquid separation, the same decanter was used as in the Biogas production system, to separate the mass and liquid. VFA's are considered to remain 100% in the liquid and NH3 is assumed to behave similar as TAN. The PHA in the biomass before extraction, is assumed to follow the same separation as the total solids.

Filtration should be considered based on the operational requirements of the PHA enrichment and accumulation stages described in the literature review. However, no detailed modelling of this step is included in this study. It is assumed that all large solids are removed and directed to the solid fraction.

Storage

No model implementations are done for the storage of the VFA liquid, only control over the concentrations inflow of the PHA process, by applying only a fraction of the total liquid.

PHA production and accumulation

The PHA production stage is modelled based on a mixed microbial culture (MMC) process consisting of an enrichment phase and a production (accumulation) phase. The influent to this system is the liquid fraction obtained after solid-liquid separation, containing volatile fatty acids (VFAs), nitrogen, and other dissolved components.

By Guho et al. [54], a distribution of the VFA liquid is applied between the enrichment phase and the production phase, which is 27 % to the enrichment and 73% of the VFA's being directed to the production reactor. The same ratio is applied in this study. For the enrichment reactor, water addition is used to obtain a correct range of VFA concentration for the growing of the biomass with the 23% of the total liquid. A OLR of 0.52 g COD VFA L^{-1} d^{-1} with SRT of 4 days is applied by Guho et al. [54], so 2 g COD_{VFA}/L is targeted (Table 2.9, section 2.7.5)

For the production reactor, no additional dilution is applied. Guho et al. [54] reports that pulse feeding is applied, with a maximum VFA concentration of approximately 0.8 g COD VFA L^{-1} (26 mmol C-VFA L^{-1}), which is maintained through controlled substrate addition.

Guho et al. [54] has measured an ammonia concentration of 100 mg $N - NH_3$/ L at the start of the enrichment in the enrichment phase and a range of 50-350 mg $N - NH_3$/L in the production phase. The concentration of ammonia is calculated in the model and should be in a similar range. Additionally, the fraction of free ammonia (NH_3) relative to total ammoniacal nitrogen (TAN) is calculated, as this influences microbial activity and potential inhibition.

Although the ammonia and phosphorus contents of the inflow are consumed in the PHA reactors, no nitrogen or phosphorus uptake is assumed in the model of this study, as the consumptions are assumed to be low. This will lead to a small overestimation of the P and N content of the outflowing liquid.

Given the measured increased pH by Guho et al. [54], it is assumed in the mass balance that OH^- is produced. The produced polymer is assumed to be poly(3-hydroxybutyrate-co-3-hydroxyvalerate) (PHBV), approximated by the empirical formula $C_9H_{14}O_4$[12]. The molecular weight and elemental composition are calculated accordingly.

The PHA synthesis reaction is formulated based on elemental mass balances and degree of reduction (γ) analysis. The average VFA composition is used as input, a PHA yield of 0.76 $gCOD_{PHA}$/$gCOD_{VFA}$ is applied based on the average of the yields of the enrichment and accumulation reactor in Guho et al. [54]. This yield is converted to both mass-based and molar yields using literature values for COD equivalents, being 1.7 gCOD/g PHA [144]. This leads to a yield of 0.186 mol PHA/ mol VFA and the following reaction stoichiometry shown in equation 3.23.

$$-1\,\mathrm{C_{3.1}H_{5.1}O_2}- - \nu_{O2}\,\mathrm{O_2} - \nu_{H2O}\,\mathrm{H_2O} + 0.186\,\mathrm{C_9H_{14}O_4(PHBV)} + \nu_{CO2}\,\mathrm{CO_2} + \nu_{OH-}\mathrm{OH^-} \quad (3.23)$$

The required oxygen demand is calculated by balancing the electron equivalents between substrate and product, using equation 3.24.

$$Y_{O2} = \frac{\nu_{VFA} - \nu_{PHA} * Y_{PHA/VFA}}{-4} \quad [\text{mol } O_2 \text{ mol}^{-1} \text{ VFA}] \quad (3.24)$$

The remaining mass balances are solved using a least-squares approach to obtain coefficients for water, carbon dioxide, and hydroxide, shown in equation 3.25

$$-1\,\mathrm{C_{3.1}H_{5.1}O_2}^- - 0.80\,\mathrm{O_2} - 1.1\,\mathrm{H_2O} + 0.20\,\mathrm{C_{14}H_9O_4(PHBV)} + 0.32\,\mathrm{CO_2} + 1.0\,\mathrm{OH^-} \quad (3.25)$$

The total moles of VFA entering the production reactor are calculated from the VFA mass and molecular weight, taking into account the fraction of 73% directed to production. The VFA is assumed to be completely consumed, given the data from Guho et al. [54]. The remaining VS in the liquid inflow of the PHA step is assumed to be only un-reacted swine manure, and thus not converted to PHA and remain in the liquid. The same is assumed for inorganic residues like ash. The process is assumed to

operate at a temperature of 16–27 °C. The pH in the production reactor is assumed to range between 8.2 and 9.1. 0.232 kg defoamer per kg produced PHA and 0.116 kg H_2SO_4 per kg PHA are assumed to be used, taken from Wu et al. [158], to consider the conditions stable PHA accumulation. As Wu et al. [158] uses H_2SO_4 and NaOH for pH control, next to defoamer (PDMS) for prevention of biomass loss in aerobic fermentors, the same amounts are applied here, 0.058 kg H_2SO_4 / kg PHA and 0.232 kg PDMS / kg PHA. During the PHA production process, oxygen is consumed and carbon dioxide is

produced, as seen in the reaction 3.23. In Del Oso et al. [30], only biogenic CO_2 emissions are reported across all scenarios. Also, Wu et al. [158] only reports biotic carbon dioxide emissions. Therefore, in this study, only CO_2 emissions are considered for the PHA production stage.

As the solid fraction of the outflow of this step is used for composting, the fate of the defoamer and sulfuric acid must be considered, as these substances may ultimately be applied to land. In Appendix A, it is further explained that both chemicals do not effect any downstream process products.

PHA extraction

From the liquid obtained during the PHA production, the PHA needs to be extracted out of the cells. The PHA extraction was done with a centrifugation step, separating the relatively heavy cells containing PHA, from the liquid. After this a sequential digestion and solvent extraction method was applied with sodium hypochlorite and chloroform to break the cells open and extract the PHA, as described in Table 2.11 [158, 110]. Wu et al. [158] has obtained a yield recovery of 85%. The purity is considered the same as measured by Wu et al. [158], being 99%. As reported by Wu et al. [158] 0.969 kg NaOCl/kg PHA and 0.747 kg chloroform/kg PHA is used to obtain these yields.

According to Del Oso et al. [30], no emissions are reported for the PHA extraction process. Similarly, the extraction method described by Wu et al. [158], which is adopted in this process design, does not include any emissions in its inventory data for the extraction and purification stages. Consequently, no emissions are considered for PHA extraction in this study.

Chloroform is recycled in the DSP, with a recycle ratio of 85% [158]. The effects of chloroform and NaOCl are discussed in Appendix A, conluding to no significant effects for the DSP.

The remaining streams are a liquid stream out of the decanter after PHA production, this contains nutrients like nitrogen, phosphorus and volatile solids, which is led into the micro-algae pond. The separation of the cells and the PHA leads to a rest stream of bio-waste, containing the residues of the broken cells.

Composting

For the composting process in the bioplastic production scenario, the same methodology is applied as in the biogas scenario. In this case, however, both the solid fraction obtained after acidogenesis and the solid fraction remaining after PHA extraction are considered as inputs to the composting process. As a result, a higher total mass of material is expected to be directed to composting, which will likely lead to an increased amount of compost .

Micro-algal cultivation

In the bioplastic scenario, the similar method is used for calculations of the amount of algae produced. The composition of the un-reacted pig manure is used as an approximation for the mixed substrate supplied to the cultivation system, given the fact that all VFA's are consumed in the PHA production steps. In reality, sludge is added for the PHA accumulation phase, but remaining particles of this in the liquid fraction is not taken into account. The composition is $C_1H_{2.6}O_{0.75}N_{0.005}$, which is very similar to the mixed substrate in the biogas production, thus the same calculations are done.

3.1.3. Energy demand

The electricity demand is determined with literature for manure management, shown in Table 3.8.

Table 3.8: Electricity demand of operators

Operator	Equation	Source
Anaerobic digester	0.251 kWh/ m^3 swine waste	[59]
Decanter	4 kWh/m3	[32, 132]
Cultivation	0.0238 kWh/day/ m^3	[101]
Composting	5 kWh / ton manure	[132]
Ammonia stripping	2.3 kWh/ m^3	[132]
VFA production	53.5 kWh	[158]
PHA enrichment and production	132.8 / reactor	[158]
PHA extraction	18.74 and 0.86 kWh/kg PHA produced	[158]

The electricity demand of the PHA production is mainly based on Wu et al. [158], as the process considered in that study is similar, differing primarily in the upstream substrate (food waste instead of manure-derived streams). In Wu et al. [158], the electricity demand is reported per kg of PHA produced. Despite the difference in scale between the pilot-scale system described in the literature and the farm-scale system considered in this study, the specific electricity demand (per kg PHA) is assumed to remain constant. The energy demand is assumed constant over different yields for the VFA production and PHA production steps. Although improvements in the yields would result in larger volumes of VFA, the bioreactor size kept constant, thus the electricity demand is considered constant. The storage unit will store the extra VFA liquid before further processing.

Heat demand of bioplastic production is considered to be inside the electricity demand as Wu et al. [158] does not mention this. For the biogas production scenario only heat consumption is needed for the anaerobic digestion[32]. The same source as the electricity demand of the AD , reports a heat demand of 0.77 kWh per cubic meter of manure [59].

3.2. Environmental impact assessment

The environmental impact is studied with a Life Cycle Assessment (LCA). Using LCA standards, first the goal and scope are explained, after which data collection and calculation procedures are explained to identify and quantify all relevant input and output flows, which is called the inventory. In Life cycle impact assessment (LCIA), the inventory results are evaluated and translated into potential environmental impacts. This step is done using the life cycle assessment software fo SimaPro. SimaPro is a widely used LCA tool that enables the modeling of complex product systems by linking foreground processes defined by the user with background data from established life cycle inventory (LCI) databases.[29].

3.2.1. Goal

The objective of the study is to identify main contributors to environmental emissions and compare which treatment scenario is best for swine manure treatment.

3.2.2. Scope

Both systems are studied using a functional unit of 1000 kg (1 ton) of manure. The system boundaries are defined as cradle-to-gate, from raw manure input to final product outputs. In line with LCA practice [45], manure is treated as a waste stream with zero upstream burden, consistent with European Regulation 2023/1185 [37]. System expansion is applied to account for avoided impacts, assuming all products (biogas, bioplastic, compost, and algae) substitute conventional equivalents. For this, the product is substituted, but not the application of product. In the *biogas production* scenario, biogas is partly used in an on-site CHP unit to meet internal heat and electricity demand, after which the surplus biogas can be substituted. Compost and algae substitute conventional fertilizers, while bioplastic replaces polyethylene terephthalate (PET). Transport is excluded due to assumed similarity between both systems. Diesel use for composting is included, covering both production and combustion emissions. In microalgae cultivation, 95% of water is recycled to achieve the required TAN concentration, with 5% supplied as fresh water [101]. Air is used as the oxygen source in PHA production [46]. The system boundaries are shown in Figures 3.3 and 3.4.

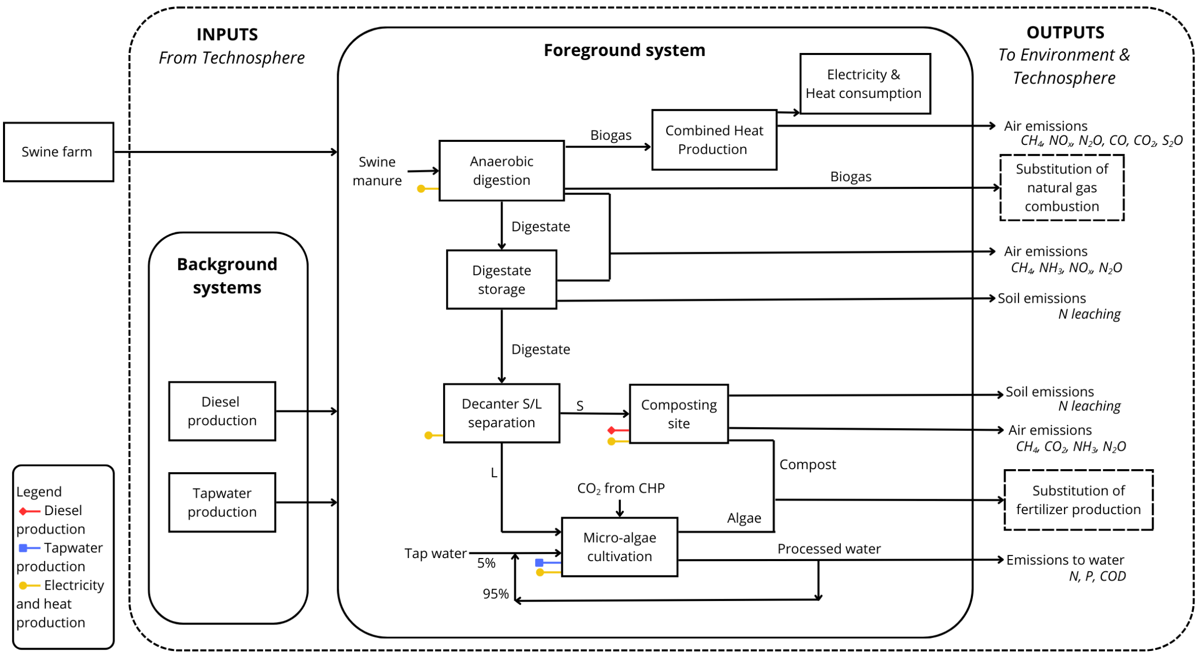


Figure 3.3: System boundaries of biogas production scenario for swine manure treatment. Anaerobic digestion with biogas production and fertilizer production from digestate, by S/L separation and micro-algae production.

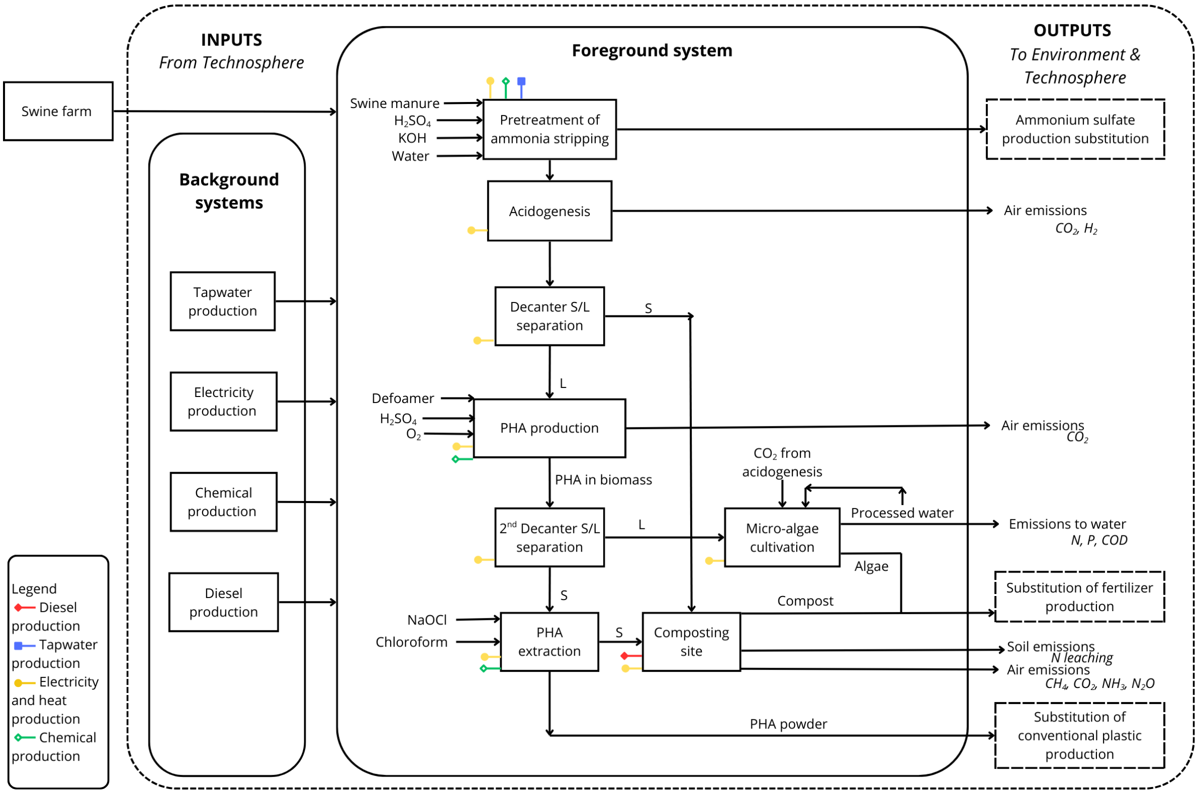


Figure 3.4: System boundaries of bioplastic production scenario for swine manure treatment. Pretreatment of ammonia stripping, acidogenesis to produce VFA's to be converted to bio plastics. Compost and algae production with liquid and solid rest streams.

3.2.3. Inventory Analysis

The life cycle inventory (LCI) is divided into foreground and background systems. The foreground system consists of all processes explicitly modelled in this study, while background processes are implemented in *SimaPro* using datasets from the ecoinvent database representative for Spain. Biogenic CO_2 emissions are considered climate-neutral and are therefore excluded from the impact assessment, but still reported in the inventory for completeness.

Air use is assumed to have no environmental impact, as it is considered an abundant resource and not associated with significant upstream burdens [97] Nitrogen oxides (NO_x) are reported by the IPCC in terms of NO_2 equivalents. Therefore, all NO_x emissions are implemented in SimaPro as NO_2 emissions [79].

All emissions are assigned to the appropriate environmental compartments in SimaPro: air emissions are allocated to the subcompartment "stratosphere+ troposphere" , soil emissions to "agricultural soil", and water emissions to "undefined", as water emissions can effect the freshwater bodies, soil and further emit to the marine waters.

Given the calculations in the model, the Inventory tables of both processes are shown in Table 3.9 and 3.10.

Table 3.9: Inventory table of biogas production process. Description of SimaPro labels. For diesel use diesel production (Diesel Europe without Switzerland | market for diesel | Cut-off, U) and consumption (Energy, from diesel burned in machinery RER Economic, U).*: NO_x emissions determined by IPCC in NO_2 equivalents [79].

Input	Value	Unit	Output	Value	Unit	Description
Anaerobic digestion system						
Swine manure	1000	kg	Digestate	995.9	kg	
Electricity (from biogas)	0.251	kWh	Biogas (66% CH4)	5.83	kg	
			Air emissions			
			CH4 (biogenic)	0.0881	kg	Methane, biogenic
			NH3	0.121	kg	Ammonia, ES
			NOx	0.0091	kg	Nitrogen dioxide, ES*
Digestate storage						
Digestate	995.9	kg	Digestate	995.6	kg	
			Soil emissions			
			N leaching	0.0520	kg	Nitrogen, ES
			Air emissions			
			CH4 (biogenic)	0.00350	kg	Methane, biogenic
			N2O	0.0944	kg	Dinitrogen monoxide
Biogas combined heat production						
Biogas (66% CH4)	3.54	kg	Electricity for own consumption	7.75	kWh	
			Air emissions			
			CH4 (biogenic)	0.000111	kg	Methane, biogenic
			CO	0.00168	kg	Carbon monoxide, biogenic
			CO2	6.22	kg	Carbon dioxide, ES
			SO2	4.20E-05	kg	Sulfur dioxide, ES
			NOx	0.00615	kg	Nitrogen dioxide, ES*
			N2O	0.000117	kg	Dinitrogen monoxide
S/L separation						
Digestate	995.6	kg	Solid fraction	90.04	kg	
Electricity (from biogas)	3.98	kWh	Liquid fraction	903.9	kg	
Composting site						
Solid fraction	90.33	kg	Compost	24.77	kg	
Electricity (from biogas)	0.29	kWh	Soil emissions			
Diesel	0.218	kg	N leaching	0.0195	kg	Nitrogen, ES
Energy from diesel burning	9.57	MJ	Air emissions (from composting)			
			CH4 (biogenic)	0.617	kg	Methane, biogenic
			CO2	8.73	kg	Carbon dioxide, ES
			N2O	0.00686	kg	Dinitrogen monoxide
			NH3	0.0488	kg	Ammonia, ES
Micro-algae cultivation						
Liquid fraction	904.3	kg	Algae	7.17	kg	
CO2 from CHP	13.12	kg	Outflowing water	1111	kg	
Electricity (from biogas)	3.07	kWh	P	0.0032	kg	Phosphate, ES
Tap water	201.3	kg	N	0.0519	kg	Nitrogen
			COD	0.285	kg	Chemical Oxygen Demand, ES

Table 3.10: Inventory table of Bioplastic production process. Description of SimaPro labels. For diesel use diesel production (Diesel Europe without Switzerland | market for diesel | Cut-off, U) and consumption (Energy, from diesel burned in machinery RER Economic, U).*: NO_x emissions determined by IPCC in NO_2 equivalents [79].

Input	Value	Unit	Output	Value	Unit	Description
Pretreatment						
Swine manure	1000	kg	Pretreated swine manure	1123	kg	
Electricity (from biogas)	2.3	kWh	Ammonium sulfate	8.93	kg N	
KOH	11.00	kg				
H2SO4	5.91	kg				
Acidogenesis						
Pretreated swine manure	1122.7	kg	Digestate	1106	kg	
Electricity	53.5	kWh	Air emissions			
			CO2	4.39	kg	Carbon dioxide, ES
			H2	0.0439	kg	Hydrogen
S/L separation						
Digestate	1106	kg	Solid fraction	83.0	kg	
Electricity	4.43	kWh	Liquid fraction	1032	kg	
PHA production						
Liquid fraction	1032	kg	PHA containing liquid	1206	kg	
Electricity	132.8	kWh	Air emissions			
H2SO4	0.348	kg	CO2	1.16	kg	Carbon dioxide, ES
Defoamer (PDMS)	0.693	kg				
O_2	2.15	kg				
Second S/L separation						
PHA containing liquid	1206	kg	PHA containing solid fraction	82.4	kg	
Electricity	4.87	kWh	Liquid fraction	1121	kg	
PHA extraction						
PHA containing solid fraction	82.4	kg	PHA product	2.98	kg	99% pure PHA powder
Electricity	57.1	kWh	Bio-waste	79.5	kg	Biomass without PHA
NaOCl	2.895	kg				
Chloroform	2.232	kg				
Composting site						
Solid fraction	82.4	kg	Compost	36.2	kg	
Bio-waste	79.5	kg	Soil emissions			
Electricity	0.533	kWh	N leaching	0.00123	kg	Nitrogen, ES
Diesel	0.395	kg	Air emissions			
Energy from diesel burning	17.31	MJ	CH4 (biogenic)	0.787	kg	Methane, biogenic
			CO2	11.1	kg	Carbon dioxide, ES
			N2O	0.0043	kg	Dinitrogen monoxide
			NH3	0.020	kg	Ammonia, ES
Micro algae cultivation						
Liquid fraction	1034	kg	Algae	1.24	kg	
CO2 from CHP	2.27	kg	Outflowing water	848.5	kg	
Electricity	0.533	kWh	P	0.00196	kg	Phosphate, ES
			N	0.0479	kg	Nitrogen
			COD	0.191	kg	Chemical Oxygen Demand, ES

3.2.4. Avoided products

In order to consider a substitute for a product, the efficiency of the use of the product is quantified.

Compost substitution

For compost, substitution is based on the equivalent amount of mineral fertilizer that would otherwise be applied. This reduced efficiency is accounted for when determining the amount of fertilizer displaced, as can be seen in equation 3.26 and 3.27.

$$N_{avoided}[kg] = N_{compost}[kg/FU] * Working\ efficiency\ N\ [\%] \tag{3.26}$$

$$P_{avoided}[kg] = P_{compost}[kg/FU] * Working\ efficiency\ P\ [\%] \tag{3.27}$$

The avoided product is modelled using the market activity for "inorganic phosphorus fertilizer, as P_2O_5" in Spain. Market activities represent the average consumption mix within a specific region, including domestic production and imports weighted by their contribution to total supply. Upstream processes include the production of phosphorus fertilizers from mineral sources such as phosphate rock and phosphoric acid, based on national statistics from the International Fertilizer Association.[136] Potassium is not considered as a separate nutrient in this study, as it is typically not applied independently but rather incorporated into mixed fertilizers with nitrogen and phosphorus. In Spain, for example, 47% of potassium fertilizers were applied as NPK mixtures in 2023. [75, 77].

Algae substitution

In this study, a nitrogen substitution efficiency of 50% and a phosphorus substitution efficiency of 30% are assumed for microalgal biomass applied as a fertilizer or soil improver, as explained in section2.6. The same equations as for compost are used.

Biogas substitution

For the biogas substitution, the amount of electricity and heat that could be generated from the remaining biogas is accounted for. The same conversion efficiencies as taken in the CHP unit used for internal consumption, are applied.

Bioplastic substitution

Since PHA powder is produced in this scenario, the avoided product is modeled in SimaPro as polyethylene terephthalate (PET) fines, with a 1 kg PHA : 0.72 kg PET ratioDel Oso et al. [30].

Ammonium sulfate

During the pretreatment step of VFA production in the bioplastic production scenario, ammonia must be removed, resulting in the production of ammonium sulfate as a co-product. This product can be considered conventional ammonium sulfate [132]. Therefore, a 1:1 substitution ratio is assumed. In SimaPro, this is modelled as the avoidance of ammonium sulfate production.

The avoided products of the biogas and bioplastic production process are summarized in Table 3.11.

Table 3.11: Comparison of avoided products for biogas and bioplastic production scenarios.

Item	Biogas scenario	Bioplastic scenario	Description
N fertilizer (kg N)	0.414	0.128	Market for inorganic nitrogen fertilizer, as N ES
- Compost	0.118	0.0774	30% working efficiency of N in compost
- Algae	0.296	0.0512	50 % working efficiency of N in algae
P fertilizer (kg P2O5)	0.383	0.408	Market for inorganic phosphorus fertilizer, as P2O5 ES
- Compost	0.293	0.392	30% working efficiency of P in compost
- Algae	0.383	0.0155	30% working efficiency of P in algae
Biogas (kg)	2.19	-	Biogas output for energy recovery
- Electricity (kWh)	4.21	-	Electricity, low voltage, market for electricity,ES
- Heat (kWh)	4.96	-	Heat, central or small-scale, other than natural gas ES
Conventional plastic (kg)	-	2.19	Polyethylene terephthalate fines RER
Ammonium sulfate (kg)	-	8.93	Market for ammonium sulfate

3.2.5. Life Cycle Impact Assessment

The Life Cycle Impact Assessment (LCIA), translates the quantified inputs and outputs of the system into their corresponding environmental impacts. In this study, midpoint impact categories are assessed using the ReCiPe 2016 Hierarchist V1.07/World (2010), which is implemented in Simapro. Hierarchic perspective is chosen, as it relies on scientific consensus, focusing on well-established impact mechanisms with predictable outcomes.

Impact categories

In literature, a wide range of impact categories is considered for the environmental assessment of swine manure treatment systems [45]. As described in the Introduction, swine manure management is associated with nutrient leaching, greenhouse gas emissions, and negative effects on ecosystems, particularly in regions such as Spain. Therefore, impact categories related to climate change, nutrient emissions (both nitrogen and phosphorus), soil acidification, and ecosystem quality are considered most relevant. Apart from this, the scope of this report needs to be considered. It has been noted that the emission of heavy metals are caused by manure management systems, but given assumption 2 this is left out of the scope. Because of this, including the toxicity impact categories is not reasonable.

Based on this, the selected impact categories are Global Warming Potential (GWP), Terrestrial Acidification (TA), Freshwater Eutrophication (FE), Marine Eutrophication (ME) and Fine Particulate Matter Formation (FPMF). GWP is included to account for greenhouse gas emissions such as CO_2, CH_4, and N_2O in both treatment methods. TA is considered due to emissions of acidifying compounds such as NH_3 and NO_x. FE and ME are included due to nitrogen and phosphorus emissions which lead to eutrophication in aquatic ecosystems. FPMF is included to capture the formation of fine particulate matter from precursor emissions such as NH_3 and NO_x.

Database

The life cycle assessment (LCA) was performed using the software package SimaPro 10.3.0.1. Cut-off definitions are used for background processes. The cut-off approach severs the environmental history of a material at the point it is generated, meaning is that the primary producer of a material bears all environmental burdens. For chemical use in a process, the production of the chemical is accounted for in its full impact, as this production process is build for this alone. If a material is a waste produced during a process, no environmental burden is applied [10, 136].

The background data used in this study is derived from the database ecoinvent, which contains standardized data on energy production, material processes, transport, and emissions. This data is compiled from a combination of measured industrial data, literature sources, and modeled processes, and is continuously updated to reflect technological and geographical differences [10].

Foreground data, representing the specific system under study are shown in the Inventory Tables. The combination of foreground and background data allows for a comprehensive assessment of environmental impacts across the full life cycle.

Characterization factors

The translation of emissions and resource use into environmental impacts is performed through life cycle impact assessment (LCIA) methods implemented in SimaPro. Characterization factors are used to convert individual emissions (e.g., CH_4, NH_3, NO_x) into a common unit within an impact category (e.g., kg CO_2-eq for global warming or kg SO_2-eq for acidification).

These characterization factors are defined by internationally recognized LCIA methods and are based on scientific models that describe the environmental mechanisms linking emissions to impacts. Although Simapro implements these factors automatically, the values are also visualized in Table 3.12 for a better understanding of the outcome values of the LCA.

Table 3.12: Characterization factors ReCiPe 2016 Midpoint Hierarchist for impact categories

Molecule / Emission	GWP (kg CO_2-eq)	TA (kg SO_2-eq)	ME (kg N-eq)	FE (kg P-eq)	FPMF (kg $PM_{2.5}$-eq)
CO_2	1.0	–	–	–	–
CH_4	34.0	–	–	–	–
N_2O	298.0	–	–	–	–
NH_3 (Air)	–	1.96	0.10^s / 0.24^w		0.11
NO_x (Air)	–	0.36	0.04^s / 0.09^w		0.24
SO_2 (Air)	–	1.00	–	–	0.29
$PM_{2.5}$ (Air)	–	–	–	–	1.00
Total N (Water/Soil)	–	–	0.13^s / 0.30^w		–
Total P (Water/Soil)	–	–		0.10^s / 1.00^w	–

^{s} Indirect emission from agricultural soil (leaching/runoff); ^{w} Direct emission to freshwater/marine bodies.
Note: GWP= Global Warming Potential; TA = Terrestrial Acidification; FE= Freshwater Eutrophication; ME = Marine Eutrophication; PM = Fine Particulate Matter Formation.

3.3. Techno-economic Analysis

A techno-economic analysis (TEA) is performed to identify parameters that influence the feasibility of the manure treatment plants. By using the mass and energy balance of the processes under comparison, the reactor size, raw material costs, revenue, and electricity or heat costs can be estimated. These calculations lead to the determination of both capital investment and operational costs, which are then used to estimate the net present values of both plants[59]

Assumptions

The plant is assumed to operate for 7,920 hours per year, labor hours per scenario differ, which is discussed later in this section. Location factors used in the cost estimation are provided in Appendix D, and CEPCI values were used to calculate the cost of in year 2025 [126]. A currency conversion rate of 1 USD = 0.86 EUR was assumed [53].

3.3.1. Capital costs

The capital expenditure (CAPEX) of the process is estimated based on the Direct Fixed Capital (DFC), which is derived from the total purchased equipment cost (PEC) . The PEC is determined by estimating the cost of each individual unit operation and summing these contributions over the entire process. The purchase cost of each equipment item is obtained from literature data and subsequently adjusted to the required process conditions using scaling relations. Three types of scaling are applied: capacity scaling, location scaling, and cost index scaling.

Capacity scaling is performed using the six-tenths rule, as is stated in equation 3.28.

$$C_{\text{reactor}} = C_{\text{reactor,ref}} \left(\frac{V_{\text{reactor}}}{V_{\text{ref}}} \right)^{0.6} \tag{3.28}$$

In this, C represents the equipment cost and V the reactor volume. If the capacity of a reactor is stated instead of the volume, the same relation applies.

Location scaling accounts for differences in installation costs between regions, for which equation 3.29 can be used.

$$C_{\text{reactor}} = C_{\text{reactor,ref}} \left(\frac{F_{\text{ISF}}}{F_{\text{ISF,ref}}} \right) \tag{3.29}$$

In equation 3.29, F_{ISF} is the installation scaling factor defined per region or country.

Cost index scaling is applied to correct for inflation over time using the Chemical Engineering Plant Cost Index (CEPCI). The CEPCI is an index used to adjust historical equipment and plant costs to present-day values by accounting for inflation in materials, labor, and construction within the chemical engineering industry. The adjusting of inflation is stated in equation 3.30.

$$C_{\text{reactor}} = C_{\text{reactor,ref}} \left(\frac{\text{CEPCI}_{2025}}{\text{CEPCI}_{\text{ref}}} \right) \tag{3.30}$$

The total purchased equipment cost (PEC) are calculated with equation 3.31. A factor of 10% for unlisted purchase cost are assumed, to account for additional auxiliary components like small dosing pumps, holding tanks for raw materials and products [141].

$$PEC = 1.1 \cdot \sum PC \tag{3.31}$$

After determining the total purchased equipment cost, the Direct Fixed Capital (DFC) is estimated using the Lang factor method, as is stated in equation 3.33 [120].

$$DFC = L_a \cdot \sum PEC \tag{3.32}$$

The Lang factor (L_a) accounts for additional direct and indirect costs required to construct and commission the plant. These include piping, instrumentation, insulation, electrical systems, buildings, yard improvements, auxiliary facilities, installation costs (listed and unlisted equipment), as well as engineering, construction, contractor fees, and contingency.

Typical Lang factors range between 3.5 and 5.03 depending on the complexity of the plant and the extent of new infrastructure required[120].

This lower value can be justified when a relatively small scale of system is applied and the assumption that the plant is integrated into an existing farm infrastructure, reducing the need for extensive additional facilities. Given the farm-scale plant size applied in this study, the biogas production plant could be considered with a lang factor of 3.5. For the bioplastic production plant, given the amount of operators needed, the same assumption cannot be done. A Lang factor of 4.09 is used for a biochemical process that is based on an initial biosynthesis stage followed by downstream extraction and purification, similar to the PHA production process in this study [121]. Even though the process differs in product and specific design, the comparable number of unit operations and the similar overall process structure can justify use of this Lang factor for this study.

The total investment cost are estimated with the direct fixed cost and a working capital of 75% of the listed purchase cost [141, 120].

$$Total\ investment\ cost = DFC + 0.75 * PEC \tag{3.33}$$

To compare obtained CAPEX values to literature, the same relation as in equation 3.28 can be used.

Operator definitions

For the plant, continuous operations are assumed, and with the flow and retention time in the reactor, the volume of the reactor that is needed is described. Over this volume, a safety factor of 1.2 is taken [30, 141].

$$V_{reactor} = Q * HRT * safety\ factor \tag{3.34}$$

Anaerobic digester For the cost estimation of the anaerobic digester, the "Biomodule", a similar AD reactor in Brazil 2022 of 700 m^3, can be scaled to the needed volume of the reactor in this study. The volume is estimated with equation 3.34, with an inflow of the average swine manure treated per year, determined per day, given the equation 3.35, the operating hours (t_{op}) and the density of the manure (ρ)

$$Q = \frac{m_{treated}}{t_{op} * \rho} \tag{3.35}$$

Digestate storage Santonja et al. [132] describes the cost in Germany considered for different covers of slurry tanks, for different dimensions. Given the assumption of an uncovered digestate storage, the investment cost is stated to be not relevant. Thus, for this model, no cost for the storage are considered.

Combined Heat and Power generation (CHP) For the estimation of the price of the CHP unit, the capacity needed first needs to be determined. The annual raw energy is determined by the mass of biogas and its lower heating value:

$$E_{raw} = M_{biogas} \cdot LHV \tag{3.36}$$

The electrical capacity is calculated based on the raw energy, electrical efficiency (η_2), and annual operating hours in equation 3.37.

$$P_e[kWe] = \frac{E_{raw} \cdot \eta_e}{t_{op}} \tag{3.37}$$

The cost of a CHP unit is defined for a 100 kWe by [149]. By scaling this capacity, next to location and year, the cost of the CHP unit can be determined.

Decanter For the liquid-solid separation, a decanter is preferred for manure separation processes. The capital cost of a decanter is determined by the capacity, the inflow of volume separated per hour. For a decanter of 1.5- 2 m^3/h, the price is determined by [132] in Europe. This can be scaled to the required flow in this model.

Composting site For the composting site, the operational equipment is based of a tractor used for infrequently turning to assist with aeration, as the process is defined by passive windrow. For the estimation of the cost, two references are used [59, 132]. Given the equations of scale, location and time, the cost of the composting site is determined per source, the average is be taken of the two to determine the price more specific.

Micro-algae cultivation pond According to the Spanish source of micro-algal cultivation on swine manure, micro-algal productivity typically ranges between 10 and 30 g biomass m^{-2} d^{-1}. [116]

Based on the calculated biomass production of the model in kg algae per day, the required pond surface area can be determined as:

$$\text{Raceway area } [\text{m}^2] = \frac{\text{Algae produced [kg/d]}}{\text{Productivity [kg m}^{-2}\text{d}^{-1}]} * fs \tag{3.38}$$

A average of of 20 g biomass m^{-2} d^{-1} is assumed in this model, an average pond depth of 0.3 m [116]. The volume can be calculated with equation.

$$\text{Raceway volume } [\text{m}^3] = Raceway\ area * 0.3 \tag{3.39}$$

The price of the raceway can be determined with the price of the 320 m^3 raceway of Palomar et al. [116] and scaled to the wanted volume.

Ammonia stripping column For the ammonia stripping column, the costs are estimated by Santonja et al. [132] for manure treatment methods. The price is described for a ammonia stripping column of 15 m^3/h in Slovenia, 2010. The flow rate of this study is approximated by the average manure treated per day.

Acidogenesis reactor For the acidogenesis reactor, a sequencing batch reactor was used by Guho et al. [54]. By Krömer et al. [88], a relation for stirred tank reactors is reported [141]. The relation is described in equation 3.40.

$$Purchase\ cost[USD] = 16,651 * (V_{fermentor})^{0.6} \tag{3.40}$$

The volume was estimated with the flow into the acidogenesis, including the added KOH and water, per day.

VFA liquid storage tank For a storage tank to store the VFA containing liquid up to 5 days, a price of 80 USD/m^3 is applied from Mudliar et al. [110], which is also scaled to location and year.

PHA production The relation of equation 3.40 is used for the enrichment and accumulation reactor, the inflow of the enrichment reactor is based on a 27 % continuous outflow of the storage tank and a HRT of 4 days. The remaining VFA liquid is applied in the accumulation reactor (73% of the VFA liquid and the outflow of the enrichment reactor), but the cycle time is estimated around 8.5 hours, obtained from Guho et al. [54]. The amount of micro-organisms added for enrichment are not considered to influence the size of the reactor. The size of the reactors can be compared to the values from [83], which treats sludge from waste water to PHA, with a fermentor of 125 m^3 and a flow of 30 m^3/d for the enrichment and production reactor.

PHA extraction The study of Mudliar et al. [110] describes a similar process and same PHA extraction, with chemical digestion of the biomass using sodium hypochlorite and solvent extraction of PHA using chloroform. The plantt consists of fermentation capacity of 100 m^3/day using a fermentor volume of 125 m^3. The chloroform is subsequently recovered with an efficiency of 90% .This reference case is used as the basis for estimating the capital costs of the extraction section in this study.[110]

The extraction process includes the following unit operations: storage of NaOCl, mixing of NaOCl with the biomass slurry, storage of chloroform, a boiling tank for chloroform extraction with biomass, filtration, storage of filtrate, a grinding system, a chloroform recovery system, and storage tanks for both recovered chloroform and the final PHA product. The total capital cost for this extraction section is reported as €3,324 for a plant capacity of 100 m^3/day.

In the extraction process, centrifugation is applied [110]. The cost of the centrifuge is estimated analogously to a decanter and is added to the total cost of the PHA extraction section. All equipment costs are subsequently scaled to the required process capacity, location, and reference year using the scaling relations.

3.3.2. OPEX definitions

For the operational costs, the assumptions that are made to estimate the costs are shown in Table 3.13 The costs for raw materials, waste management and labor are discussed separately. The electricity demand is evaluated in the model based on assumptions in Table 3.8.

Activity	Assumption	Source
Maintenance	10% of listed PEC	[141]
Insurance	1 % of DFC	[141]
Local taxes	2% of DFC	[141]
Factory expense	5% of DFC	[141]
Electricity demand	0.15 EUR/ kWh	[50]
Laboratory (quality) control	15% of Labor costs	[122], [141]

Table 3.13: Assumptions OPEX

Raw materials

In line with Sánchez-Martín et al. [131], manure is treated as a waste stream and therefore assigned no purchase cost. It is assumed to be freely available, although its intrinsic nutrient value and potential future market developments should be acknowledged. Tap water is priced at 1.24 EUR/m^3, based on data for Castilla y León, Spain [73]. Prices for sulfuric acid, potassium hydroxide, sodium hypochlorite, chloroform, and defoamer are derived from European market analyses for 2025 [71, 69, 70, 67, 18].

Products

The prices for the products need to be defined in order to determine how much revenue is created per process.

Compost The compost value in Spain is in 2026 around 0,10 EUR/ kg [23] . Based on the nutrients in the compost, per N, P, and K a price can be attributed. In December 2025 the price per nutrient was EUR/ kg nutrient 0,40 EUR/kg N, 0.61 EUR/kg P, 0.36 EUR/kg K. [39], which leads to higher total price of the compost. For now, the selling price of compost as complete product is used for the Techno-economic evaluation.

Micro-algae as fertilizer The micro-algae are sold as soil conditioner, as the algae are only separated from the pond water and centrifuged, it cannot be sold as food-grade powder. Given the characterization of algae as high-value organic slow-release fertilizer, this can be sold for a price of 11 EUR/kg N.[24]. The algae biomass is typically sold as an agricultural feed additive or for aquaculture nutrition, as it is not processed to food-grade. The algae can be sold as nutrient for animals, which price is estimated between 2.83 and 315 USD/ kg in 2025. [108, 1]

Biogas sold The biogas that is left after our consumption is sold as electricity and heat with 0.15 and 0.16 EUR/kWh, the same as pricing assumed for electricity bought from the net.[50]

CO_2 avoided emissions as revenue in Spain As Sánchez-Martín et al. [131] pointed out, the avoided carbon dioxide can be seen as a revenue in Spain. Normally, fossil fuel would have to be burned, but with biogas only biogenic CO_2 is emitted.In order to calculate the tons of CO_2 avoided by replacement of fossil fuels, equation 3.41 is applied. The calculation is based on the emission factor for natural gas, which is 0.182 kg CO2/kWh [131], and the energy value of the biogas obtained.

$$CO_2 avoided[kg/yr] = Emission\ factor\ [kgCO_2/kWh] * Net\ biogas\ production\ [kWh/yr] \tag{3.41}$$

To calculate the revenue from this avoided CO_2, the average value of the cost of CO_2 in 2022 is used, which is 84.79 EUR/tCO2 [131], shown in equation 3.42.

$$CO_2 revenue[EUR/yr] = CO_2 avoided\ [kgCO_2/yr] * CO_2\ cost[EUR/kg] \tag{3.42}$$

PHA powder The market price of PHA varies significantly depending on production scale, product grade, and application. As explained earlier, the PHA powder produced in this study can be applied for packaging. A European market analysis by IMARC Group [68] estimates a potential selling price of 2.65 EUR/kg for PHA powder.Montoya [106] states that PHA prices typically range from 5 to 9 USD/kg in 2024. The variation in price is further illustrated by Helian Polymers B.V. [56], which reports a selling price of 12.50 EUR/kg for large quantities in the Netherlands.

The price for commodity fossil-based plastics such as PE and PP are in comparison between 1.10–1.40 USD/kg [106]. PLA, currently the most widely adopted biodegradable bio-plastic with an annual production of approximately 3.2 million tonnes in 2024, is priced at around 2.40–3.00 USD/kg. Thus the price of 2.65 EUR/kg can be competitive to market values.[91].

Ammonium sulfate Ammonium sulfate is a product commonly used as a fertilizer, and market analyses in Europe in 2025 indicate a selling price of approximately 0.23 EUR/kg. [66] Price fluctuations in the European region are primarily driven by seasonal autumn and spring agricultural applications, logistical constraints (such as river water levels and port congestion in Rotterdam or Hamburg), and the cost of feedstocks like ammonia and sulfur, but the 23 cents per kilo is a good estimation. [33].

Waste management
The water is reduced of its nutrients in the micro algae pond. However, not all nutrients are removed from the water, which means that contaminated water is leaving the system. Per contaminant, a price needs to be paid, which is specific per country. Generalitat de Catalunya [48] estimates 0.7610 EUR/kg for nitrogen and 1.5222 EUR/kg for P. The COD contamination is described as MO, and for this a calculation is used of

$$MO = \frac{2 \cdot BOD_5 + COD}{3}$$

which is priced at 1.0023 EUR/kg. 50% of the COD is assumed to be BOD_5.

Labor
According to Girón-Rojas et al. [50], the labor requirement for an anaerobic digestion plant in Spain is estimated at 1.3–2 hours per day. In this study, composting is not included; however, given the passive windrow design of this step, the required labor is assumed to be minimal. Therefore, the upper limit reported by Girón-Rojas et al. [50] is used, resulting in 730 hours per year.

For the bioplastic production scenario, the labor requirement is expected to be higher due to the larger number of unit operations. Khan et al. [83] report a personnel factor of 3.22 for the total process. In that study, personnel factors are defined per unit operation, with most operations comparable to those considered here. By multiplying this factor by the annual operating hours, a total of 25,502 labor hours per year is obtained.

The labor cost in this sector in Spain in 2024 is estimated at €50.24 per hour. This value is assumed to be the same for both scenarios. [50]

3.3.3. Key parameter indications

The depreciation is the amount of money per year that is lost because the value of the machinery in the plant is decreased. This is caused by the use of the equipment. Depreciation is an accounting method that spreads the cost of a tangible asset (like machinery, vehicles, or buildings) over its useful life, reflecting its gradual loss of value due to wear, tear, or obsolescence. It can be calculated with the amount of years the capital can be used (n_{yr}) and the interest rat (ir), with equation 3.43.

$$D = DFC * \frac{ir * (1 + ir)^{n}_{yr}}{(1 + ir)^{n_{yr}} - 1} \tag{3.43}$$

The depreciation also indicates the cashflow needed per year to buy the capital costs.

The actual cash flow is calculated with equation 3.44.

$$CF = Revenue - OPEX - Depreciaiton \tag{3.44}$$

Over the actual cash flow, the gross profit, taxes need to be paid, which leads to the Net profit (eq. 3.45. Tax of 25% is assumed in this study [141].

$$Net\ profit = CF - CF * 0.25 \tag{3.45}$$

The net present value is to be calculated with equation 3.46.

$$\text{NPV} = \sum \frac{CF_n}{(1 + ir)^n} - DFC \tag{3.46}$$

Interest rate
An interest rate of 3.24% is assumed for both plants, corresponding to the business interest rate in Spain in December 2025 [146]. Comparable values are reported in literature, including an interest rate of 4.0% for a similar anaerobic digestion plant Hollas et al. [59], and 3.8% for an anaerobic digestion system with integrated PHB production and a microalgae pond Guillen et al. [55]. However, due to

differences in geographical location and economic conditions, the Spain-specific interest rate of 3.24% is applied in this study.

4 Results

4.1. Biogas production

The total mass flows in the biogas production process for 1 ton of manure is shown in Figure 4.1.

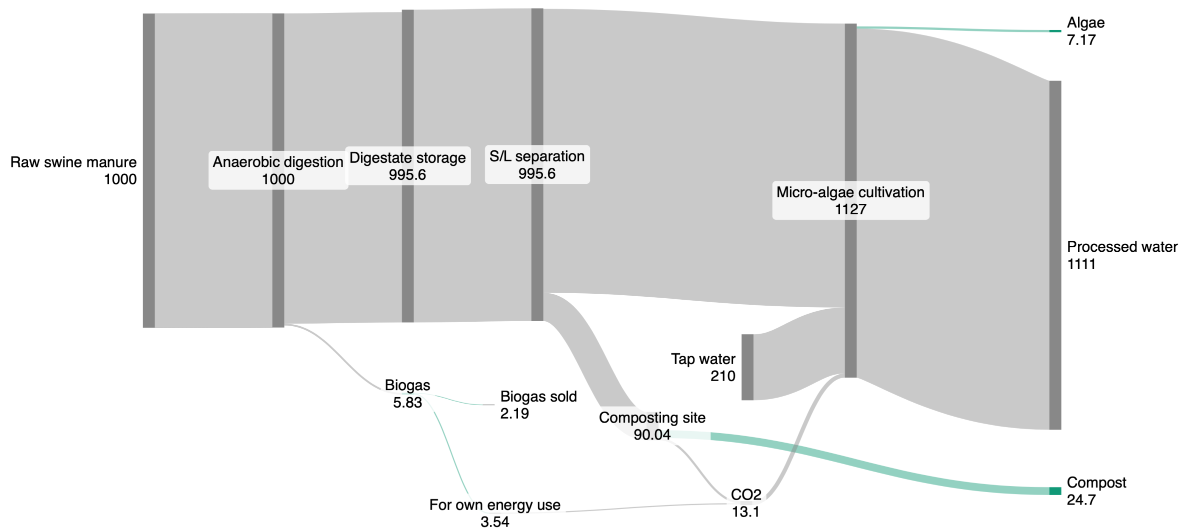


Figure 4.1: Mass balance total process biogas production in kg with input of 1000 kg swine manure (without emissions visualized). Imbalance: 7.4% (air intake during composting not captured in model).

As is shown in Figure 4.1, given an input biodegradability of the manure of 30%, the amount of biogas produced is 5.8 kg, of which 2.2 kg can be sold. This means that the sellable biogas corresponds to 0.54% of the total input mass and 8.9% of the total solids (TS).The other products are 25 kg compost and 7.2 kg algae. Of the 1000 kg manure with TS of 24.7 kg, a total amount of 37 kg product is made. The compost is however only 37% DM, which explains the larger mass of the main products. The total efficiency of the process is 3.7 % of the inflow raw manure ends up in a useful product. The compost was in right nutrient composition to be described as fertilizer according EU legislation. The water leaving the system contains 0.0003 % P, 0.0046 % N and 0.025 % COD, regarding regulations in Spain, management of this water is still needed. The economical cost and environmental cost are discussed in the TEA and LCA.

The in and outflow per operator are evaluated on accuracy, which led to the largest error being around 7.4% in the composting process. This is explained by the consumption of air that is not incorporated in the model. All other processes have a inaccuracy smaller than 0.5 %, indicating almost perfect consistency of the model.

The resulting biogas composition is 63% CH_4 and 37% CO_2. The LHV of 6.3 kWh per m^3 biogas is found. From the produced biogas, the majority is utilized for on-site energy generation for the total electricity demand of 7.7 kWh per ton manure. The operator with the largest electricity demand was the decanter, with 51% of total (Appendix B). For the total electricity demand, 65% of the produced biogas is used for own energy. The remaining 35% is available for external electricity generation or can be exported. The heat demand was estimated to be 0.77 kWh per ton manure. With the required electricity demand, 9.2 kWh heat is produced. The enthalpy effects (heat generation) from microbial metabolic activity during anaerobic digestion are very small, so is neglected in this study [84]. The CO_2 produced during CHP combustion is directed to the microalgae pond, where it is utilized for algal growth, combined with the carbon dioxide from the composting, as can be seen in Figure 4.1.

The total mass in and outflow is shown in Table 4.1.

Table 4.1: Mass balance of inputs and outputs of biogas production scenario

Input	Value	Unit	Output	Value	Unit
Swine manure	1000	kg	Biogas (63% CH_4)	2.2	kg
Diesel	0.22	kg	Algae	7.17	kg
Tap water	210	kg	Compost	24.7	kg
			- 37% DM		
			- 1.6% N		
			- 1.7% P		
			- 1.4% K		
			-12% C		

Microalgae cultivation

The separation performed by the decanter centrifuge yields results consistent with expectations. Phosphorus is predominantly retained in the solid fraction and directed to the composting process, while the majority of the ammonia remains in the liquid fraction. To prevent ammonia inhibition in the microalgae cultivation stage, dilution of the liquid fraction is required. A minimum of 4.2 m^3 of water is needed to reduce ammonia concentrations to acceptable levels. Based on the mass balance, approximately 4.2 m^3 of water must be added per functional unit to achieve the target TAN concentration. For this, a operation of a larger cultivation pond to maintain lower nutrient concentrations is used.

The nutrient composition of the liquid stream entering the microalgae system is evaluated at 1.8 g/L total nitrogen (TN) and 0.19 g/L phosphorus (P), corresponding to an N:P ratio of approximately 9.2:1. Comparison with the optimal ratio for micro-algae growth (4.5:1) indicates that nitrogen is present in excess, while phosphorus is the limiting nutrient.

The total phosphorus available in the liquid stream is 0.168 kg P per ton manure. Assuming a phosphorus removal efficiency of 91%, this results in a phosphorus uptake of 0.153 kg P/t. Given a phosphorus content of 1.82% in algal biomass, the theoretical biomass production is estimated at 7.17 kg algae per ton manure treated.

4.2. Bioplastic production

The bioplastic production scenario is evaluated on mass flows in Figure 4.2.

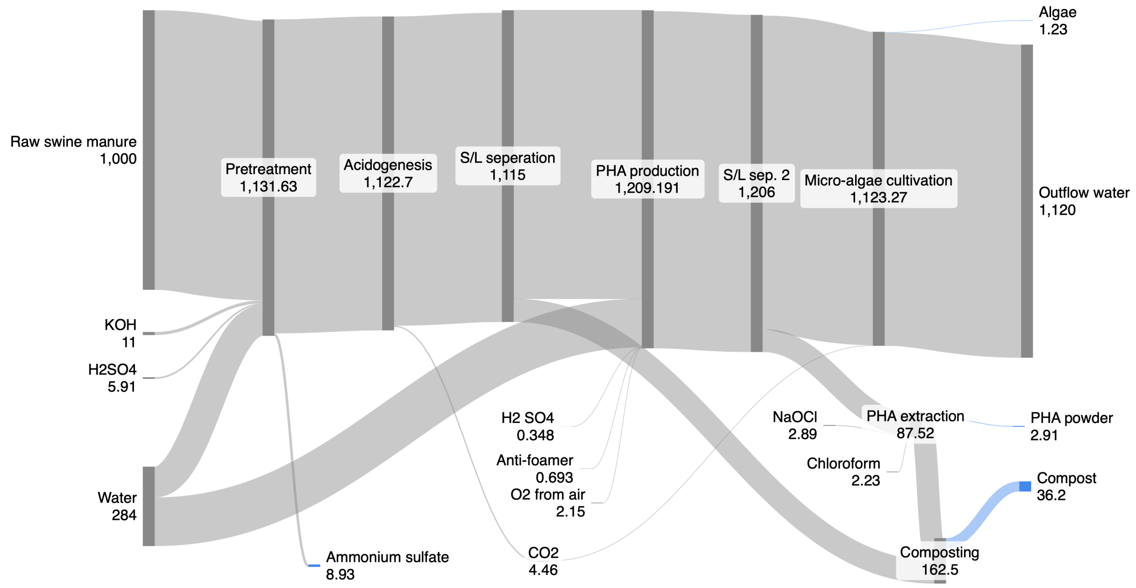


Figure 4.2: Mass balance total process bioplastic production in kg with input of 1000 kg swine manure (without emissions visualized). Imbalance: 3.0% (air intake during composting not captured in model)

As shown in Figure 4.2, the bioplastic production amounts to 2.9 kg. The co-products include 8.9 kg ammonium sulfate, 36 kg compost, and 1.2 kg algae. From 1000 kg of manure, containing 24.7 kg total solids (TS), a total of 49 kg of products is generated. The compost has a relatively low dry matter (DM) content of 24%, based on the calculations. The yield of the main product (bioplastic) is relatively low, corresponding to 0.3% of the incoming manure and 12% of the total solids. When considering all products, 3.6% of the manure is converted into useful outputs, assuming that no added chemicals contribute to the final product mass.

The biomass of the sludge used in the enrichment reactor is not included in this mass balance, which may lead to a slight underestimation of the DM in the compost. The compost meets the required nutrient composition to be classified as a fertilizer according to EU legislation. The liquid fraction contains 0.00023% phosphorus (P), 0.0056% nitrogen (N), and 0.022% chemical oxygen demand (COD), for which environmental and economic impacts are accounted for in the LCA and TEA.

The largest imbalance over the separate operators was 3.1%, also in the composting step caused by unaccounted aeration. The next largest imbalance is 1% in the acidogenesis step.

In the VFA production, per ton manure, 8.4 kg VFA is produced (12.8 kg COD VFA) is produced. With the used 22.8 kg VS, the acidification degree is perfectly applied, as the acidification degree was 0.38 kg VFA/ kg VS. For the gas fraction, 75 vol% H_2 and 25 vol% CO_2 was observed, before setting this to 1% H_2 leading to a biogas production of 2.3 kg. This adjustments has led to a electron imbalance of 1.74 e^- per reaction, 35 % of total electrons in one Cmol-manure.

For the PHA production, 6.7 kg COD PHA was produced in the liquid, in line with the yield of 0.76 g COD PHA/ g COD VFA, as 73% of the liquid was used in the accumulation reactor. This leads to a total yield of approximately 52% over the VFA produced with the enrichment phase included.

A total yield for the acidogenesis and PHA production of 0.18 g COD PHA/g COD PM is found before PHA extraction. After extraction, an overall yield of 0.15 g PHA/g COD is observed.

The bioplastic production scenario is sensitive to high ammonia concentrations, thus ammonia concentrations through important operators are stated in Table 4.2.

Table 4.2: Ammonia nitrogen concentrations before acidogenesis, PHA production and micro-algae cultivation

Inflow	Acidogenesis	PHA production	Micro-algae cultivation
TAN [g/L]	0.28	0.58	0.49
FAN [g/L]	0.089	0.39	0.33

Compared to the constraints discussed in the Literature review and Table A.2 (Appendix A), all ammonia concentration are in the correct range, except for the cultivation pond. This leads to 0.79 m^3 water that is added per ton manure, to prevent ammonia inhibition. The nutrient concentrations of the inflow of the pond were 0.35 g/L TN and 0.026 g/L P, corresponding to an N:P ratio of approximately 14:1. Comparing the required ratio (4.5:1) with the available ratio (N:P 14:1), shows that there is a excess of N, so the phosphorus is still the limiting nutrient [55].

The electricity consumption per operator is shown in Appendix B, with a total of 262 kWh. The largest fraction of the electricity is used by the PHA productions steps, being 51 %.

Additions of chemicals and fuel are not considered mass flows of the model, but are shown in Table 4.3.

Table 4.3: Mass balance of inputs and outputs bioplastic production scenario

Input	Value	Unit	Output	Value	Unit
Swine manure	1000	kg	PHA product	2.91	kg
Electricity	262	kWh	Ammonium sulfate	8.93	kg N
KOH	11.0	kg	Algae	1.23	kg
H_2SO_4	6.29	kg	Compost	36.2	kg
Tap water	14.8	kg	-24% DM		
Defoamer (PDMS)	0.693	kg	-0.71% N		
NaOCl	2.82	kg	-1.6% P		
Chloroform	2.17	kg	-4.2% K		
Diesel	0.395	kg	-10% C		

4.3. Sensitivity analysis

A sensitivity analysis is performed to evaluate the effect of improved process yields in the anaerobic digester and acidogenesis step. Yield ranges of 30-70% biodegradability for AD and 54-99% efficiency of acidogenesis are considered as stated in the method.

The resulting product outputs across these yield ranges are shown in Figure 4.3.

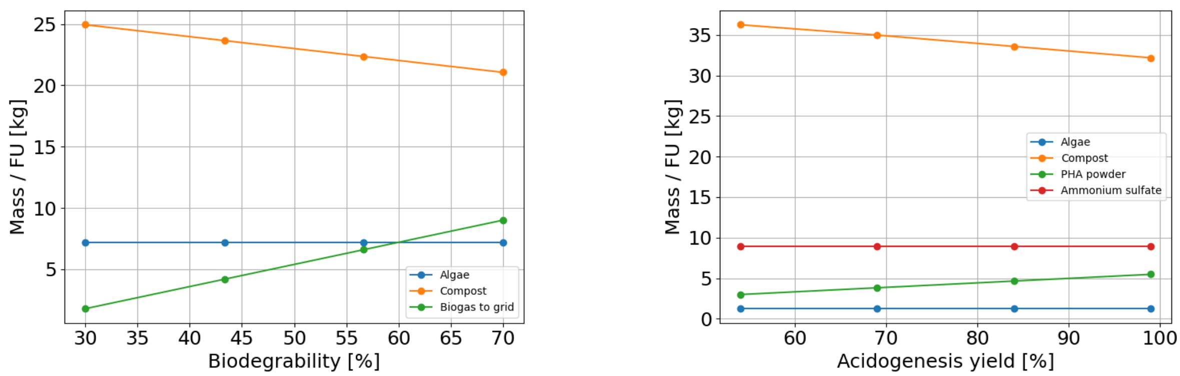


Figure 4.3: Products over range of yield of AD and acidogenesis form 1000 kg manure. Range for anaerobic digestion 30%-70%, for acidogenesis 50-99% C conversion

In Figure 4.3 it is shown that the biogas production increases over the improved biodegradability. The algae production remains constant across both ranges. This is expected, as algae growth is determined by the available phosphorus content, which is not affected by changes in methane yield or acidogenesis in the model. The amount of compost decreases with increasing biodegradability, as more volatile solids are converted into biogas, resulting in less digestate available for composting.

For the bio-plastic production scenario, similar trends are observed for algae and compost, next tot the increasing PHA production. The ammonium sulfate production also remains constant, which is expected as this step occurs upstream of the acidogenesis. The PHA powder production increases with acidogenesis efficiency, which is in line with expectations, as more substrate for PHA synthesis is provided.

For higher yields, a slight linear decrease in electricity demand is observed for biogas production, shown in Appendix B. This is because higher biogas yields result in lower amounts of digestate, thereby reducing the volume that needs to be handled. However, this effect is minimal, amounting to approximately 0.05 kWh per ton of manure.

In contrast, the bioplastic production scenario shows the opposite trend. With increasing acidogenesis yields, the electricity demand rises, as more PHA is produced and subsequently requires extraction. This leads to higher energy consumption during the downstream processing stage, with the energy demand rising to 313 kWh/ton manure.

4.4. Nutrient recovery

To analyze the total process on the nutrient recovery, per carbon, nitrogen, phosphorus and potassium, the flow thought the total process is analyzed. The mass flows are defined per kg element. The useful products are considered to be methane in biogas, compost, algae product and PHA powder.

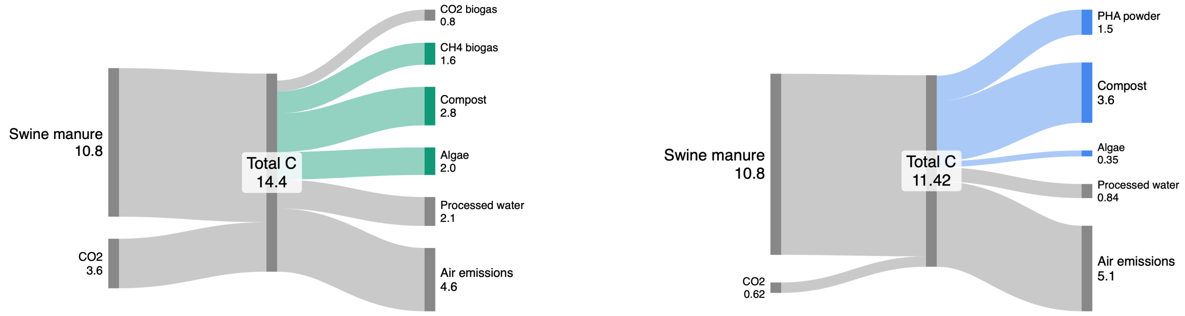


Figure 4.4: Carbon mass balance of total production process Biogas (left) and bioplastic (right) in kg C (input 1000 kg raw manure). Imbalance 0.1 kg C 0.7% and 0.03 kg C (0.3%). Commercially viable products are colored in.

The carbon distribution analysis in Figure 4.4 shows that 51% of the carbon ends up in useful products for the biogas production, while 55% is recovery is seen in the bio-plastic production. This means that in regard to carbon, more of the element ends up in a useful product in the bio-plastic production treatment of swine manure, compared to biogas production. The imbalances are both around below 0.7%.

When maximum achievable yields are applied, the carbon recovery increases to 59% for biogas production and 60% for bioplastic production. This corresponds to improvements of 5% and 9%, respectively.

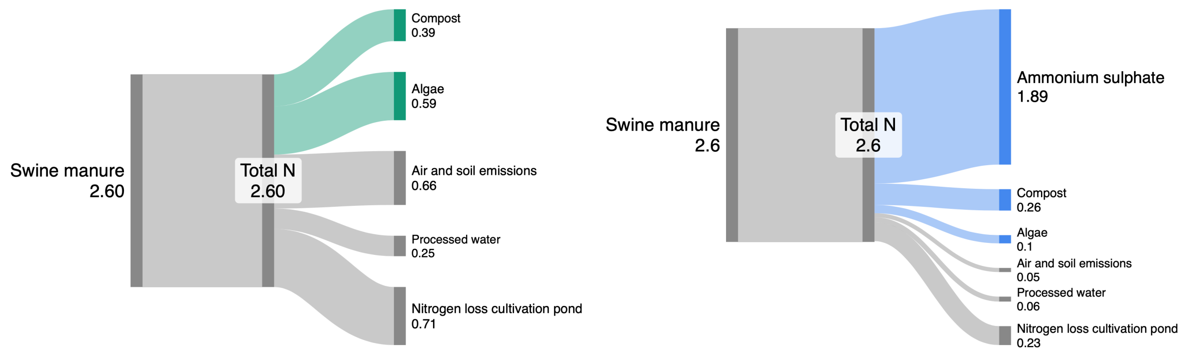


Figure 4.5: Nitrogen mass balance of total production process Biogas (left) and bioplastic (right) in kg N (input 1000 kg raw manure). Imbalance: 0.007 kg N 0.3% and 0.007 kg N 0.3%. Commercially viable products are colored in.

The nutrient recovery of nitrogen is shown in Figure 4.5, with largest inaccuracy of 3.3%. Nitrogen retention is 38% for the biogas scenario and 85%for the bioplastic one, which means that in the bio-plastic production, the nitrogen is better preserved compared to the biogas production.

It should be noted, that a fraction of nitrogen is not accounted for in the model, labeled with ”Nitrogen loss cultivation pond” in Figure 4.5, which correspondents with 27 and 8.8 % of the total in the biogas and bioplastic production.

Nutrient recovery into useful products was not appreciably improved by higher biogas or PHA yields (change <0.15%, data not shown).

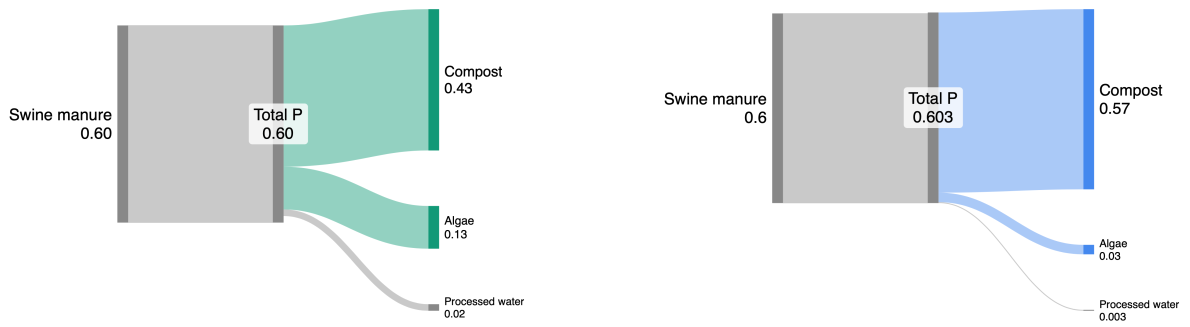


Figure 4.6: Phosphorus mass balance of total production process Biogas (left) and bioplastic (right) in kg P (input 1000 kg raw manure). Imbalance: 0.02 kg P 3.3% and 0.003 kg P 0.05%. Commercially viable products are colored in.

As is seen in Figure 4.6, the retention of phosphorus in biogas is 93 % and in bioplastic 99%. Both values indicate almost perfect retention of P in the system. The largest fraction of P ends up in the compost according to this model, with higher separation in the bioplastic scenario due to multiple decanters.

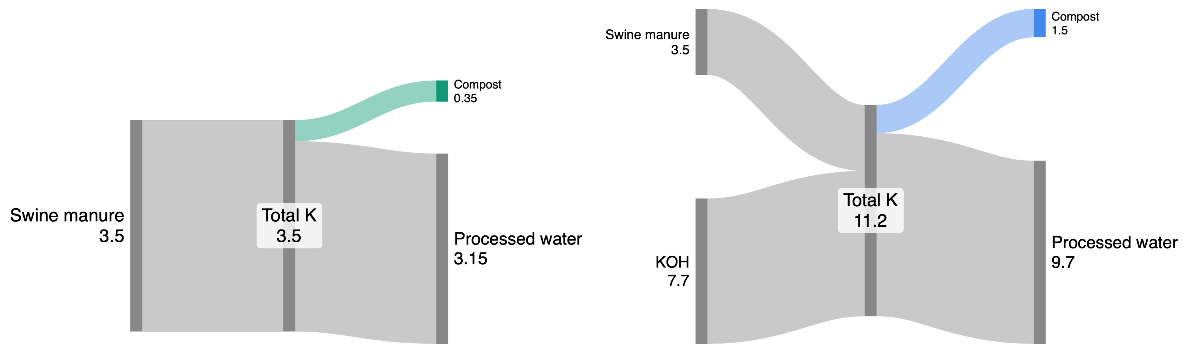


Figure 4.7: Potassium mass balance of total production process Biogas (left) and bioplastic (right) in kg K (input 1000 kg raw manure). Imbalance: 0.0 kg K and 0.0 kg K. Commercially viable products are colored in.

In Figure 4.7, it is shown that the majority of potassium ends up in the processed water, with nutrient recovery efficiencies of 10% and 14% for the biogas and bioplastic production scenarios, respectively. This distribution is consistent with the assumption that potassium predominantly remains in the liquid fraction during processing. As a result, a relatively high potassium concentration is observed in the processed water. However, since potassium is not typically considered a contributing factor to eutrophication, this does not pose a significant environmental concern within the scope of this study.

For potassium and phosphorus, no change in recovery was observed when the yields were improved, because the change in yields to not affect the phosphorous and potassium streams.

4.5. Environmental assessment

The interpretation of the environmental impact of both scenarios, the main contributors and possible changes are evaluated in this section.

4.5.1. Midpoint evaluation

The absolute values on impact categories of both processes is shown in Table 4.4.

Impact category	Unit	Biogas production	Bioplastic production
Global warming (GWP)	kg CO_2 eq	2.3×10^{1}	1.2×10^{2}
Terrestrial acidification (TA)	kg SO_2 eq	3.9×10^{-1}	3.0×10^{-1}
Marine eutrophication (ME)	kg N eq	1.9×10^{-2}	1.5×10^{-2}
Freshwater eutrophication (FE)	kg P eq	-8.3×10^{-4}	3.3×10^{-2}
Fine particulate matter formation (FPMF)	kg $PM_{2.5}$ eq	2.5×10^{-2}	1.1×10^{-1}

Table 4.4: Midpoint values for impact categories of biogas production scenario and bioplastic production scenario with functional unit of the treatment of 1 ton swine manure.

As shown in Table 4.4, the biogas production process has a lower overall environmental impact compared to the bioplastic production process, except for the terrestrial acidification and marine eutrophication categories.

4.5.2. Contribution analysis

To see what the impact categories are based on, a contribution analysis is done and shown in Figure 4.8 and 4.9 show the impact per process as a percentile representation of the total.

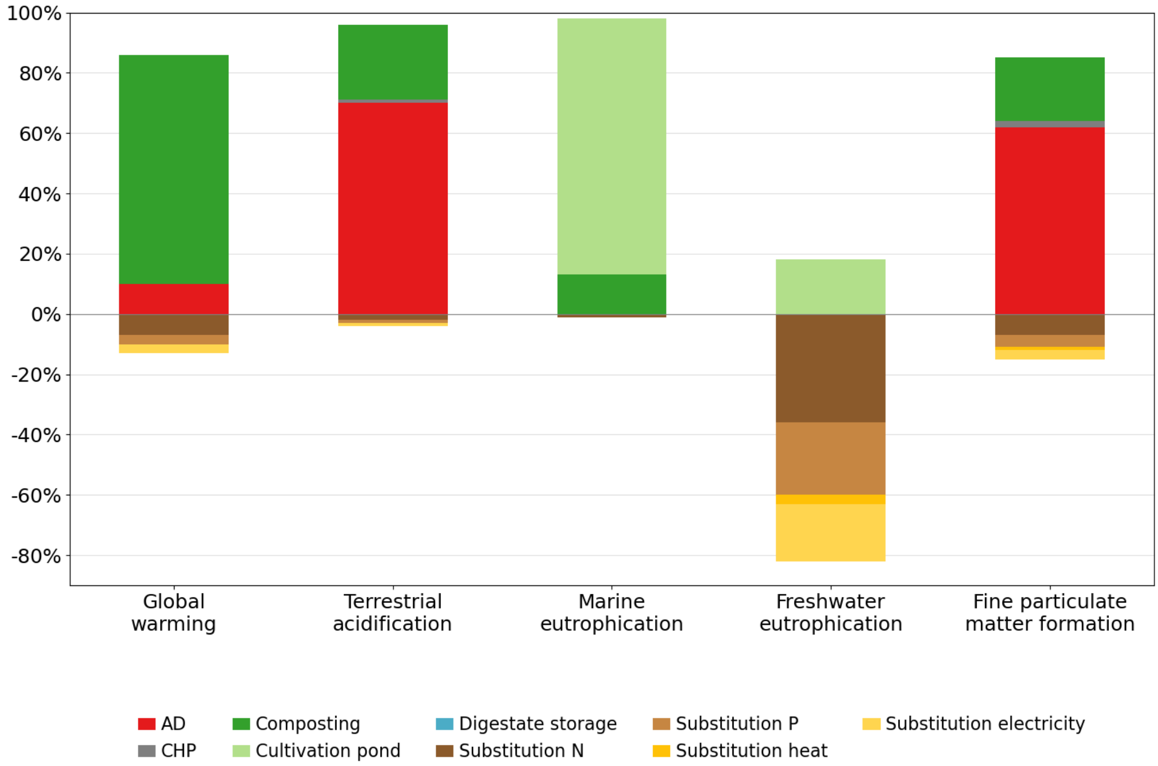


Figure 4.8: Contribution analysis of impact categories for biogas production scenario based on functional unit of 1 ton swine manure.

The contribution analysis in Figure 4.8 shows that for the biogas production process, composting dominates the GWP (approximately 80%), followed by the anaerobic digestion (AD) step (approximately 10%).

The high impact of composting is mainly caused by methane emissions, which are assumed to account for 8% of the carbon content. Given the high characterization factor of methane (approximately 40 times CO_2), even relatively small emissions significantly contribute to the overall GWP.

The terrestrial acidification impact category is mainly driven by emissions of NO_x and NH_3, which, according to the inventory Table 3.9, originate from the anaerobic digestion process (70%) and composting stage (25%). Only 1% of the TA is caused by the CHP. A small negative contribution is observed from fertilizer substitution, accounting for approximately 1% reduction for nitrogen and phosphorus substitution.

The marine eutrophication impact is mainly driven by nitrogen emissions from the outflowing water of the cultivation pond (85%) and the composting site (13%). These contributions can largely be explained by nitrogen in the effluent after the cultivation pond. During composting both air emissions and nitrogen leaching cause a impact on the ME. Given the small contribution of digestate storage to this category, the nitrogen leaching in this is not considered high.

The freshwater eutrophication is exclusively caused by the outflowing water of the foreground process, as also shown in Figure 4.8. A significant negative contribution is observed from the substitution of fertilizers, with approximately 36% attributed to nitrogen fertilizer substitution, 24% to phosphorus fertilizer substitution and 19 % to substituted electricity. Additionally, substitution of heat contributes to a further reduction in the overall impact.

The fine matter particulate formation originates 62% of the impact from the AD reactor and 21% from composting. The CHP unit contributes only 2% to the total impact. This relatively small contribution can be explained by differences in characterization factors between emitted substances, as well as lower emission associated with the CHP process. The effect of the AD on this category is caused by NH_3 emissions during this process.

Given the absolute values of the impact categories, the anaerobic digestion and composting site are identified as the processes impacting the environment most.

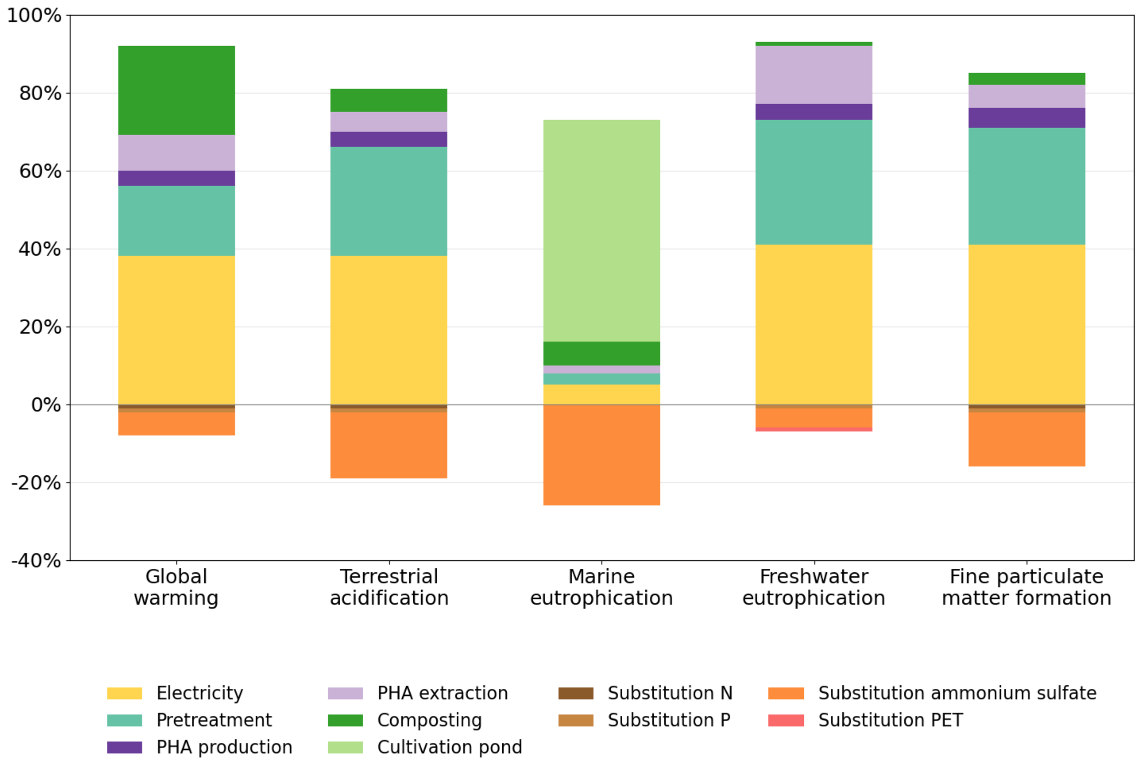


Figure 4.9: Contribution analysis of impact categories for bioplastic production scenario based on functional unit of 1 ton swine manure.

In Figure 4.9, a more fragmented contribution of individual process steps is observed compared to the biogas scenario. As shown in the inventory table, direct air emissions are primarily associated with the composting process, while for other foreground processes only CO_2 emissions are considered, which are considered to have no impact on all categories. Consequently, a significant part of the impacts originates from background processes such as the production of chemicals, electricity, and fuels.

The use of chemicals, such as chloroform, contributes indirectly to multiple impact categories due to the energy-intensive nature of their production. Chloroform is typically produced via chlorination of methane at elevated temperatures (400–500 °C), which results in substantial energy demand and associated emissions [101, 29]. This contributes to global warming potential (GWP) as well as eutrophication through the emission of nitrogen oxides (NO_x) from fossil fuel combustion, which subsequently deposit into aquatic ecosystems [97, 29]. A comparable effect is observed for potassium hydroxide (KOH) production, which is based on electrolysis and therefore strongly dependent on electricity use within the system boundaries [81].

Similarly, electricity use contributes to all impact categories, as the Spanish electricity mix includes a significant share of fossil-based generation. This leads to emissions such as fossil CO_2, NO_x and SO_x which drive global warming potential, freshwater and marine eutrophication.

As shown in Figure 4.8, the pretreatment step contributes more to all impact categories than the PHA extraction process. Although this may seem counterintuitive, it can be explained by the relatively higher mass and energy demand associated with pretreatment chemicals (e.g., KOH), compared to the lower amounts of chloroform required for extraction.

Composting contributes only significantly to GWP (23 %) in the bioplastic scenario. This can be attributed to increased diesel use, as approximately twice the mass is processed in the composting stage compared to the biogas scenario, as also reflected in the inventory table. Also, more mass is inserted in the composting, thus more methane is produced.

The cultivation pond remains the dominant contributor to marine eutrophication (57 %), similar to the biogas production scenario, due to comparable nutrient emissions.

Finally, the substitution of ammonium sulfate has a substantial influence across impact categories. This effect could be allocated to the pretreatment step, where the substitution occurs, which would reduce its apparent impact. The impact of the pretreatment step would be decreased by 12% for GWP, 11% TA, 29% ME, 27% FE and 15% FPMF. But as the ammonium sulfate positively effects the complete process, this does not influence the overall outcome.

In contrast, the substitution of conventional plastics has a negligible effect in Figure 4.9, except for a small contribution to freshwater eutrophication (approximately 1%).

The first and second contributing processes are summarized in Table 4.5.

Impact category	GWP	TA	ME	FE	FPMF
Biogas	Composting (80%) AD (10%)	AD (70%) Composting (25%)	Cultivation pond (85%) Composting (13%)	Cultivation pond (100%) -	AD (62%) Composting (21%)
Bioplastic	Electricity (38%) Composting (23%)	Electricity (38%) Pretreatment (28%)	Cultivation pond (57%) Composting (6%)	Electricity (41%) Pretreatment (32%)	Electricity (41%) Pretreatment (30%)

Table 4.5: First and second most contributing processes to biogas and bioplastic production scenarios.

4.5.3. Sensitivity analysis

Using the same maximum yields as before, the effect on the total system performance was evaluated for each impact category. The impact categories are stated in absolute values, in comparison between the base yield and maximal yield for both processed, shown in Figure 4.10

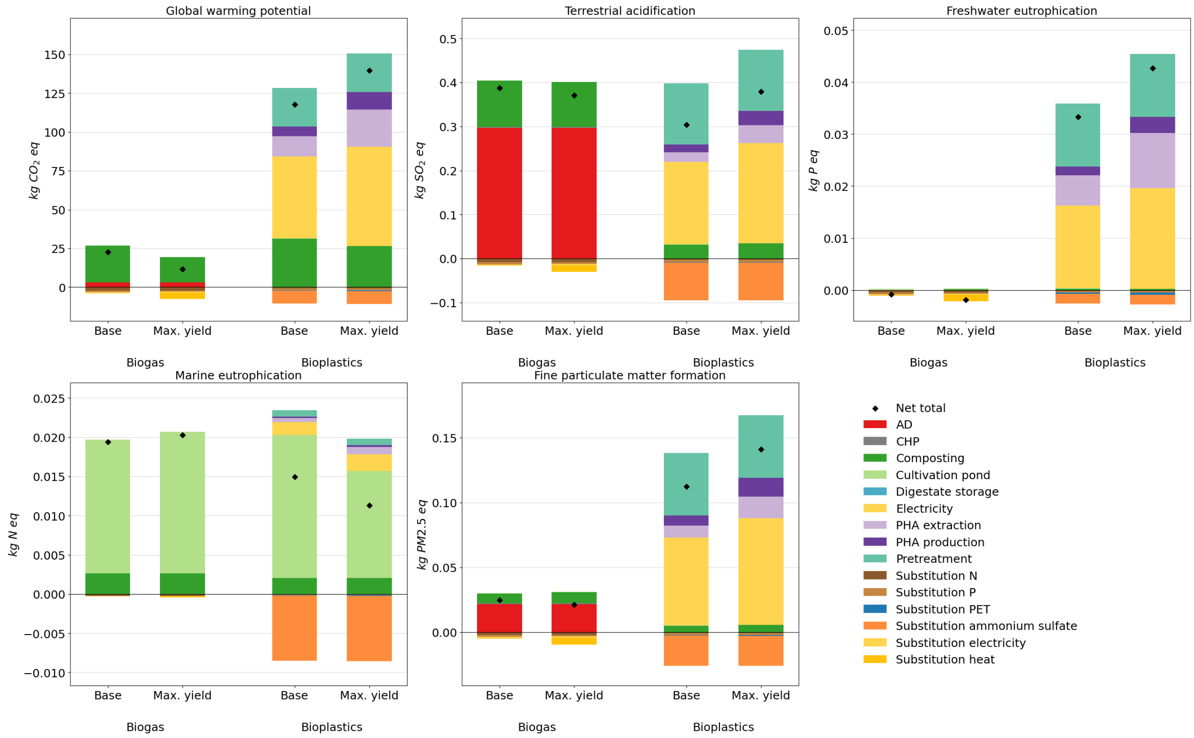


Figure 4.10: Environmental impact for all scenarios in absolute values for the treatment of 1 ton swine manure. For biogas (left) the scenarios of the base yield (30%) and maximal yield (70%), for bioplastic the scenarios of base yield (54%) and maximal yield (99%).

For the biogas scenario, yield improvements lead to a reduction in digestate production, resulting in lower emissions after anaerobic digestion. In addition, increased biogas production enhances the substitution of electricity and heat, thereby improving the overall environmental performance. The only exception is observed in the marine eutrophication category, where a slight increase in impact is detected. This can be explained by the reduced digestate volume, which requires less dilution water to achieve the desired TAN concentration in the cultivation pond. Consequently, the effluent becomes more concentrated in nitrogen, leading to a higher calculated impact. However, this effect is a result of the modelling approach and does not reflect an increase in the absolute nitrogen load. Therefore, it can be considered an artefact and does not significantly affect the interpretation.

In contrast, for the bioplastic production scenario, higher yields result in increased impacts across all categories. This is mainly attributed to the higher demand for electricity of the foreground process and chemicals consumption with background process effects. As more PHA is accumulated intracellular, additional energy and solvent input are required for recovery, increasing the overall environmental burden, leading to specific increased impact of the PHA extraction step..

Only within the marine eutrophication category, the contribution of the cultivation pond decreases with higher yields. This is due to the reduced digestate flow, which lowers the total nitrogen input into the cultivation system, thereby reducing nutrient-related emissions from this stage.

The changes in total value are presented in Table 4.6, reflecting the change compared to the base outcomes.

Table 4.6: Effect of yield improvements inside biogas and bioplastic production scenarios on impact categories. Effect defined as fractional change.

Impact category	Base value (biogas production)	Maximum yield	Base value (bioplastic production)	Maximum yield
Global warming $kg\ CO_2\ eq$	2.3×10^1	−48%	1.2×10^2	19%
Terrestrial acidification $kg\ SO_2\ eq$	4.3×10^{-1}	−4%	3.0×10^{-1}	25%
Marine eutrophication $kg\ N\ eq$	2.6×10^{-2}	5%	1.5×10^{-2}	−24%
Freshwater eutrophication $kg\ P\ eq$	-8.3×10^{-4}	−122%	3.3×10^{-2}	28%
Fine particulate matter formation $kg\ PM2.5\ eq$	3.4×10^{-2}	−15%	1.1×10^{-1}	26%

4.6. Techno-economic analysis

For the techno-economic analysis, first the purchase cost for the equipment are estimated, which lead to the capital investment costs. After that the operational costs are discussed per year and the potential revenue per year. This lead to a final conclusion on the feasibility of the biogas and bioplastic production.

4.6.1. Capital investment

The cost of the equipment is estimated with literature values and determination of the scale needed for the process is calculated in line with the mass flows of the model of both scenarios. The exact sizing and prices are shown in Appendix D, the results are visualized in Figure 4.11.

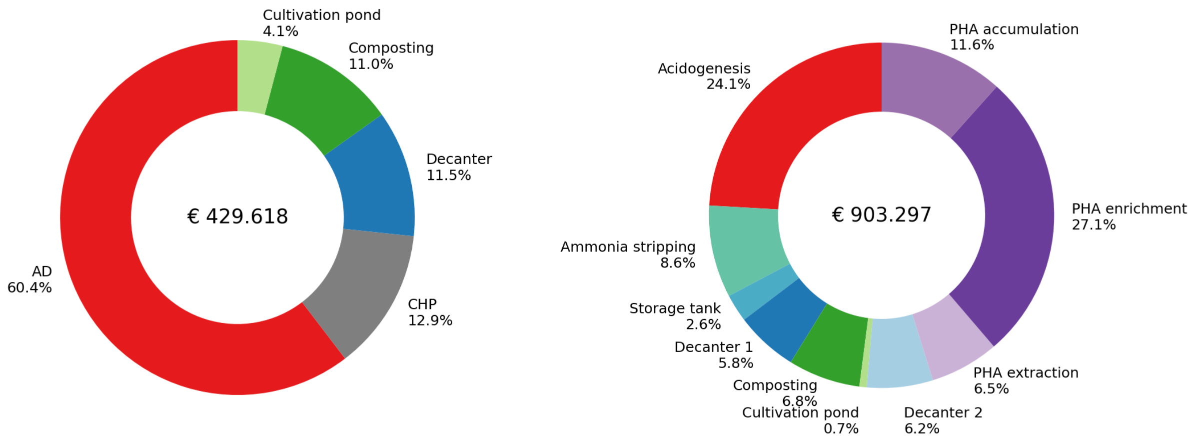


Figure 4.11: Total purchase cost equipment (PC). Left: biogas scenario, right: bioplastic scenario.

In the biogas production scenario, the total purchase cost for the equipment is estimated to be €432,423 in total. The anaerobic digestion (AD) reactor represents the largest contributor. This is in line with expectations, as the AD reactor is the first and largest unit operation in the process, requiring a significant volume of 927 m^3. The estimated cost of €262,239 for the reactor is relatively moderate. According to Girón-Rojas et al. [50], a cost relation for the anaerobic digester, CHP, and digestate storage infrastructure results in a price of €431,366 for the AD reactor, after subtracting the costs of the CHP and storage used in this study. This is approximately twice as high. However, given that the AD reactor cost is explicitly reported by Hollas et al. [59] (2022), and considering the inclusion of additional costs through the Lang factor, the price used here is considered appropriate.

The total CAPEX of Girón-Rojas et al. [50] for the comparable system, excluding composting and microalgae cultivation, is €457,508 which is in reasonable agreement with our estimation of total purchase costs.

The total purchase cost of the bioplastic production scenario is approximately twice as high as that of the biogas scenario. However, considering that this process includes four additional unit operations, this increase is not excessive. The acidogenesis, PHA accumulation, and PHA enrichment steps require the largest capital, given the required bioreactors. The cost estimations are taken from Krömer et al. [88], the acidogenesis reactor (52 m^3) is estimated at a price €217,638. After the bioreactors, the ammonia stripping column is the large contributor that consists of €78,014[141]

For the bioplastic scenario, Khan et al. [83] report a CAPEX of €3,871,377 for a PHA production capacity of 5,000 t/y, corresponding to a primary sludge flow of 843,313 t/y. Scaling this value down to the capacity considered in this study yields an estimated capital cost of €245,427. However, directly applying a down-scaling factor of 1000 to the CAPEX introduces significant uncertainty and is therefore not fully reliable. Even at a reduced processing capacity of 8,500 t/y of manure, the process still requires essential unit operations such as pumps, decanters, reactor vessels, and control systems. These components represent fixed or semi-fixed costs and do not scale linearly with throughput. As a result, the total investment does not decrease proportionally with capacity, and assuming a reduction to 1% of the original CAPEX likely underestimates the true cost. This estimation also assumes that the same yield is maintained across scales. If the system were instead scaled based on actual PHA production, the resulting CAPEX would be even lower than the value derived here, further highlighting the limitations of simple linear scaling.

Compared to the Equipment Purchase Cost of Ventura and Ventura [153] for PHB production from corn stover, a scaled value of 13.5 million euros is calculated, which is around 15 times larger than the purchase cost of this study. When the purchase cost is scaled to the PHB production in this study and the observed production of Ventura and Ventura [153], a equipment cost fo 1.95 million is observed. This is in the same ball park as the estimation done in his study. A comparison of a similar size would improve the comparison, but no mention of this is found.

The total investment required for the treatment plants is presented in Table 4.7, where the total purchased equipment cost includes an additional 10% to account for unlisted equipment.

	Biogas production	*Bioplastic production*
Total purchased equipment costs (PEC)	€ 475,665	€ 971,846
Direct Fixed Capital (DFC)	€ 1.664.828	€ 3.974.850
Working capital	–	€ 728.884
Total investment cost	€ 1.990.010,45	€ 4.415.715,40

Table 4.7: Total investment cost for capital. Lang factor of 3.5 for biogas and 4.09 for bioplastic production assumed. For biogas (left) the scenarios of the base yield (30%) and maximal yield (70%), for bioplastic the scenarios of base yield (54%) and maximal yield (99%).

The direct fixed capital (DFC) required for the biogas production plant is €1,664,828, considering a Lang factor of 3.5. The DFC of the bioplastic production scenario is €3,974,850, including a higher Lang factor due to the more complex process design. The total investment cost of the biogas production plant is assumed to be equal to the DFC, as no working capital is considered necessary. For the bioplastic production plant, working capital is included, resulting in a total investment cost of €4,415,715.

4.6.2. Operational costs

For the operational cost per year, the assumptions stated in the Methods (section 3.3.2) are applied on the DFC, the listed PEC and labor cost. For the laboratory quality control of the biogas production plant, it is assumed that only 5 % lab control has to be applied to this system. This is, given the low demand of control over the system, as the system is very robust in operation, and low control is needed [119]. The required labor hours are taken from literature, without incorporating the size of the reference plant, as it is assumed that the labor demand depends primarily on the number and complexity of the unit operations rather than the plant size [83, 50]. In Figure 4.12 the operational cost are shown.

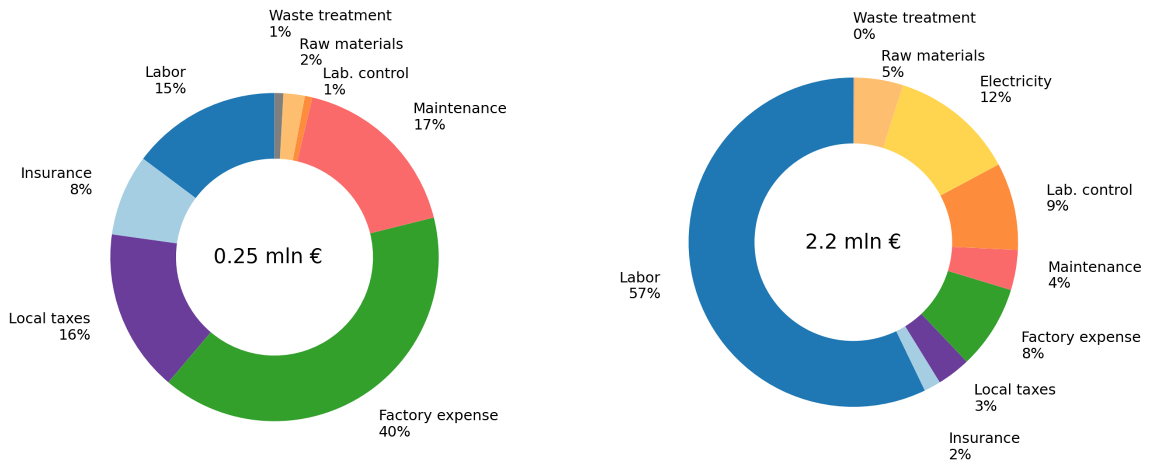


Figure 4.12: Annual operational cost of biogas production plant (left) and bio-plastic production plant (right).

As shown in Figure 4.12, the main contributor to the operational costs in the biogas production scenario is the factory expense. Factory expenses, also referred to as manufacturing overhead, include indirect costs required to operate the plant, such as indirect materials, indirect labor, and quality control staff that cannot be directly assigned to a specific product. In this study, the factory expense is estimated as 5% of the direct fixed capital (DFC), which is a commonly applied approximation [141].

In contrast, the bioplastic production scenario is dominated by labor costs. This difference is primarily driven by the significantly higher labor demand: 730 hours per year for the biogas production scenario compared to 25,502 hours per year for the bioplastic production scenario. The same hourly wage is applied in both cases. Although a more complex plant would typically require higher-skilled (and potentially higher-paid) personnel, the applied wage of €50.24/h is considered reasonable, as Khan et al. [83] report a lower wage of €31.2/h for a comparable PHB production plant in Sweden.

The remaining operational costs amount to €915,788 and include maintenance, raw materials, utilities, and other minor contributions.

A comparison with literature highlights notable differences. For the biogas production scenario, an operational cost of €46,242 is reported by Girón-Rojas et al. [50], when scaled to the capacity considered in this study. This value is approximately five times lower than the result obtained here. However, in that study only maintenance costs are included in the OPEX, while other operational activities are assumed to have negligible cost. As this represents a simplified approximation, the reported value is considered to underestimate the true operational costs. By Hollas et al. [59], a scaled operational cost of 36 thousand € per year is estimated, including a composting process, but no micro-alage pond. However, this is a calculation done for Brazilian markets, which can lead to significant lower costs in utility and labor.

For the bioplastic production scenario, scaling the literature value by capacity results in an estimated annual operational cost of €140,842.94, which is approximately 15 times lower than the value obtained in this study [83]. This discrepancy can largely be explained by the dominant contribution of labor in the present analysis. As shown in Figure 4.12, labor accounts for 56% of the total operational cost. Since the labor hours were not scaled with capacity, but instead taken directly from the reference study, the resulting OPEX is significantly higher than the scaled literature value.

Furthermore, the contribution of electricity costs in the bioplastic production scenario is only 0.1% of the total OPEX. This is not consistent with literature values, where Khan et al. [83] report electricity to account for 61% of the operational cost. In the same study, labor contributes only 6% to the total OPEX, which is also significantly lower than the value obtained here. This discrepancy can be explained by the different scaling behavior of cost components: labor is largely independent of plant capacity, while electricity demand typically scales linearly with process throughput [119].

4.6.3. Revenue

Based on the mass balances obtained from the model, the revenue can be estimated. For the biogas scenario, the largest share of income is generated by the algae, for the bio-plastic scenario this is done by the PHA powder. The revenue is shown in Table 4.8

Table 4.8: Annual revenue streams for biogas and bioplastic production

Biogas production	Revenue [€]	*Bioplastic production*	Revenue [€]
Algae	37,116 (52%)	Algae	6,367 (5%)
Compost	20,463 (29%)	Compost	29,990 (25%)
Electricity & heat sold	11,425 (16%)	PHA powder	65,483 (55%)
CO_2 avoided	1,537 (2%)	Ammonium sulfate	17,772 (15%)
Total	**700,541 €**		**119,614 €**

When the revenue is related to the gross profit, the results are presented in Table 4.9. For this calculation, the depreciation is determined using an interest rate of 3.24% [146] and Equation 3.43. Depreciation can on one side represents the annual cost associated with the capital investment, on the other side reflect a non-cash expense, meaning that no taxes are paid over this amount, and it can therefore be considered an indirect financial benefit. For the gross profit calculations, the depreciation is used to account for the annual cost of the direct fixed capital.

Accordingly, the gross profit is calculated by subtracting both the depreciation and the operational costs from the total revenue ,seen in Table 4.9

	Biogas production [mln. €]	*Bio-plastic production* [mln. €]
Operational cost	-0.25	-2.2
Depreciation	-0.14	-0.31
Revenue	0.071	0.11
Gross profit	- 0.32	-2.4

Table 4.9: Profit per year

As shown in Table 4.9, both systems are not economically feasible.

To further assess whether profitability can be achieved, the price of the main products is analyzed relative to the operational costs, by using equation 4.1.

$$Operational\ cost\ MP = \frac{OPEX}{Mass\ MP\ produced\ per\ year} \tag{4.1}$$

For biogas, this results in 11.48 € /kWh, and for bio-plastic, 102.58 € /kg. Compared to the respective selling prices of 0.15 € /kWh and 2.65 € /kg PHA, the operational costs are significantly higher.

To address this issue, process optimization is investigated, for which several scenarios are evaluated. Improving process efficiency (Scenario I) could increase product yields and thus enhance revenue.

Since algae and compost are by-products in the biogas and bio-plastic production, their quantities cannot be directly increased through yield improvements. Instead, they depend on the residual nutrients in the system. However, improved yields affect the total revenue and allow for a reassessment of raw material and waste management costs. The overall impact is illustrated in Figure 4.13.

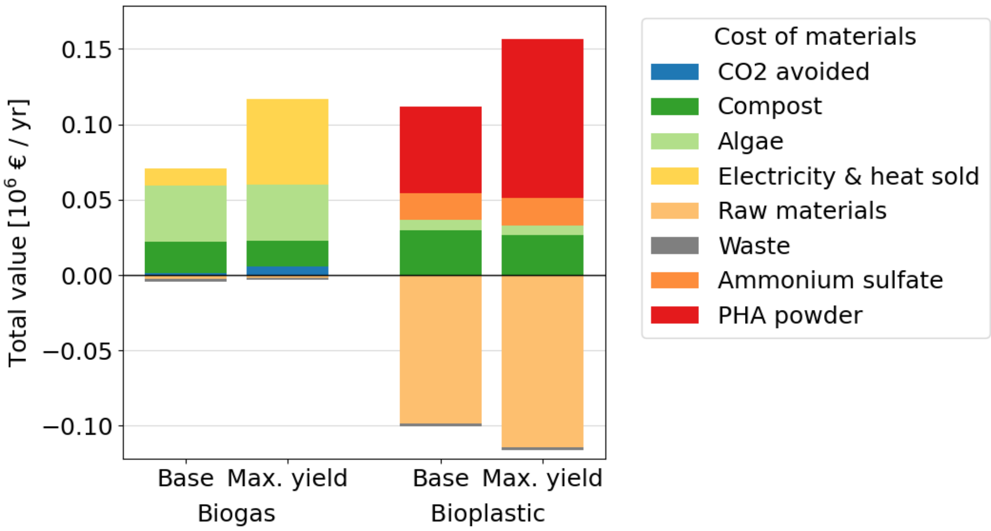


Figure 4.13: Impact of yield improvement on revenue of products and costs of raw materials and waste

In Figure 4.13, the change in revenue, raw materials and waste is shown over improved yield of the anaerobic digestion and acidogenesis. With improved yields in the biogas scenario 57% more revenue is produced, leading to a effect of 42 thousand euros. The bioplastic revenue was 29% improved, but the raw materials need as well and a effect of 28 thousand euros more was observed. In Table 4.9, it is shown that to reach a break-even point for the gross profit, a gap of €0.32 million (biogas) and €2.4 million (bio-plastic) needs to be closed. As shown in Figure 4.13, this gap cannot be closed within realistic yield improvements for the biogas and bioplastic scenario.

To estimate the required production for a break-even situation, the necessary mass of the main product is calculated. Biogas and PHA powder are considered the primary products, as their production increases with higher reactor yields, as shown in Figure 4.13.

To achieve break-even, the required production is calculated using Equation 4.2.

$$Mass\ MP[kg] = \frac{OPEX + Depreciation}{sellingprice[/kg]} \tag{4.2}$$

Earlier, in the sensitivity analysis of the model, Figure 4.3 showed that at maximum yields, biogas production reaches 7.65×10^4 kg/year and PHA powder 3.99×10^4 kg/year. In terms of scale, for the biogas production approximately 4.92 $\times 10^5$kg/year would be required, to obtain a break even point. This is 31 times higher than the maximum achievable production. For bio-plastic production, 7.09×10^5 kg/year would be needed, which is 18 times higher than the theoretical maximum production at 99% yield.

Since yield improvements alone are insufficient to achieve a neutral gross profit, the impact of increased selling prices is also evaluated (Scenario II). The prices of algae, compost, and PHA powder may be higher than the conservative estimates used. In contrast, the prices of biogas (converted to electricity and heat) and ammonium sulfate are observed to remain relatively stable over time [43], IMARC Group [66], with fluctuations per month due to political events left out of this consideration.

Applying higher-end market prices for algae, compost, and PHA results in annual revenues of €0.27 mln and €0.58 mln for the biogas and bio-plastic scenarios, respectively. When these improved prices are combined with yield improvements, the gross profit for the scenarios stay negative. The results are summarized in Table 4.10, after possible improvements in the capital cost are considered.

4.6.4. Sensitivity analysis of the capital investment

Further improvements in the gross profit could be achieved if the capital costs are lower than currently estimated. As discussed in Section 3, the Lang factor is used to estimate all additional costs beyond the equipment purchase costs. For the biogas scenario, this factor is already set at the lower bound of the typical range, as the biogas production plant is assumed to require minimal additional construction [120].

For the bioplastic scenario, however, Lang factor reduction is investigated. Although the expected value is set at 4.09 [121], a theoretical reduction of this factor would decrease the direct fixed capital by approximately €0.5 million. Nevertheless, due to the uncertainty and lack of justification for such a reduction, this scenario is not included in the analysis.

On another note, it is observed that biogas production plants in Europe often receive financial support from the European Union in the form of subsidies for green energy generation. In the case study by Martínez-Hernando et al. [101], which considers bio-methane upgrading using micro-algae from swine manure, a grant of €1,237,605 was awarded [40]. Given the similarities to the biogas scenario in this work, it is assumed that a comparable grant could be obtained.

For the bioplastic scenario, a comparison can be made with the CIRCULAR BIOCARBON project, which developed a first-of-its-kind commercial-scale biorefinery that converts municipal organic waste and sewage sludge into high-value bio-based products such as bioplastics, green graphene, and fertilizers. The total project cost was €23 million, of which €15 million was funded by the European Union, corresponding to approximately 65% of the total budget[38]

Based on this ratio, a similar subsidy fraction is assumed for Scenario III, resulting in an estimated grant of €2,870,215. It should be noted, however, that EU grants are subject to strict requirements, and factors such as economic feasibility and process efficiency are evaluated before funding is awarded.

When these subsidies are subtracted from the direct fixed capital, the annual capital cost represented by the depreciation is recalculated and implemented in the gross profit in Table 4.10, for the base scenario of revenue and the improved yield, with altered selling prices and EU grant scenario.

Table 4.10: Gross profit scenarios

Gross profit	*Biogas production* [mln. €]	*Bioplastic production* [mln. €]
Base scenario	-0.32	-2.4
Scenario I	-0.28	-2.4
Scenario II	-0.096	-1.8
Scenario III	-0.21	-2.2
Scenario I+II+ III	0.033	-1.5

In Table 4.10, it is observed that in scenario I, a slight improvement in gross profit is made for the biogas production, but no substantial difference in the bioplastic production. For scenario II the gross profit of biogas and bioplastic production is less negative (thus better). The improved scenario III alone results in less negative gross profits for both scenarios compared to base and only with all modifications, the gross profit becomes positive for the biogas production plant. But, with a tax of 25%, the net profit becomes only € 25 thousand. This leads to a Net Present Value of -4.1 thousand € for a project life time of 15 years. The NPV of the bioplastic production plant is - 443 thousand €. This means that the plant does not generate enough annual profit to justify the initial capital cost under these specific conditions. So in the end, given all possible modifications, the process stays unfeasible from a economic point of view.

5
Discussion

The result are discussed in this section in relation to literature and accuracy to reality.

5.0.1. Model biogas

The diesel consumption in the composting step in this study is relatively high, with 2.8 L per ton of input applied. In comparison, significantly lower values of 0.91 L diesel per ton of input have been reported in the literature [30]. This discrepancy suggests that both operational costs and associated emissions could be reduced under more accurate assumptions.

The 7.17 kg of algae produced is estimated based on phosphorus limitation. However, compared to the study of Martínez-Hernando et al. [101], significantly higher algae production could be achieved. In that study, 7.3×10^2 kg of algae is produced per day in a pond of 320 m^3, whereas only 7.7 kg of algae per day is produced in the 140 m^3 pond of this study. This difference is substantial. This discrepancy can be explained by the method used to estimate algae production. Although the approach from Guillen et al. [55] was applied and seems in theoretically logical, the assumed growth limitation may not accurately reflect a actual system. As a result, the true biomass growth of algae is likely higher than currently estimated.

In the microalgae pond, a mass loss of nitrogen is observed in the nitrogen balance over this unit operation. Since phosphorus is the limiting nutrient, the amount of algae growth was determined based on phosphorus availability. Consequently, a nutrient removal efficiency is applied in the calculations, but not all nitrogen is assimilated into the algal biomass. The remaining nitrogen can partition into several pathways. One possible route is volatilization to the atmosphere as NH_3, as algal growth increases the pH of the pond, the chemical equilibrium shifts towards gaseous ammonia, promoting NH_3 emissions. This is supported by Zimmo, Steen, and Gijzen [165]. Another explanation is the presence of anaerobic micro-zones within the pond, where denitrification may occur. In these zones, nitrate can be reduced to N_2 gas and subsequently released into the atmosphere. However, due to the absence of measured nitrogen gas emissions in comparable studies, no definitive conclusion can be drawn regarding the exact fate of nitrogen in [101].

Nevertheless, given the significant nitrogen mass loss observed in this system, in combination with literature findings, it is most likely that nitrogen is emitted in gaseous forms such as NH_3, NO_x, N_2O, or N_2. This leads to unaccounted environmental impacts for possibly terrestrial acidification, marine eutrophication, fine particulate matter formation and global warming potential [117, 16].

Upon revising the manure input data, significant variability is observed in Table 3.2. The variation amounts to 89% for total solids, 64% for volatile solids, and 25% for total ammoniacal nitrogen. These parameters strongly influence nearly all stages of the process. An increase in VS directly enhances methane production potential in the anaerobic digestion step, or higher VFA production yield. With higher TAN concentrations, earlier ammonia inhibition in the microalgae pond would occur, requiring increased dilution and consequently larger pond volumes to maintain stable operation. But since the values were averages over annual observations in multiple farms, the averages can be used with certainty.

Biogas production

In the biogas production process, the base scenario results in 5.4 kg biogas is produced, which is 4.7 m^3, per ton manure. According to [84], 23 m^3 biogas per wet tonne is produced by swine manure, but the TS of this manure is also 10 times higher. Santonja et al. [132] reports general biogas production for pig and cattle slurry of 14 - 25 m^3 per tonne in Europe. The yield improvement to 70 % biodegradability is more in this range, as around 11 m^3 biogas is produced in this case. However, the methane yield depends on the temperature of the reactor, which should be considered in comparisons.

The biogas compositions and LHV are as expected (63 % CH_4, 6.2 kWh/ m^3) [84, 148]. In regard of real life applicability, the H_2S in biogas should be considered as this is required to be scrubbed, as high concentrations are expected in biogas from manure [84].

The total electricity demand of 7.6 kWh / ton is considered to be low. In the anaerobic digestion, a electricity demand of 0.25 kWh/ ton manure was assumed [59]. By Martínez-Hernando et al. [101] an electricity demand of AD of 7.6 kWh/ ton manure is noted, which is considerably much higher than our value. Also, Girón-Rojas et al. [50] notes an electricity demand of the total process of 16.2 kWh/ ton manure is observed, for the AD, CHP and S/L separation only. Both studies indicate that the electricity demand of this study is underestimated, affecting both LCA and TEA.

The total electricity demand of 7.6 kWh per ton of input is considered relatively low. In this study, an electricity demand of 0.25 kWh per ton of manure was assumed for the anaerobic digestion process, based on [59]. However, [101] reports a significantly higher electricity demand of 7.6 kWh per ton of manure for AD alone. Furthermore, [50] reports a total electricity demand of 16.2 kWh per ton of manure for a system including AD, CHP, and solid-liquid separation. These values are substantially higher than those assumed in this study. Both studies suggest that the electricity demand in the present analysis is likely underestimated, which may lead to an underestimation of both environmental impacts in the LCA and operational costs in the TEA.

The same is observed for the heat demand of the AD. By Hollas et al. [59] the assumption of 0.77 kWh heat is taken in this study, but given the location of this plant, being Brazil, this heat could be underestimated for Spain, given climate differences. For example, Girón-Rojas et al. [50] accounts for a heat demand of 22 kWh/ ton manure in Spain for AD, CHP and S/L separation. In the study of Martínez-Hernando et al. [101], no heat is accounted for in the AD, but severe lower yields of biogas in the digester were observed. If a higher heat demand was required, fo 22 kWh/ ton, the total combustion of the biogas would not lead to enough heat production, even if the CHP efficiency of Girón-Rojas et al. [50] is used, being 55% thermal efficiency compared to 46% implemented in this study. This means that biogas yield are probably even lower than used here, or heat provided from an other source should be used.

Bioplastic production

A total PHA production of 3 kg PHA per ton of manure is obtained, corresponding to 0.13 kg PHA per kg VS. Comparable yields have been reported in studies on food waste, which is in line with expectations [158]. This similarity can be explained by the comparable extraction efficiency applied, as well as similar VFA production yields. In the food waste study, VFA production ranged between 0.35–0.5 g COD_{VFA}/g VS, which is consistent with the value of 0.57 g COD_{VFA}/g VS obtained in this study.

The assumption that hydrogen gas is not inline with the stoichiometric reaction, is counter intuitive. The electron sink that after this adjustment needs to be filled is 1.74 e^- /reaction. As this cannot be completely related to biomass growth, it is expected that these electrons are accepted in the production of other intermediates of the anaerobic digestion pathway in Figure 2.1. In acidogenesis, production of ethanol is observed normally, which is not implemented in the mass balance now, but could explain the electron sink [135].

The PHA production yield of 0.45 g COD$_{VFA}$/g COD$_{PHA}$ over the enrichment and accumulation phases is in line with literature values reported by [30], which range between 0.50–0.80 g COD/g COD for VFA-to-PHA conversion.

A range between 0.11 and 0.26 g PHA/g COD is reported by Wu et al. [158] for different food waste streams over the total process. The yield obtained in this study (0.15 g COD PHA/ g COD manure) is significantly higher than values reported for manure-based systems . For example, Guho et al. [54] report a yield of 0.035 g PHBV/g COD for dairy manure at pilot scale, which is mainly attributed to a low acidification efficiency of approximately 12% (gCOD$_{VFA}$/gCOD$_{in}$).

It should be noted that the assumed acidogenesis yield is based on laboratory-scale data and typically decreases upon scale-up. Guho et al. [54] report a reduction of approximately 18% when scaling from bench-scale to pilot-scale. A similar decrease may be expected in this study; however, this effect has not been accounted for and may lead to an overestimation of the PHA yield. Next to this, the VFA distribution used, is just an representative of what the VFA mixture could contain. Multiple studies show different distributions in the acidogenesis of swine manure, changing over different conditions [62].

Overall, the yield obtained in this study falls within the expected range .

The electricity demand of the bioplastic production process also introduces uncertainty, as it is based on pilot-scale PHA production data [158]. Since the values are expressed per kg of PHA produced, the assumption of equal efficiency between pilot- and industrial-scale systems would justify this approach. However, as process efficiencies generally improve at larger scales, the electricity demand may be overestimated.

5.1. Environmental impact

5.1.1. General considerations

Considering the boundaries of LCA results is crucial in comparing the values to literature. The functional unit of this study was chosen as 1 ton manure, which is done broadly in literature, but what has been pointed out before, the content of the manure determines also the environmental impact. Higher DM contents results in potential more methane emission, and so on. So, for the comparison in literature, studies are indicated by origin and if possible chosen on locations in Spain. This will make a fairer comparison, but slight differences can still effect the results.

A comparison between the end of life effects of the products would also improve the comparison. For example, PHA has better biodegradability in natural marine and freshwater conditions, and can be used as a more circular product, as it has been reported as a substrate for AD. In contrast, conventional plastics are often incinerated, leading to substantial negative impacts on human health, ecosystems, and resources [57, 13]. The algae and compost that are produced in both processes are ultimately used as a fertilizer, but emissions of this process was not included for now. Application can significantly contribute to terrestrial acidification and freshwater eutrophication, but can also improve the performance of both scenarios in this study.

Conventional mineral fertilizers provide immediately available nutrients, resulting in high short-term plant uptake (60%) and rapid yield response, but also cause significant environmental losses through volatilization and leaching, contribute to soil degradation, and have high energy demands during production. In contrast, compost acts as a slow-release fertilizer with lower initial nutrient uptake (30%) but provides long-term nutrient supply, improves soil structure and biology, and supports soil health, although it may lead to heavy metal accumulation over time. Microalgae-based fertilizers also function as slow-release systems but additionally offer biostimulant properties, enhancing plant growth, stress tolerance, and soil microbial activity. Compared to conventional fertilizers, microalgae have a lower environmental footprint by recycling nutrients from waste streams and reducing risks such as soil salinization. Factors such as initial nutrient levels, pH, microbial activity, and soil composition strongly influence nutrient mineralization and plant uptake. As these effects are largely dependent on soil conditions and crop type of the farm using the fertilizer, no definite conclusions could have ben made in this study [161, 3, 21, 136, 9].

This also highlights the effect of the substitution assumptions of the fertilizers. As the soil conditions determine the need for nutrients and the ability to absorb them, the assumptions of 30 % working efficiency of the compost and 30 and 50 % for the algae fertilizers could be studies in a site-specific situation, to further improve the accuracy.

The same can be said for the emission factors of the composting process, where case specific experiments would also improve the accuracy of this study. In absence of specific data, the emission factors are based on digestate from the co-digestion of dairy manure with maize silage in Germany. This was done, as Kupper et al. [89] has explained that emissions of raw manure and digested manure are different during composting. Compared to raw manure emission factors of the IPCC, higher methane (8.1 % of C versus1.7% of C [76]) and N_2O emissions (1.4% versus 0.5% of TN) were applied. In contrast, for NH_3 and NO_x emissions lower factors were used (20% of NH4-N versus 65% of TN). But, to take into account more context-specific conditions, more representative factors could be determined, leading to higher accuracy of the results.Kupper et al. [89],

An other emission factor that effects the outcomes of the LCA, is the NH_3 emissions in the AD step. The IPCC indicated that a significant fraction of NH3 and NOx emissions originates from post-digestion storage rather than the digestion process itself. This could have led to overestimation of emissions in the AD step. In the metabolic pathway, the absolute NH_3 production due to AD can be used to verify if this emission factor was overestimated (section 3.1.1). The mass balance resulted in 0.154 kg NH_3 / t manure, which is close to the gas emissions reported in the inventory table, based on IPCC values (0.121 kg NH_3 / t manure).

At last, the choice of impact categories determine overall performance of a process. The impact categories were considered most appropriate for swine manure management and this study specific, but considering the chemicals used in the PHA extraction, more impact categories could be added to further investigate this effects, next to the impact of potential hazardous heavy metals in the products. For the goal to compare two manure systems, these impact categories were however appropriate.

For the heat and electricity demand of the biogas and bioplastic production, more realistic estimates could further improve the accuracy of the outcome. Biogas production performs well because of emissions avoided by the use of biogas as source of heat and electricity. To keep the analysis as realistic as possible, electricity from the grid is used as a energy source. In the future, changes renewable electricity in the electricity grid could lead to better outcomes for the bioplastic scenario. In a study modeling manure management it was found that that simply replacing a standard national electricity mix with a renewable electricity mix in SimaPro reduced the overall single environmental impact score by 26% lower [136].

The separate impact categories are now discussed in their relation to other comparable systems.

Global warming potential (GWP)

As can be seen in Table 5.1, the GWP of the biogas scenario is around five times smaller than the bioplastic scenario. The GWP of the biogas production system is in the same order of magnitude as values reported in literature. Compared to a system involving little to no intervention, an impact of 39.2 kg CO_2-eq per ton manure was observed. This value is approximately twice as high as biogas scenario, indicating this is favored over no treatment for GWP impact [4].

For the bioplastic production scenario, a higher environmental impact than no treatment is observed in Table 5.1, meaning that for GWP, the bioplastic scenario is worse. Studies on more intensive treatment of manure, like membrane separation, showed similar values [52]

Applying only AD and no composting or algae cultivation, much less impact has been observed by Álvaro et al. [4]. The contributional analysis (Figure 4.8) further narrows this difference down to only the composting step, meaning that a large differennce in GWP could be observed if no composting was done.

Table 5.1: Comparison of global warming potential (GWP) impacts expressed in kg CO_2 eq. per ton of treated manure.

Process category	Description	GWP [kg CO_2 eq.]	Location	Source
Biogas production scenario	AD, biogas combustion, S/L separation, composting and micro-algae cultivation	2.3 *10^1	Spain	This study
Bio-plastic production scenario	Ammonia stripping, VFA production, PHA production and extraction, composting, micro-algae cultivation	1.2 *10^2	Spain	This study
No treatment	Open storage (45 days), transport, application on the land	39.2	Spain	[4]
	Covered storage (45 days), transport, application on the land	31.4	Spain	[4]
	Open lagoon storage (6 months), transport, application on the land	113	Spain	[52]
AD	AD, biogas combustion, outside digestate storage (28 days), transport, application on the land	8.95	Spain	[4]
	AD, S/L separation, micro-algae cultivation & biogas upgrading to biomethane, digestate application	30.4	Spain	[101]
Membrane separation	Closed storage, GPM separation, ammonium sulfate and treated manure transported and applied on land	97	Spain	[52]
PHA production	OFMSW use for PHA production, with composting, PHA extraction with chloroform	114	Italy	[30]
	OFMSW use for PHA production, with composting, PHA extraction with ethyl acetate	-25.9	Italy	[30]

Terrestrial acidification (TA)

Terrestrial acidification is driven by NH_3, NO_x and SO_2 emissions, which performed approximately 1.3 times better in the bioplastic scenario compared to the biogas scenario (0.30 versus 0.39 kg SO_2 eq.) This is largely attributed to the ammonia stripping as pretreatment step, which captures 95% of the total ammonia into ammonium sulfate.

In Table 5.2, it is shown that both processes perform better than literature values. This can be explain by three reasons. First, the reference systems include the application of compost to land, which accounts for 80% of the terrestrial acidification in the study of Hollas et al. [60].

Secondly, the reduction in NH_3 emissions in the digestate storage leads to lower TA impact. This is supported by Hollas et al. [60], that compare systems with and without AD, resulting in decreased NH_3 emissions during storage and land application of digestate.

As third reason, the nitrogen mass loss identified in the micro-algae pond of this model could lead to underestimation of the results, if NH_3 emissions were emitted. This should however first be confirmed before any conclusions can be drawn, as similar LCA's do not report these emissions [101].

Table 5.2: Comparison of terrestrial acidification (TA) impacts expressed in kg SO_2 eq. per ton of treated substrate.

Process category	Process description	TA [kg SO_2 eq.]	Location	Source
Biogas production	AD, biogas combustion, solid/liquid separation, composting, and microalgae cultivation	0.39	Spain	This study
Bioplastic production	Ammonia stripping, VFA production, PHA production and extraction, composting, micro-algae cultivation	0.30	Spain	This study
No treatment	Open lagoon storage (6 months), transport, application on the land	1.95	Spain	[52]
	Closed storage, GPM separation, ammonium sulfate and treated manure transported and applicated on land	2.29	Spain	[52]
AD	AD, biogas combustion, S/L separation, composting and application	1.40	Brazil	[60]
PHA production	OFMSW use for PHA production, with composting, PHA extraction with chloroform	2.37	Italy	[30]
	OFMSW use for PHA production, with composting, PHA extraction with ethyl acetate	1.64	Italy	[30]

Marine eutrophication (ME)

The bioplastic generation impacts the marine eutrophication potential 23 % less compared to the biogas system. The considerably low ME values of both scenarios (seen in Table 5.3) can be the result of the algae cultivation and its nitrogen removal of the processed water. In a study using a similar pond, 6.5×10^{-3} kg N-eq per ton of manure remains when the land application of manure and digestate is excluded (95 % of the total). This small value highlights that microalgae-based nutrient removal from the liquid phase can significantly reduce marine eutrophication impacts and explain the low observed values of this study.

On the other side, the reported value of 1.5×10^{-4} kg N eq. by Girón-Rojas et al. [50], is approximately two orders of magnitude lower marine eutrophication than the values obtained in this study. As this system includes only anaerobic digestion and CHP, no nitrogen leaching is included for digestate handling, explaining this low reported value. Álvaro et al. [4] did show reduced nitrogen leaching during land application of digestate compared to raw manure, indicating positive effects for composting later in the process.

Table 5.3: Comparison of marine eutrophication (ME) impacts expressed in kg N eq. per ton of treated substrate. Differences between studies are mainly explained by system boundaries, particularly the inclusion of land application, digestate management, and composting processes.

Process category	Process description	ME [kg N eq.]	Location	Source
Biogas production	AD, biogas combustion, solid/liquid separation, composting, and microalgae cultivation	0.019	Spain	This study
Bio-plastic production	Ammonia stripping, VFA production, PHA production and extraction, composting, micro-algae cultivation	0.015	Spain	This study
No treatment	Open storage (45 days), transport, application on the land	0.28	Spain	[4]
	Covered storage (45 days), transport, application on the land	0.28	Spain	[4]
AD	Anaerobic digestion, biogas combustion, digestate handling, and land application	0.21	Spain	[4]
	AD with CHP (no composting or digestate management)	1.5×10^{-4}	Spain	[50]
	AD with biomethane upgrading combined with micro-algae cultivation, composting and application on land	0.13	–	[101]

Freshwater eutrophication (FE)

The freshwater eutrophication potential performs significantly better (41 times) in the biogas scenario compared to the bioplastic, as the biogas scenario has a negative value of - 8.3 $\times 10^{-4}$ compared to 3.3 $\times 10^{-2}$, resulting in a difference of 3.4 $\times 10^{-2}$. With the negative value, the biogas production process even has a positive effect on the freshwater eutrophication, as it avoids more P equivalents than it produces. The effect of the pond is also observed by Martínez-Hernando et al. [101], who report a considerable low impact of 6.0×10^{-4} kg P-eq per ton of manure, with the largest attribution related to manure and digestate application.

The higher values of other studies shown in Table 5.4, are mainly attributed to the inclusion of land application of fertilizers within the system boundaries, as application of fertilizers cause leaching of P into the soil [50, 60, 59] . The higher FE impact of bioplastic is caused by the background processed

of the electricity use [29]. As can be seen in Table 5.4, the bioplastic scenario perform only better than the treatment of OFMSW to PHA [30].

Table 5.4: Comparison of freshwater eutrophication (FWE) impacts expressed in kg P eq. per ton of treated substrate.

Process category	Process description	FE [kg P eq.]	Location	Source
Biogas production	AD, biogas combustion, solid/liquid separation, composting, and microalgae cultivation	- 8.3×10^{-4}	Spain	This study
Bio-plastic production	Ammonia stripping, VFA production, PHA production and extraction, composting, microalgae cultivation	3.3×10^{-2}	Spain	This study
No treatment	Open and closed storage (45 days), transport, application on the land	4.23×10^{-4}	Spain	[4]
AD	Anaerobic digestion with land application of digestate	4.17×10^{-4}	Spain	[4]
	AD, composting and land application	5×10^{-3}	Brazil	[60]
	Biomethane upgrading combined with microalgae cultivation, composting and application on land	6.03×10^{-4}	Spain	[101]
Membrane treatment	GPM system with fertilizer substitution	-4.35×10^{-3}	Spain	[52]
PHA production	OFMSW treatment without PHA production	0.73	Italy	[30]
	OFMSW with PHA (chloroform extraction)	1.70	Italy	[30]
	OFMSW with PHA (ethyl acetate extraction)	1.08	Italy	[30]

Fine particulate matter formation (FPMF)

Fine particulate matter formation (FPMF) is mainly caused by air emissions of NH_3 in this process. The total impact is 5 times larger in the bioplastic scenario compared to the biogas produciton. As shown in Figures 4.8 and 4.9, these emissions primarily originate from foreground processes such as anaerobic digestion and composting, as well as background processes related to chemical and electricity production. FPMF is influenced by NO_x emissions associated with electricity use, which is also significant for chemicals that require highly energy-intensive production processes

In Table 5.5, a large difference is observed in reference studies. The storage time of the no treatment scenarios largely impacts the comparability to his study, as this also influences the results fo the AD and membrane processes. The digestate storage does not influence the results of the biogas scenario, as no NH_3 were concluded, making it hard tto compare tthe values.

The effect of AD on the FPMF waas not observed by González-García et al. [52], as no NH_3 emission were reported, which is against general expectations of anaerobic digestion.

What can be concluded is that biological extraction of PHA showed 9 times lower impact 9 times lower than the bioplastic scenario, indicating that novel PHA extraction methods may lead to improved environmental performance with respect to FPMF. [96]

Table 5.5: Comparison of fine particulate matter formation (FPMF) impacts expressed in kg $PM_{2.5}$-eq per ton of treated substrate.

Process category	Process description	FPMF [kg $PM_{2.5}$ eq.]	Location	Source
Biogas production	AD, biogas combustion, solid/liquid separation, composting, and microalgae cultivation	2.5 $\times 10^{-2}$	Spain	This study
Bio-plastic production	Ammonia stripping, VFA production, PHA production and extraction, composting, microalgae cultivation	1.1 $\times 10^{-1}$	Spain	This study
No treatment	Open storage (45 days), transport, application on the land	2.50×10^{-5}	Spain	[4]
	Covered storage (45 days), transport, application on the land	2.41×10^{-5}	Spain	[4]
	Open lagoon storage (6 months), transport, application on the land	2.3 $\times 10^{-1}$	Spain	[52]
AD	Anaerobic digestion with storage and land application	2.50×10^{-5}	Spain	[4]
Membrane separation	Gas-permeable membrane treatment, storage and land application	2.7 $\times 10^{-1}$	Spain	[52]
Biological PHA extraction	Sterilization, PHA production, S/L separation and biological PHA extraction using mealworms	1.3 $\times 10^{-2}$	Malaysia	[96]

5.2. Economic evaluation

Both processes seem economically unfeasible.

For the biogas production plant, a similar scale for swine treatment with AD has been found, that also concludes a economically unfeasible process [50]. Only in collaboration with other farms a economically feasible process could be obtained. Why other biogas production plants are economically feasible in Europe, is first explained by economies of scale. Larger biogas facilities are significantly more profitable, as Hollas et al. [60] concludes that a mono-digestion biogas plant of swine manure should have a capacity of at least 740 kWe, in Brazil. The capacity of this study is 5 kWe for the total biogas production[131]. If the yield of the biogas production would be maximal, a electrical output of 12 kWe could be obtained, still around 60 times too small, if the same assumptions could be done as this study uses for a plant in Brazil, which is most likely even less favorable in Spain. Studies in Europe also explain that, small-scale or on-farm digesters often face prohibitive initial capital investments and typically require external financial support to be viable [132, 150] Also, digesting animal manure alone (mono-digestion) usually yields limited economic returns due to its low biogas potential [132, 35]. Adding highly degradable co-substrates with a high carbon content, like vegetable waste, the biogas yields can be boosted [50]. Co-digestion can produce 70% more electricity and up to 5.5 times higher economic benefits compared to mono-digestion [59, 50]. However, this is only feasible if the costs of acquiring these co-substrates are kept low [35].

An other option to obtain a economically feasible biogas plant, upgrading of the gas to a high-purity biomethane, can be considered. This can be used for vehicle transport and has a higher-value on the market, 410 EUR/L for for quantities of 1L, compared to 0.96 EUR/m3 for biogas [138] . The price should be handled with care, as this is the price for laboratory use. [101, 116].

For the bioplastic production pathway, economical infeasibility was even clearer shown. Yield improvements did not necessarily lead to better economic performance. Although higher PHA yields increase product output, they are accompanied by higher operational costs (OPEX), particularly due to increased demand for energy and chemicals during downstream processing. Ventura and Ventura [153] indicate that improved process efficiencies can reduce raw material requirements per unit of product, which may partially offset OPEX, but this effect appears limited in the present system.

Based on large scale PHA plants, the capital expenditure (CAPEX) of the bioplastic system was observed high, but given the amount of reactors and complexity, the CAPEX is relatively moderate. It is the operational costs per unit of PHA produced, that significantly impact the results.

A key limitation of the system studied is its small production scale. Large-scale projects based on municipal wastewater and mixed microbial cultures, such as the European PHARIO and WOW! initiatives, indicate that a minimum production capacity of approximately 5,000 tons of PHA per year is required for commercial viability [83]. In contrast, the system analyzed in this study produces only 25 tons per year, which is several orders of magnitude lower.

The importance of scale is further supported by Mudliar et al. [110], who show that increasing yields of PHA production from 40% to 70% can make systems with capacities of around 50 m^3/day economically feasible. However, the capacity considered in this study (26 m^3/day) remains below this threshold, suggesting that yield improvements alone are insufficient to achieve economic viability.

Overall, plant capacity has a dominant influence on the unit cost of PHA production. Larger-scale facilities benefit from economies of scale, as capital investment and labor costs are distributed over a greater production volume, thereby reducing the minimum selling price per kilogram of PHA [110, 147, 153]. In addition, labor requirements tend to increase less than proportionally with scale, further improving economic performance at larger plant sizes.

Besides this, the use of different substrates would lead to easier to manage production. The PHA extraction method is for example now chosen for food waste substrate, but using manure a substrate can lead to even higher operational cost. For this, sulfide gasses and heavy metals should be taken into account, to calculate the cost for extra operational steps to use manure as a feedstock. This is not incorporated in this study.

As is highlighted before, VFA's are of its own value and can be used in multiple production processes as precursor (section 2.2.8). In the hypothetical scenario that the VFA mixture would be pure acetic acid, the process could become economically better. However, given assumptions and calculations stated in Table D.5 in section D, the operating cost of this would be 4.95 EUR/kg, which is not feasible given the selling price of 0.79 EUR/kg for pure acetic acid [65].

6 Conclusion

By answering the main question, the conclusion of this study is drawn:

What treatment strategy for swine manure performs best in terms of environmental impact reduction, nutrient recycling, and economic feasibility in a farm-scale plant?

The subquestions were (i) how treatment process steps influence the overall performance of a treatment, (ii) which of the two scenarios performs best across all defined criteria and at last (iii) how do improvements in key process yields affect the overall system performance on all criteria.

Anaerobic digestion is considered a suitable treatment for swine manure, leading to lower emissions during storage. The composting step strongly influences environmental impacts, but compost is also a significant revenue stream and a slow-release fertilizer. Without composting, the mass of the digestate would be approximately twice as large, containing around 80% more water. This would increase transport requirements and associated impacts. It would also result in lower nutrient concentrations, making the product less attractive for sale. The microalgae pond has a positive effect on eutrophication impacts.

The energy demand of both the biogas and bioplastic scenarios strongly influences environmental and economic results. Electricity demand remains a critical uncertainty and should be further investigated to accurately assess energy requirements for both processes. The use of sustainable energy would improve the comparison between the two processes. The chemical use in PHA production also strongly influences the process, and as more sustainable methods are developed, these effects may significantly change the results.

In comparison of both scenarios, under the assumptions made in this study, nutrient recoveries of 50 and 55% are observed for biogas and bioplastic production for carbon, 38 and 86% for nitrogen, 93 and 99% for phosphorus, and 10 and 14% for potassium recovery. Bioplastic production performs better, specifically for nitrogen recovery, as a result of the ammonia stripping column. A nitrogen loss of 29 and 12% of the total is observed in the cultivation pond due to mass balance calculations based on phosphorus limitation. In terms of circularity, phosphorus and nitrogen (in the bioplastic scenario) are retained very well, carbon is moderately retained, and potassium is poorly retained.

From an environmental perspective, the biogas scenario impact is approximately 5 times lower in global warming potential, 41 times in freshwater eutrophication and 5 times fine particulate matter formation than the bioplastic scenario. The biogas scenario performs also better compared to no treatment of manure. Only for terrestrial acidification and marine eutrophication does the bioplastic production have a lower negative impact. This is mostly caused by NH_3 retention in ammonium sulfate. The results are largely affected by the assumptions made and the system boundaries defined. To improve precision in future assessments, the composting and AD emission factors, the application of fertilizer on land, and the electricity demand should be further investigated.

The economic feasibility of the biogas production is, under optimistic conditions, better than that of bioplastic production, but still insufficient to achieve a positive net present value. Labor demands of the PHA production have a substantial effect on the outcome of the bioplastic production scenario. Economies of scale should be applied to both processes to achieve economic feasibility, roughly 31 times or 18 times more product (under improved yields) is needed for the biogas and bioplastic scenario to outplay the operational cost, indicating that both processes are structurally under-scaled.

Yield improvements do not strongly affect nutrient recovery (5–9% for carbon as the largest increase). The environmental effect of biogas production improves(between 4–122%), whereas the effect on the bioplastic scenario worsens(between 19–28%). This is mainly caused by energy demands. Yield improvements lead to higher revenues (57% and 29%), but these increases are still insufficient to significantly improve the economic performance. Additionally, increased raw material requirements are observed in the bioplastic scenario.

Overall, the biogas production scenario outperforms the bioplastic pathway environmentally across most impact categories and is closer to economic viability, while the bioplastic pathway provides superior nitrogen recovery. Neither process is economically feasible at the farm scale of 8,500 t/yr considered.

6.1. Recommendations

For the recommendations of this study, a distinction is made between (i) improvements to the quality of this study, (ii) further developments of the model, and (iii) improvements to the realistic treatment of swine manure.

Recommendations to improve the quality of this study include, first, applying a sensitivity analysis to the composition of the manure. This could provide further insight into which biodegradable fractions impact the outcome, in mass of end products and mass of emissions. The variability of manure has been identified as being strongly influenced by factors such as nutrition, climate, and location. By testing the variations of the nutrients in the manure, analysis could be made on the effects of specific contents. For the environmental impact assessment, it is recommended to extend the system boundaries to include land application and potentially even the end-of-life stage of bioplastics, in order to obtain a more complete picture. Additionally, more impact categories, particularly human toxicity and eco-toxicity, could be included to assess if any unaddressed impact categories might change the picture for the bioplastic production scenario. For the bioplastic scenario, multiple downstream processing methods should be considered to evaluate how technological advances in PHA extraction influence environmental impacts. Furthermore, emission factors for composting should preferably be measured for more accurately estimations.

Further modeling steps could include expanding the current model by incorporating additional technologies and varying yields based on different locations in Europe. A model could be developed that uses different manure characteristics as input and provides outputs on environmental impact and economic performance. Economic parameters could be integrated into the Python model to directly assess feasibility. However, due to the generalized assumptions and background data used in SimaPro, a fully integrated LCA within such a model may not be feasible. Instead, the model could generate specific input parameters for SimaPro, allowing users input this in SimaPro. If successful, the model could be extended to other types of manure, such as cattle or poultry manure. This tool should be developed with a focus on European conditions and have a clear output on needed scale for economic feasibility. Also, the focus on bioplastic production should be shifted towards different end products, as this process has shown multiple limitations.

Finally, alternative process configurations should be investigated to improve overall yields. These may include co-digestion, the use of algae for biogas upgrading to high-purity biomethane and PHA co-digestion during acidogenesis. The main focus of future studies should be on increasing yields in the biogas production process, to produce more valuable products, to avoid more fossil fuels and generate sufficient revenue to offset operational costs.

References

[1] Acrres Wageningen UR. *AlgaEconomics bio economic production models of microalgae*. Tech. rep. Wageningen University and Research, 2018. URL: https://www.acrres.nl/wp-content/uploads/2018/03/3e5c6f85-d5bc-431d-ae25-85d949b327b0_WP2A7.10-report-Business-economics-microalgae-and-DSP.pdf.

[2] Teodorita Al Seadi et al. "Biogas digestate quality and utilization". In: *The Biogas Handbook*. Cambridge, UK: Woodhead Publishing Limited, 2013. Chap. 12, pp. 267–301. DOI: 10.1533/9780857097415.2.267.

[3] Ana Álvarez-González et al. "Environmental and economic benefits of using microalgae grown in wastewater as biofertilizer for lettuce cultivation". In: *Bioresource Technology* 424 (Feb. 2025), p. 132230. DOI: 10.1016/j.biortech.2025.132230. URL: https://doi.org/10.1016/j.biortech.2025.132230.

[4] Alfonso García Álvaro et al. "Life cycle assessment of swine manure management: A comparison of different management systems with Montecarlo simulation". In: *Journal of Cleaner Production* 512 (Mar. 2025), p. 145368. DOI: 10.1016/j.jclepro.2025.145368. URL: https://doi.org/10.1016/j.jclepro.2025.145368.

[5] Ayesha Algade Amadu et al. "A review of biopolymer (Poly-β-hydroxybutyrate) synthesis in microbes cultivated on wastewater". In: *The Science of The Total Environment* 756 (Nov. 2020), p. 143729. DOI: 10.1016/j.scitotenv.2020.143729. URL: https://doi.org/10.1016/j.scitotenv.2020.143729.

[6] Mujidat O. Aremu, Mofoluwake M. Ishola, and Mohammad J. Taherzadeh. "Polyhydroxyalkanoates (PHAs) Production from Volatile Fatty Acids (VFAs) from Organic Wastes by Pseudomonas oleovorans". In: *Fermentation* 7.4 (Nov. 2021), p. 287. DOI: 10.3390/fermentation7040287. URL: https://doi.org/10.3390/fermentation7040287.

[7] Arkema. *Global product strategy safety summary for hydrogen peroxide and sodium hypochlorite*. Tech. rep. Arkema, 2013. URL: https://www.arkema.com/files/live/sites/shared_arkema/files/downloads/socialresponsability/safety-summuries/hydrogen-peroxide-sodium-hypochlorite-gps-2013-02-10-v0.pdf.

[8] Merve Atasoy et al. "Bio-based volatile fatty acid production and recovery from waste streams: Current status and future challenges". In: *Bioresource Technology* 268 (July 2018), pp. 773–786. DOI: 10.1016/j.biortech.2018.07.042. URL: https://doi.org/10.1016/j.biortech.2018.07.042.

[9] Giulia Atzori et al. "Algae and Bioguano as promising source of organic fertilizers". In: *Journal of Applied Phycology* 32.6 (Oct. 2020), pp. 3971–3981. DOI: 10.1007/s10811-020-02261-7. URL: https://doi.org/10.1007/s10811-020-02261-7.

[10] PRé Sustainability B.V. *Life Cycle Assessment (LCA) glossary - SimaPro*. Jan. 2026. URL: https://simapro.com/insights/life-cycle-assessment-lca-glossary/.

[11] BIOBOX Water. *Industrial wastewater regulations in spain*. 2024. URL: https://biobox-water.com/en/news/industrial-wastewater-regulations-in-spain/.

[12] Atanu Biswas, Huai N. Cheng, and John C. Edwards. "Poly(3-Hydroxybutyrate-Co-3-Hydroxyvalerate) Produced from Food-Related Wastes: Solid-State NMR Analysis". In: *Macromol—A Journal of Macromolecular Research* 5.4 (Dec. 2025), p. 61. DOI: 10.3390/macromol5040061. URL: https://doi.org/10.3390/macromol5040061.

[13] Francesco Borda et al. "Environmental performance of PET and biopolymers: a comparative LCA of end-of-life scenarios". In: *The International Journal of Advanced Manufacturing Technology* 142.5-6 (Jan. 2026), pp. 2599–2613. DOI: 10.1007/s00170-025-17057-7. URL: https://doi.org/10.1007/s00170-025-17057-7.

[14] Anca Bordianu et al. “Impact of Polypropylene Sutures Phisical Properties on Lymphaticovenous Anastomosis in Lymphedema Patients”. In: *Materiale Plastice* 61.1 (2024), pp. 111–117. DOI: 10.37358/MP.24.1.5707. URL: https://revmaterialeplastice.ro/pdf/10%20BORDIANU%201%2024.pdf.

[15] Claudio Brienza et al. *Final report on mass and energy balances, product composition and quality and overall technical performance of the demonstration plants*. Deliverable D1.5 D1.5. Horizon 2020 Grant Agreement No. 730400. SYSTEMIC Project, 2022. URL: https://systemicproject.eu/.

[16] T. Cai, S. Y. Park, and Y. Li. “Nutrient removal from wastewater by integrated microalgae-based biofuel production—A review”. In: *Renewable and Sustainable Energy Reviews* 19 (2013), pp. 356–369. DOI: 10.1016/j.rser.2012.11.030.

[17] Qitao Cao et al. “Revealing mechanism of micro-aeration for enhancing volatile fatty acids production from swine manure”. In: *Bioresource Technology* 365 (Oct. 2022), p. 128140. DOI: 10.1016/j.biortech.2022.128140. URL: https://doi.org/10.1016/j.biortech.2022.128140.

[18] ChemAnalyst. *Polydimethylsiloxane antifoam emulsion pricing data and market analysis*. 2026. URL: https://www.chemanalyst.com/Pricing-data/polydimethylsiloxane-antifoam-emulsion-3050.

[19] Zhihui Chen et al. “Exploration of Microalgal Species for Nutrient Removal from Anaerobically Digested Swine Wastewater and Potential Lipids Production”. In: *Microorganisms* 9.12 (Nov. 2021), p. 2469. DOI: 10.3390/microorganisms9122469. URL: https://doi.org/10.3390/microorganisms9122469.

[20] Hong L. Choi, Sartika I.A. Sudiarto, and Anriansyah Renggaman. “Prediction of livestock manure and mixture higher heating value based on fundamental analysis”. In: *Fuel* 116 (Sept. 2013), pp. 772–780. DOI: 10.1016/j.fuel.2013.08.064. URL: https://doi.org/10.1016/j.fuel.2013.08.064.

[21] Qingnan Chu et al. “Hydrothermal carbonization of microalgae for phosphorus recycling from wastewater to crop-soil systems as slow-release fertilizers”. In: *Journal of Cleaner Production* 283 (Oct. 2020), p. 124627. DOI: 10.1016/j.jclepro.2020.124627. URL: https://doi.org/10.1016/j.jclepro.2020.124627.

[22] Erik R. Coats, Benjamin S. Watson, and Cynthia K. Brinkman. “Polyhydroxyalkanoate synthesis by mixed microbial consortia cultured on fermented dairy manure: Effect of aeration on process rates/yields and the associated microbial ecology”. In: *Water Research* 106 (Sept. 2016), pp. 26–40. DOI: 10.1016/j.watres.2016.09.039. URL: https://doi.org/10.1016/j.watres.2016.09.039.

[23] COGERSA (Compañía para la Gestión de Residuos Sólidos en Asturias, S.A.U.) *Venta de compost: Composición, características y usos*. Accessed: 2026-03-26. 2026. URL: https://cogersa.es/recuperacion-de-residuos/venta_compost/.

[24] Joeri Coppens et al. “The use of microalgae as a high-value organic slow-release fertilizer results in tomatoes with increased carotenoid and sugar levels”. In: *Journal of Applied Phycology* 28.4 (Dec. 2015), pp. 2367–2377. DOI: 10.1007/s10811-015-0775-2. URL: https://doi.org/10.1007/s10811-015-0775-2.

[25] Laiza Santos Dagnaisser et al. “Microalgae as Bio-fertilizer: a New Strategy for Advancing Modern Agriculture, Wastewater Bioremediation, and Atmospheric Carbon Mitigation”. In: *Water Air & Soil Pollution* 233.11 (Nov. 2022). DOI: 10.1007/s11270-022-05917-x. URL: https://doi.org/10.1007/s11270-022-05917-x.

[26] Alexia Saleme Aona De Paula Pereira et al. “Microalgae organomineral fertilizer production: A life cycle approach”. In: *Algal Research* 71 (Feb. 2023), p. 103035. DOI: 10.1016/j.algal.2023.103035. URL: https://doi.org/10.1016/j.algal.2023.103035.

[27] Sanne M. De Smit et al. “Continuous n-valerate formation from propionate and methanol in an anaerobic chain elongation open-culture bioreactor”. In: *Biotechnology for Biofuels* 12.1 (May 2019), p. 132. DOI: 10.1186/s13068-019-1468-x. URL: https://doi.org/10.1186/s13068-019-1468-x.

[28] J.W. De Vries et al. “Comparing environmental consequences of anaerobic mono- and co-digestion of pig manure to produce bio-energy – A life cycle perspective”. In: *Bioresource Technology* 125 (Sept. 2012), pp. 239–248. DOI: 10.1016/j.biortech.2012.08.124. URL: https://doi.org/10.1016/j.biortech.2012.08.124.

[29] M. Saavedra Del Oso, M. Mauricio-Iglesias, and A. Hospido. “Evaluation and optimization of the environmental performance of PHA downstream processing”. In: *Chemical Engineering Journal* 412 (Nov. 2020), p. 127687. DOI: 10.1016/j.cej.2020.127687. URL: https://doi.org/10.1016/j.cej.2020.127687.

[30] Mateo Saavedra Del Oso et al. “Prospective LCA to provide environmental guidance for developing waste-to-PHA biorefineries”. In: *Journal of Cleaner Production* 383 (Nov. 2022), p. 135331. DOI: 10.1016/j.jclepro.2022.135331. URL: https://doi.org/10.1016/j.jclepro.2022.135331.

[31] Liangwei Deng et al. “Effect of temperature on continuous dry fermentation of swine manure”. In: *Journal of Environmental Management* 177 (Apr. 2016), pp. 247–252. DOI: 10.1016/j.jenvman.2016.04.029. URL: https://doi.org/10.1016/j.jenvman.2016.04.029.

[32] Na Duan et al. “Life cycle assessment of anaerobic digestion of pig manure coupled with different digestate treatment technologies”. In: *Environment International* 137 (Jan. 2020), p. 105522. DOI: 10.1016/j.envint.2020.105522. URL: https://doi.org/10.1016/j.envint.2020.105522.

[33] ECHEMI. *Ammonium sulfate Granular Caprolactam Northwest Europe FOB Price Curve and Change*. Accessed: 2026-03-27. ECHEMI Price Database. 2026. URL: https://www.echemi.com/price-curve/northwest-europe-fob-granular-caprolactam-temppid160705011796-2.html.

[34] Lars Elsgaard, Anne B. Olsen, and Søren O. Petersen. “Temperature response of methane production in liquid manures and co-digestates”. In: *The Science of The Total Environment* 539 (Sept. 2015), pp. 78–84. DOI: 10.1016/j.scitotenv.2015.07.145. URL: https://doi.org/10.1016/j.scitotenv.2015.07.145.

[35] Pedro Esperanço, António Ferreira, and José Ferreira. “A Comparative Systematic Review of Life-Cycle Assessments of Treatment Strategies for Swine Slurry with a Focus on Anaerobic Co-Digestion”. In: *Processes* 14.3 (Feb. 2026), p. 573. DOI: 10.3390/pr14030573. URL: https://doi.org/10.3390/pr14030573.

[36] European Commission. *Clean air and agriculture in the EU*. Last updated information refers to CAP 2023-27, relevant content published 26 June 2023. European Commission, Directorate-General for Agriculture and Rural Development. 2023. URL: https://agriculture.ec.europa.eu/cap-my-country/sustainability/environmental-sustainability/natural-resources/clean-air%5C_en.

[37] European Commission. *Commission Delegated Regulation (EU) 2023/1185 of 10 February 2023 supplementing Directive (EU) 2018/2001 of the European Parliament and of the Council by establishing a minimum threshold for greenhouse gas emissions savings of recycled carbon fuels and by specifying a methodology for assessing greenhouse gas emissions savings from renewable liquid and gaseous transport fuels of non biological origin and from recycled carbon fuels*. 2023. URL: https://eur-lex.europa.eu/eli/reg_del/2023/1185/oj/eng.

[38] European Commission. *CORDIS Project Fact Sheet: CIRCULAR BIOCARBON (Turning carbon of complex organic urban waste streams into value-added products)*. Accessed: 11 April 2026. 2021. URL: https://cordis.europa.eu/project/id/101023280.

[39] European Commission. *Fertiliser prices*. 2026. URL: https://agridata.ec.europa.eu/extensions/DashboardFertiliser/FertiliserPrices.html.

[40] European Commission. *Processing of livestock waste, for the production of biomethane for use in agricultural vehicles and biofertilizers*. Accessed: 11 April 2026. 2024. URL: https://webgate.ec.europa.eu/life/publicWebsite/project/LIFE19-CCM-ES-001206/processing-of-livestock-waste-for-the-production-of-biomethane-for-use-in-agricultural-vehicles-and-biofertilizers.

[41] European Environment Agency (EEA). *Greenhouse gas emissions from agriculture in Europe*. Published 31 Oct 2024; Accessed: 31 October 2025. 2024. URL: https://www.eea.europa.eu/en/analysis/indicators/greenhouse-gas-emissions-from-agriculture.

[42] European Parliament Research Service (EPRS). *Greenhouse gas emissions from agriculture: A problem for the environment, farmers and policymakers*. European Parliament, Briefing Document. 2018. URL: https://www.europarl.europa.eu/pdfs/news/expert/2018/3/story/20180301STO98928/20180301STO98928_en.pdf (visited on 10/30/2025).

[43] Eurostat. *Electricity price statistics*. Accessed: 11 April 2026. 2025. URL: https://ec.europa.eu/eurostat/statistics-explained/index.php?title=Electricity_price_statistics#Electricity_prices_for_non-household_consumers.

[44] Eurostat. *Value of agriculture output down by 0.9% in 2024*. Accessed: April 20, 2026. 2025. URL: https://ec.europa.eu/eurostat/web/products-eurostat-news/w/ddn-20251107-1.

[45] José Ferreira et al. "Environmental Assessment of Pig Manure Treatment Systems through Life Cycle Assessment: A Mini-Review". In: *Sustainability* 16.9 (Apr. 2024), p. 3521. DOI: 10.3390/su16093521. URL: https://doi.org/10.3390/su16093521.

[46] Nicola Frison et al. "Effects of the Sludge Retention Time and Carbon Source on Polyhydroxyalkanoate-Storing Biomass Selection under Aerobic-Feast and Anoxic-Famine Conditions". In: *ACS Sustainable Chemistry & Engineering* 9.28 (July 2021), pp. 9455–9464. DOI: 10.1021/acssuschemeng.1c02973. URL: https://doi.org/10.1021/acssuschemeng.1c02973.

[47] Maria Cruz García-González et al. "Treatment of swine manure: case studies in European's N-surplus areas". In: *Scientia Agricola* 73.5 (Oct. 2016), pp. 444–454. DOI: 10.1590/0103-9016-2015-0057. URL: https://doi.org/10.1590/0103-9016-2015-0057.

[48] Generalitat de Catalunya. *Llei 2/2023, del 16 de marc, de pressupostos de la generalitat de catalunya per al 2023*. Art. 42. 2023. URL: https://aplicacions.economia.gencat.cat/wpres/AppPHP/2023/pdf/VOL_L_ART.pdf.

[49] Luis Getino et al. "The Effectiveness of Polyhydroxyalkanoate (PHA) Extraction Methods in Gram-Negative Pseudomonas putida U". In: *Polymers* 17.2 (Jan. 2025), p. 150. DOI: 10.3390/polym17020150. URL: https://doi.org/10.3390/polym17020150.

[50] Cecilia Girón-Rojas et al. "Enhancing Energy Self-Sufficiency in rural regions: Environmental and Financial assessment of anaerobic digestion of pig slurry." In: *Waste and Biomass Valorization* (Oct. 2025). DOI: 10.1007/s12649-025-03366-1. URL: https://doi.org/10.1007/s12649-025-03366-1.

[51] Cristina González-Fernández, Beatriz Molinuevo-Salces, and Maria Cruz García-González. "Evaluation of anaerobic codigestion of microalgal biomass and swine manure via response surface methodology". In: *Applied Energy* 88.10 (2011), pp. 3448–3453. DOI: 10.1016/j.apenergy.2010.12.035.

[52] I. González-García et al. "Environmental sustainability performance of a membrane-based technology for livestock wastewater treatment with nutrient recovery". In: *Journal of environmental chemical engineering* 10.2 (Jan. 2022), p. 107246. DOI: 10.1016/j.jece.2022.107246. URL: https://doi.org/10.1016/j.jece.2022.107246.

[53] Google Finance. *Historical Exchange Rate: USD to EUR (April 1, 2026)*. Alphabet Inc. Accessed: 2026-04-17. 2025. URL: https://www.google.com/finance/quote/USD-EUR.

[54] Nicholas M. Guho et al. "Pilot-scale production of poly-3-hydroxybutyrate-co-3-hydroxyvalerate from fermented dairy manure: Process performance, polymer characterization, and scale-up implications". In: *Bioresource Technology Reports* 12 (Oct. 2020), p. 100588. DOI: 10.1016/j.biteb.2020.100588. URL: https://doi.org/10.1016/j.biteb.2020.100588.

[55] Donna Post Guillen et al. "An Eco-Friendly System for the Production of Value-Added Materials from Dairy Manure". In: *JOM* 70.10 (July 2018), pp. 1946–1957. DOI: 10.1007/s11837-018-2995-9. URL: https://doi.org/10.1007/s11837-018-2995-9.

[56] Helian Polymers B.V. *PB3430 P34HB (15% 4 HB) powder*. Accessed: 2026-03-27. 2026. URL: https://shop.helianpolymers.com/products/pb3430-p34hb-15-4-hb-powder.

[57] Dolores Hidalgo et al. “Targeted acidogenic fermentation of waste streams for the selective production of volatile fatty acids as bioplastic precursors”. In: *Applied Sciences* 15.11 (May 2025), p. 5923. DOI: 10.3390/app15115923. URL: https://doi.org/10.3390/app15115923.

[58] M. Hjorth et al. “Solid—liquid separation of animal slurry in theory and practice. A review”. In: *Agronomy for Sustainable Development* 30.1 (June 2009), pp. 153–180. DOI: 10.1051/agro/2009010. URL: https://doi.org/10.1051/agro/2009010.

[59] C.E. Hollas et al. “Swine manure treatment technologies as drivers for circular economy in agribusiness: A techno-economic and life cycle assessment approach”. In: *The Science of The Total Environment* 857 (Oct. 2022), p. 159494. DOI: 10.1016/j.scitotenv.2022.159494. URL: https://doi.org/10.1016/j.scitotenv.2022.159494.

[60] Camila Ester Hollas et al. “Life cycle assessment of waste management from the Brazilian pig chain residues in two perspectives: Electricity and biomethane production”. In: *Journal of Cleaner Production* 354 (Apr. 2022), p. 131654. DOI: 10.1016/j.jclepro.2022.131654. URL: https://doi.org/10.1016/j.jclepro.2022.131654.

[61] Yuansheng Hu et al. “Filamentous microalgae as an advantageous co-substrate for enhanced methane production and digestate dewaterability in anaerobic co-digestion of pig manure”. In: *Waste Management* 119 (2021), pp. 399–407. DOI: 10.1016/j.wasman.2020.10.041.

[62] Weiwei Huang et al. “Volatile fatty acids (VFAs) production from swine manure through short-term dry anaerobic digestion and its separation from nitrogen and phosphorus resources in the digestate”. In: *Water Research* 90 (2016), pp. 344–353. DOI: 10.1016/j.watres.2015.12.044.

[63] M.A.J. Huijbregts et al. *ReCiPe 2016: A harmonized life cycle impact assessment method at midpoint and endpoint level*. Tech. rep. 2016, pp. 2–191. URL: https://www.rivm.nl/bibliotheek/rapporten/2016-0104.pdf.

[64] IFOAM EU and FiBL (Research Institute of Organic Agriculture). *Organic Farming, Climate Change Mitigation and Beyond*. Policy Report. 2016. URL: https://www.organicseurope.bio/content/uploads/2020/06/ifoameu_advocacy_climate_change_report_2016.pdf.

[65] IMARC Group. *Acetic acid pricing report: Price analysis and forecast*. Accessed: April 22, 2026. 2026. URL: https://www.imarcgroup.com/acetic-acid-pricing-report.

[66] IMARC Group. *Ammonium sulfate prices, trend, chart, demand, market analysis, news, historical and forecast data report 2026 edition*. 2026. URL: https://www.imarcgroup.com/ammonium-sulfate-pricing-report.

[67] IMARC Group. *Chloroform prices, trend, chart, demand, market analysis, news, historical and forecast data report 2026 edition*. 2026. URL: https://www.imarcgroup.com/chloroform-pricing-report.

[68] IMARC Group. *Polyhydroxyalkanoates (PHA) Prices, Trend, Chart, demand, market Analysis, news, Historical and Forecast Data Report 2026 Edition*. 2026. URL: https://www.imarcgroup.com/polyhydroxyalkanoates-pricing-report.

[69] IMARC Group. *Potassium hydroxide prices, trend, chart, demand, market analysis, news, historical and forecast data report 2026 edition*. 2026. URL: https://www.imarcgroup.com/potassium-hydroxide-pricing-report.

[70] IMARC Group. *Sodium hypochlorite prices, trend, chart, demand, market analysis, news, historical and forecast data report 2026 edition*. 2026. URL: https://www.imarcgroup.com/sodium-hypochlorite-pricing-report.

[71] IMARC Group. *Sulfuric acid prices, trend, chart, demand, market analysis, news, historical and forecast data report 2026 edition*. 2026. URL: https://www.imarcgroup.com/sulfuric-acid-pricing-report.

[72] Heribert Insam, María Gómez-Brandón, and Judith Ascher. “Manure-based biogas fermentation residues – Friend or foe of soil fertility?” In: *Soil Biology and Biochemistry* 84 (Feb. 2015), pp. 1–14. DOI: 10.1016/j.soilbio.2015.02.006. URL: https://doi.org/10.1016/j.soilbio.2015.02.006.

[73] Instituto Nacional de Estadística (INE). *Survey on Water Supply and Sewerage (Year 2022)*. Tech. rep. Official statistical report detailing regional water prices across Spain, including the 1.24 EUR/m3 rate for Castilla y León. Government of Spain, 2022. URL: https://www.ine.es/dyngs/Prensa/en/ESSA2022.pdf.

[74] International Energy Agency (IEA). *Outlook for biogas and biomethane: Prospects for organic growth*. Report. Licence: CC BY 4.0. Paris, France: International Energy Agency (IEA), 2020. URL: https://www.iea.org/reports/outlook-for-biogas-and-biomethane-prospects-for-organic-growth.

[75] International Fertilizer Industry Association. *Fertilizer production and trade statistics*. WWW Document. Accessed: 2026-04-22. 2023. URL: http://ifadata.fertilizer.org/ucSearch.aspx.

[76] IPCC. "Chapter 10: Emissions from Livestock and Manure Management". In: *2019 Refinement to the 2006 IPCC Guidelines for National Greenhouse Gas Inventories*. Ed. by E. Calvo Buendia et al. Vol. 4: Agriculture, Forestry and Other Land Use. Published: IPCC, Switzerland. Geneva, Switzerland: IPCC, 2019. URL: https://www.ipcc-nggip.iges.or.jp/public/2019rf/vol4.html.

[77] IPCC. "Chapter 11: N2O emissions from managed soils, and CO2 emissions from lime and urea application". In: *2019 Refinement to the 2006 IPCC Guidelines for National Greenhouse Gas Inventories*. Ed. by E. Calvo Buendia et al. Geneva, Switzerland: Intergovernmental Panel on Climate Change, 2019. URL: https://www.ipcc-nggip.iges.or.jp/public/2019rf/vol4.html.

[78] IPCC. "Chapter 3: Consistent Representation of Lands". In: *2019 Refinement to the 2006 IPCC Guidelines for National Greenhouse Gas Inventories*. Ed. by E. Calvo Buendia et al. Vol. 4: Agriculture, Forestry and Other Land Use. Accessed: 2026-01-22. Geneva, Switzerland: IPCC, 2019. URL: https://www.ipcc-nggip.iges.or.jp/public/2019rf/pdf/4_Volume4/19R_V4_Ch03_Land%20Representation.pdf.

[79] IPCC. "Precursors and Indirect Emissions". In: *2006 IPCC Guidelines for National Greenhouse Gas Inventories*. Ed. by H. S. Eggleston et al. Vol. 1: General Guidance and Reporting. Prepared by the National Greenhouse Gas Inventories Programme. Hayama, Japan: Institute for Global Environmental Strategies (IGES), 2006. Chap. 7. URL: https://www.ipcc-nggip.iges.or.jp/public/2006gl/pdf/1_Volume1/V1_7_Ch7_Precursors_Indirect.pdf.

[80] P. Jun, M. Gibbs, and K. Gaffney. *CH_4 Emissions from Manure Management*. Background Paper. Good Practice Guidance and Uncertainty Management in National Greenhouse Gas Inventories. IPCC, 2000. URL: https://www.ipcc-nggip.iges.or.jp/public/gp/bgp/4_3_CH4_Animal_Manure.pdf.

[81] Niels Jungbluth et al. *Life Cycle Inventories of Bioenergy*. Ecoinvent report 17. Dübendorf, Switzerland: Swiss Centre for Life Cycle Inventories, 2007. URL: https://esu-services.ch/fileadmin/download/jungbluth-2007-17_Bioenergy.pdf.

[82] G. J. Kasper and B. Peters. *Monovergisting varkensmest op boerderijschaal*. Dutch. Rapport 632. Lelystad, The Netherlands: Wageningen University
Research, Livestock Research, Aug. 2012. URL: https://edepot.wur.nl/231001.

[83] Mohammed Nazeer Khan et al. *Techno economic assessment of producing bioplastics from sewage*. Tech. rep. VITO and Interreg North West Europe WOW, Nov. 2021. URL: https://vb.nweurope.eu/media/16741/1wpt1_deliverable_3_1_pha_tea_final.pdf.

[84] Robbert Kleerebezem. "Biochemical Conversion". In: *Biomass as a Sustainable Energy Source for the Future*. John Wiley
Sons, Ltd, 2014. Chap. 14, pp. 441–468. ISBN: 9781118916643. DOI: https://doi.org/10.1002/9781118916643.ch14. eprint: https://onlinelibrary.wiley.com/doi/pdf/10.1002/9781118916643.ch14. URL: https://onlinelibrary.wiley.com/doi/abs/10.1002/9781118916643.ch14.

[85] Robbert Kleerebezem. *Extra Advanced Stoichiometry*. Brightspace. PowerPoint slides. Delft University of Technology, Dec. 2024.

[86] Robbert Kleerebezem and Mark C. M. Van Loosdrecht. “A Generalized Method for Thermodynamic State Analysis of Environmental Systems”. In: *Critical Reviews in Environmental Science and Technology* 40.1 (2010), pp. 1–54. DOI: 10.1080/10643380802000974. URL: http://dx.doi.org/10.1080/10643380802000974.

[87] Zygmunt Kowalski, Agnieszka Makara, and Kamil Fijorek. “Changes in the properties of pig manure slurry”. In: *Acta Biochimica Polonica* 60.4 (2013), pp. 845–850. URL: www.actabp.pl.

[88] Jens O. Krömer et al. “Economic Process Evaluation and Environmental Life-Cycle Assessment of Bio-Aromatics Production”. In: *Frontiers in Bioengineering and Biotechnology* 8 (May 2020), p. 403. DOI: 10.3389/fbioe.2020.00403. URL: https://doi.org/10.3389/fbioe.2020.00403.

[89] Thomas Kupper et al. “Ammonia and greenhouse gas emissions from slurry storage - A review”. In: *Agriculture Ecosystems & Environment* 300 (May 2020), p. 106963. DOI: 10.1016/j.agee.2020.106963. URL: https://doi.org/10.1016/j.agee.2020.106963.

[90] R. G. Lehmann et al. “Life Cycle of Silicone Polymer, from Pilot-Scale Composting to Soil Amendment”. In: *Compost Science & Utilization* 7.3 (June 1999), pp. 72–81. DOI: 10.1080/1065657x.1999.10701976. URL: https://doi.org/10.1080/1065657x.1999.10701976.

[91] Ian Levett et al. “Technoeconomic analysis of extreme halophilic manufacture of polyhydroxyalkanoate bioplastics from sugar: Understanding cost sensitivity to feedstock price, fermentation performance and the extraction method”. In: *Bioresource Technology* 445 (Jan. 2026), p. 134078. DOI: 10.1016/j.biortech.2026.134078. URL: https://doi.org/10.1016/j.biortech.2026.134078.

[92] Dan Li et al. “Review of Biopolymer polyhydroxybutyrate (PHB) and blends: Modification of thermal and mechanical properties via additive manufacturing processing”. In: *Polymers* 17.22 (Nov. 2025), p. 3083. DOI: 10.3390/polym17223083. URL: https://doi.org/10.3390/polym17223083.

[93] Ruirui Li et al. “Co-digestion of chicken manure and microalgae Chlorella 1067 grown in the recycled digestate: Nutrients reuse and biogas enhancement”. In: *Waste Management* 70 (Sept. 2017), pp. 247–254. DOI: 10.1016/j.wasman.2017.09.016. URL: https://doi.org/10.1016/j.wasman.2017.09.016.

[94] Wanwu Li et al. “Methane production through anaerobic digestion: Participation and digestion characteristics of cellulose, hemicellulose and lignin”. In: *Applied Energy* 226 (July 2018), pp. 1219–1228. DOI: 10.1016/j.apenergy.2018.05.055. URL: https://doi.org/10.1016/j.apenergy.2018.05.055.

[95] Xiaohui Li, Yujie Wang, Rui Chen, et al. “Environmental impacts of livestock manure management and mitigation strategies”. In: *Science of the Total Environment* 806 (2022), p. 159494. DOI: 10.1016/j.scitotenv.2022.159494.

[96] “Life Cycle Assessment on Global Warming and Fine Particulate Matter Formation for Biological Extraction Method in Polyhydroxyalkanoate Production”. In: *Chemical Engineering Transactions* 113 (2024).

[97] Rong Er Lim et al. “Life cycle assessment on methods for polyhydroxyalkanoate extraction from aerobic granules”. In: *Environmental Science Water Research & Technology* 11.8 (Jan. 2025), pp. 1966–1976. DOI: 10.1039/d5ew00335k. URL: https://doi.org/10.1039/d5ew00335k.

[98] Anna Lymperatou et al. “Improving the Anaerobic Digestion of Swine Manure through an Optimized Ammonia Treatment: Process Performance, Digestate and Techno-Economic Aspects”. In: *Energies* 14.3 (Feb. 2021), p. 787. DOI: 10.3390/en14030787. URL: https://doi.org/10.3390/en14030787.

[99] Mohamed H. Madkour et al. “PHA Recovery from Biomass”. In: *Biomacromolecules* 14.9 (July 2013), pp. 2963–2972. DOI: 10.1021/bm4010244. URL: https://doi.org/10.1021/bm4010244.

[100] Marta Marszałek, Zygmunt Kowalski, and Agnieszka Makara. “Physicochemical and microbiological characteristics of pig slurry”. English. In: *Technical Transactions. Chemistry* 1-Ch (2014). Original title: Czasopismo Techniczne. Chemia, pp. 81–91.

[101] María-Pilar Martínez-Hernando et al. "Sustainability assessment of a novel vehicular biomethane production from swine manure compared to conventional techniques". In: *Waste Management* 209 (Nov. 2025), p. 115218. DOI: 10.1016/j.wasman.2025.115218. URL: https://doi.org/10.1016/j.wasman.2025.115218.

[102] A. McCulloch. "Chloroform in the environment: occurrence, sources, sinks and effects". In: *Chemosphere* 50.10 (Feb. 2003), pp. 1291–1308. DOI: 10.1016/s0045-6535(02)00697-5. URL: https://doi.org/10.1016/s0045-6535(02)00697-5.

[103] Enrique Medina-Martos et al. "Techno-economic and life cycle assessment of an integrated hydrothermal carbonization system for sewage sludge". In: *Journal of Cleaner Production* 277 (July 2020), p. 122930. DOI: 10.1016/j.jclepro.2020.122930. URL: https://doi.org/10.1016/j.jclepro.2020.122930.

[104] Zhang Mengqi et al. "Comprehensive review on agricultural waste utilization and high-temperature fermentation and composting". In: *Biomass Conversion and Biorefinery* 13.7 (Mar. 2021), pp. 5445–5468. DOI: 10.1007/s13399-021-01438-5. URL: https://doi.org/10.1007/s13399-021-01438-5.

[105] Ministerio de Agricultura Alimentacion y Medio Ambiente. *Orden aaa 2056 2014 por la que se aprueban los modelos oficiales de declaracion de vertido*. 2014. URL: https://www.boe.es/eli/es/o/2014/10/27/aaa2056/dof/spa/pdf.

[106] Juliana Montoya. *Green but in the red: Can bioplastics become economically viable?* May 2025. URL: https://www.plasticsengineering.org/2025/05/green-but-in-the-red-can-bioplastics-become-economically-viable-008722/#!.

[107] R. Moral et al. "Estimation of nutrient values of pig slurries in Southeast Spain using easily determined properties". In: *Waste Management* 25.7 (Dec. 2004), pp. 719–725. DOI: 10.1016/j.wasman.2004.09.010. URL: https://doi.org/10.1016/j.wasman.2004.09.010.

[108] Mordor Intelligence. *Algae based animal feed market size, share and 2025 to 2030 outlook*. 2025. URL: https://www.mordorintelligence.com/industry-reports/algae-based-animal-feed-market.

[109] Fernando Morgan-Sagastume et al. "Production of polyhydroxyalkanoates in open, mixed cultures from a waste sludge stream containing high levels of soluble organics, nitrogen and phosphorus". In: *Water Research* 44.18 (June 2010), pp. 5196–5211. DOI: 10.1016/j.watres.2010.06.043. URL: https://doi.org/10.1016/j.watres.2010.06.043.

[110] S. N. Mudliar et al. "Techno-economic evaluation of PHB production from activated sludge". In: *Clean Technologies and Environmental Policy* 10.3 (July 2007), pp. 255–262. DOI: 10.1007/s10098-007-0100-0. URL: https://doi.org/10.1007/s10098-007-0100-0.

[111] Tirthankar Mukherjee, Eric Trably, and Prasad Kaparaju. "Critical Assessment of Hydrogen and Methane Production from 1G and 2G Sugarcane Processing Wastes Using One-Stage and Two-Stage Anaerobic Digestion". In: *Energies* 16.13 (June 2023), p. 4919. DOI: 10.3390/en16134919. URL: https://doi.org/10.3390/en16134919.

[112] Sarah L. Nordahl et al. "Greenhouse Gas and Air Pollutant Emissions from Composting". In: *Environmental Science & Technology* 57.6 (Jan. 2023), pp. 2235–2247. DOI: 10.1021/acs.est.2c05846. URL: https://doi.org/10.1021/acs.est.2c05846.

[113] Isabel Noya et al. "Environmental assessment of the entire pork value chain in Catalonia – A strategy to work towards Circular Economy". In: *The Science of The Total Environment* 589 (Mar. 2017), pp. 122–129. DOI: 10.1016/j.scitotenv.2017.02.186. URL: https://doi.org/10.1016/j.scitotenv.2017.02.186.

[114] OpenAI. *ChatGPT visualization of a pig on a farm integrating nutrient cycling based on user-provided images and prompts (DALL-E 3) [Artificial intelligence model]*. Accessed: April 20, 2026. 2026. URL: https://chat.openai.com.

[115] Isaac Owusu-Agyeman et al. "Conceptual system for sustainable and next-generation wastewater resource recovery facilities". In: *The Science of The Total Environment* 885 (Apr. 2023), p. 163758. DOI: 10.1016/j.scitotenv.2023.163758. URL: https://doi.org/10.1016/j.scitotenv.2023.163758.

[116] César Ruiz Palomar et al. "Pre-Commercial demonstration of a photosynthetic upgrading plant: Investment and operating cost analysis". In: *Processes* 12.12 (Dec. 2024), p. 2794. DOI: 10.3390/pr12122794. URL: https://doi.org/10.3390/pr12122794.

[117] J. B. K. Park and R. J. Craggs. "Wastewater treatment and algal production in high rate algal ponds with carbon dioxide addition". In: *Water Science and Technology* 61.3 (2010), pp. 633–639. DOI: 10.2166/wst.2010.951.

[118] Maxine Passero et al. "Dairy wastewaters for algae cultivation, polyhydroxyalkanote reactor effluent versus anaerobic digester effluent". In: *BioEnergy Research* 8.4 (Apr. 2015), pp. 1647–1660. DOI: 10.1007/s12155-015-9619-9. URL: https://doi.org/10.1007/s12155-015-9619-9.

[119] Joana Carvalho Pereira. *Capital Cost Estimation and Depreciation of Capital: OPEX and Finance*. Presentation slides, LM3731: Design of Sustainable Biotechnological Processes. TU Delft. 2026.

[120] Joana Carvalho Pereira. *Techno Economic Evaluation: CAPEX*. Presentation slides, LM3731: Design of Sustainable Biotechnological Processes. TU Delft. Mar. 2026.

[121] Víctor Pérez et al. "Ectoine Production from Biogas in Waste Treatment Facilities: A Techno-Economic and Sensitivity Analysis". In: *ACS Sustainable Chemistry & Engineering* 9.51 (Dec. 2021), pp. 17371–17380. DOI: 10.1021/acssuschemeng.1c06772. URL: https://doi.org/10.1021/acssuschemeng.1c06772.

[122] Max S. Peters, Klaus D. Timmerhaus, and Ronald E. West. *Plant Design and Economics for Chemical Engineers*. 5th. McGraw-Hill Education, 2003.

[123] Andriamahefasoa Rajaonison et al. "Detailed and simplified parameterized LCA models for early-stage energy projects: A microalgae-based biogas case". In: *Cleaner Environmental Systems* 20 (Dec. 2025), p. 100384. DOI: 10.1016/j.cesys.2025.100384. URL: https://doi.org/10.1016/j.cesys.2025.100384.

[124] T. Rehl and J. Müller. "Life cycle assessment of biogas digestate processing technologies". In: *Resources Conservation and Recycling* 56.1 (Oct. 2011), pp. 92–104. DOI: 10.1016/j.resconrec.2011.08.007. URL: https://doi.org/10.1016/j.resconrec.2011.08.007.

[125] B. Riaño and M.C. García-González. "On-farm treatment of swine manure based on solid–liquid separation and biological nitrification–denitrification of the liquid fraction". In: *Journal of Environmental Management* 132 (Nov. 2013), pp. 87–93. DOI: 10.1016/j.jenvman.2013.10.014. URL: https://doi.org/10.1016/j.jenvman.2013.10.014.

[126] Tom Rodgers. *Chemical Engineering Plant Cost Index*. The University of Manchester, Department of Chemical Engineering. Accessed: 2026-04-17. URL: https://personalpages.manchester.ac.uk/staff/tom.rodgers/Interactive_graphs/CEPCI.html?reactors/CEPCI/index.html.

[127] Joana Rodrigues et al. "Animal Slurry Sanitization through pH Adjustment: Process Optimization and Impact on Slurry Characteristics". In: *Agronomy* 11.3 (Mar. 2021), p. 517. DOI: 10.3390/agronomy11030517. URL: https://doi.org/10.3390/agronomy11030517.

[128] Ivan Rodriguez-Verde et al. "Assessing anaerobic co-digestion of pig manure with agroindustrial wastes: The link between environmental impacts and operational parameters". In: *The Science of The Total Environment* 497-498 (Aug. 2014), pp. 475–483. DOI: 10.1016/j.scitotenv.2014.07.127. URL: https://doi.org/10.1016/j.scitotenv.2014.07.127.

[129] Rtw. *De samenstelling van organische meststoffen | NutriNorm*. Feb. 2024. URL: https://nutrinorm.nl/meststoffen/de-samenstelling-van-organische-meststoffen/.

[130] I. Saidu et al. "Biochemical Composition of Chlorella vulgaris Grown on Sugarcane Molasses". In: *Journal of Engineering Research and Reports* (May 2021), pp. 115–122. DOI: 10.9734/jerr/2021/v20i617334. URL: https://doi.org/10.9734/jerr/2021/v20i617334.

[131] Laura Sánchez-Martín et al. "Cost model for biogas and biomethane production in anaerobic digestion and upgrading. Case study: Castile and Leon". In: *Materials* 16.1 (Dec. 2022), p. 359. DOI: 10.3390/ma16010359. URL: https://doi.org/10.3390/ma16010359.

[132] Germán Giner Santonja et al. *Best Available Techniques (BAT) Reference Document for the Intensive Rearing of Poultry or Pigs*. JRC Science for Policy Report EUR 28674 EN. Luxembourg: Joint Research Centre (JRC), European Commission, 2017. DOI: 10.2760/020485.

[133] Andreia D. Santos et al. "A biochemical methane potential of pig slurry". In: *Energy Reports* 8 (Feb. 2022), pp. 153–158. DOI: 10.1016/j.egyr.2022.01.127. URL: https://doi.org/10.1016/j.egyr.2022.01.127.

[134] Cristian A. Sepúlveda-Muñoz et al. "Photosynthetic treatment of piggery wastewater in sequential purple phototrophic bacteria and microalgae-bacteria photobioreactors". In: *Journal of Water Process Engineering* 47 (May 2022), p. 102825. DOI: 10.1016/j.jwpe.2022.102825. URL: https://doi.org/10.1016/j.jwpe.2022.102825.

[135] Xiaoxiao Shi et al. "Decoding butyrate fermentation with glucose as a model substrate: Parameter optimization and metagenomic insights". In: *Bioresource Technology* 439 (Sept. 2025), p. 133365. DOI: 10.1016/j.biortech.2025.133365. URL: https://doi.org/10.1016/j.biortech.2025.133365.

[136] Doyun Shin, Youngyeon Kim, and Hee Sun Moon. "Fate and toxicity of spilled chemicals in groundwater and soil environment I: strong acids". In: *Environmental Health and Toxicology* 33.4 (Dec. 2018), e2018019. DOI: 10.5620/eht.e2018019. URL: https://doi.org/10.5620/eht.e2018019.

[137] Amer Ait Sidhoum et al. "The role of rational decisions in technical inefficiency analysis of Spanish pig farms: The influence of water use management". In: *Resources Conservation and Recycling* 199 (Oct. 2023), p. 107278. DOI: 10.1016/j.resconrec.2023.107278. URL: https://doi.org/10.1016/j.resconrec.2023.107278.

[138] Sigma-Aldrich. *Ethyl Acetate Anhydrous, 99.8%*. Accessed: April 22, 2026. 2026. URL: https://www.sigmaaldrich.com/NL/en/product/aldrich/490229.

[139] Yapeng Song et al. "Microbiological and Technological Insights on Anaerobic Digestion of Animal manure: a review". In: *Fermentation* 9.5 (May 2023), p. 436. DOI: 10.3390/fermentation9050436. URL: https://doi.org/10.3390/fermentation9050436.

[140] Alfred M. Spormann. *Fermentative Metabolism*. Springer, Dec. 2023, pp. 215–269. DOI: 10.1007/978-3-031-28218-8\{_}9. URL: https://doi.org/10.1007/978-3-031-28218-8_9.

[141] Adrie Straathof. *Process assessment: Bioprocess Integration*. Lecture slides. TU Delft, Department of Biotechnology, Oct. 2024.

[142] Jiaxin Sun et al. "Anaerobic co-biodegradation of polyhydroxyalkanoate and swine manure for volatile fatty acid production: The impact of C/N ratios and microbial dynamics". In: *Bioresource Technology* 418 (Dec. 2024), p. 131995. DOI: 10.1016/j.biortech.2024.131995. URL: https://doi.org/10.1016/j.biortech.2024.131995.

[143] Ziyan Sun et al. "Deciphering the impact of lignin on anaerobic digestion: Focus on inhibition mechanisms and methods for alleviating inhibition". In: *ACS Omega* 9.44 (Oct. 2024), pp. 44033–44041. DOI: 10.1021/acsomega.4c04375. URL: https://doi.org/10.1021/acsomega.4c04375.

[144] Jelmer Tamis et al. "Enrichment of Plasticicumulans acidivorans at pilot-scale for PHA production on industrial wastewater". In: *Journal of Biotechnology* 192 (Oct. 2014), pp. 161–169. DOI: 10.1016/j.jbiotec.2014.10.022. URL: https://doi.org/10.1016/j.jbiotec.2014.10.022.

[145] Xue Tao et al. "Techno-economic evaluation of microalgae harvesting and dewatering systemsRecent advances in polyhydroxyalkanoate production from volatile fatty acids derived from food waste fermentation". In: *Frontiers in Microbiology* 16 (Oct. 2025), p. 1693596. DOI: 10.3389/fmicb.2025.1693596. URL: https://doi.org/10.3389/fmicb.2025.1693596.

[146] TheGlobalEconomy. *Spain business credit interest rate*. Data from Bank of Spain in percent. 2026. URL: https://www.theglobaleconomy.com/Spain/business_credit_interest_rate/.

[147] Isabel Thiele and Sebastian L. Riedel. “How does downstream processing influence the sustainability and techno-economics of polyhydroxyalkanoates production?” In: *Journal of Cleaner Production* 521 (July 2025), p. 146257. DOI: 10.1016/j.jclepro.2025.146257. URL: https://doi.org/10.1016/j.jclepro.2025.146257.

[148] TU Delft OpenCourseWare. *Summary table with heating values and CO2 emissions*. Energy Supply Systems for Buildings. Accessed: 2026-01-21. URL: https://ocw.tudelft.nl/wp-content/uploads/Summary_table_with_heating_values_and_CO2_emissions.pdf.

[149] United States Environmental Protection Agency. *Catalog of CHP technologies*. Tech. rep. United States Environmental Protection Agency, 2015.

[150] Universidad Politécnica de Madrid et al. *Layman Report: Processing of livestock waste for the production of biomethane for use in agricultural vehicles and biofertilizers*. Layman’s Report LIFE19 CCM/ES/001206. Accessed: 2026-01-22. Brussels, Belgium: LIFE Programme, European Commission, 2025. URL: https://webgate.ec.europa.eu/life/publicWebsite/project/LIFE19-CCM-ES-001206/processing-of-livestock-waste-for-the-production-of-biomethane-for-use-in-agricultural-vehicles-and-biofertilizers.

[151] Francesco Valentino et al. “Carbon recovery from wastewater through bioconversion into biodegradable polymers”. In: *New Biotechnology* 37.Pt A (June 2016), pp. 9–23. DOI: 10.1016/j.nbt.2016.05.007. URL: https://doi.org/10.1016/j.nbt.2016.05.007.

[152] Tony J. Van Der Weerden et al. “Ammonia and nitrous oxide emission factors for excreta deposited by livestock and land□applied manure”. In: *Journal of Environmental Quality* 50.5 (July 2021), pp. 1005–1023. DOI: 10.1002/jeq2.20259. URL: https://doi.org/10.1002/jeq2.20259.

[153] Jey-R Ventura and Ruby Lynn Ventura. “Techno-Economic Feasibility of Polyhydroxybutyrate (PHB) Production from Corn Stover”. In: *The Philippine Agricultural Scientist* 107.1 (Mar. 2024). DOI: 10.62550/ds037022. URL: http://dx.doi.org/10.62550/ds037022.

[154] Ruben Vingerhoets et al. “Enhancing swine manure treatment: A full-scale techno-economic assessment of nitrogen recovery, pure oxygen aeration and effluent polishing”. In: *Journal of Environmental Management* 356 (Mar. 2024), p. 120646. DOI: 10.1016/j.jenvman.2024.120646. URL: https://doi.org/10.1016/j.jenvman.2024.120646.

[155] Jun Wang and Wayne R. Curtis. “Proton stoichiometric imbalance during algae photosynthetic growth on various nitrogen sources: toward metabolic pH control”. In: *Journal of Applied Phycology* 28.1 (Apr. 2015), pp. 43–52. DOI: 10.1007/s10811-015-0551-3. URL: https://doi.org/10.1007/s10811-015-0551-3.

[156] Meng Wang et al. “Anaerobic Co-digestion of Swine Manure and Microalgae Chlorella sp.: Experimental Studies and Energy Analysis”. In: *BioEnergy Research* 9.4 (2016), pp. 1204–1215. DOI: 10.1007/s12155-016-9769-4.

[157] Zhiwei Wang et al. “Continuous Production of Volatile Fatty Acids (VFAs) from Swine Manure: Determination of Process Conditions, VFAs Composition Distribution and Fermentation Broth Availability Analysis”. In: *Water* 14.12 (June 2022), p. 1935. DOI: 10.3390/w14121935. URL: https://doi.org/10.3390/w14121935.

[158] Menghan Wu et al. “Comprehensive Techno-environmental Evaluation of a Pilot-Scale PHA Production from Food Waste in China”. In: *Environmental Science & Technology* 57.3 (Dec. 2022), pp. 1467–1478. DOI: 10.1021/acs.est.2c05976. URL: https://doi.org/10.1021/acs.est.2c05976.

[159] Wei Wu, Liang-Chiung Cheng, and Jo-Shu Chang. “Environmental life cycle comparisons of pig farming integrated with anaerobic digestion and algae-based wastewater treatment”. In: *Journal of Environmental Management* 264 (Apr. 2020), p. 110512. DOI: 10.1016/j.jenvman.2020.110512. URL: https://doi.org/10.1016/j.jenvman.2020.110512.

[160] Yasemin Dilsad Yilmazel and Metin Duran. “Biohydrogen production from cattle manure and its mixtures with renewable feedstock by hyperthermophilic Caldicellulosiruptor bescii”. In: *Journal of Cleaner Production* 292 (Jan. 2021), p. 125969. DOI: 10.1016/j.jclepro.2021.125969. URL: https://doi.org/10.1016/j.jclepro.2021.125969.

[161] Hiroko Yoshida et al. "Long-Term emission factors for land application of treated organic municipal waste". In: *Environmental Modeling Assessment* 21.1 (Aug. 2015), pp. 111–124. DOI: 10.1007/s10666-015-9471-5. URL: https://doi.org/10.1007/s10666-015-9471-5.

[162] Guoyan Zhang et al. "The carry-over effect of dietary manipulation on pig manure storage and composting: influence on carbon and nitrogen flows and footprints". In: *Journal of Cleaner Production* 521 (July 2025), p. 146222. DOI: 10.1016/j.jclepro.2025.146222. URL: https://doi.org/10.1016/j.jclepro.2025.146222.

[163] Ziying Zhang et al. "Effect of composition of volatile fatty acids on yield of polyhydroxyalkanoates and mechanisms of bioconversion from activated sludge". In: *Bioresource Technology* 385 (July 2023), p. 129445. DOI: 10.1016/j.biortech.2023.129445. URL: https://doi.org/10.1016/j.biortech.2023.129445.

[164] Jialiang Zhou et al. "Pretreatment of pig manure liquid digestate for microalgae cultivation via innovative flocculation-biological contact oxidation approach". In: *The Science of The Total Environment* 694 (Aug. 2019), p. 133720. DOI: 10.1016/j.scitotenv.2019.133720. URL: https://doi.org/10.1016/j.scitotenv.2019.133720.

[165] O. R. Zimmo, N. P. van der Steen, and H. J. Gijzen. "Comparison of ammonia volatilisation rates in algae and duckweed-based waste stabilisation ponds treating domestic wastewater". In: *Water Research* 37.19 (2003), pp. 4587–4594. DOI: 10.1016/j.watres.2003.08.013.

A

Appendix: Background information

A.1. Compost emission factors

Table A.1: Variations in emissions to air and soil during composting process in percentage of inflowing stream.

Description	Raw manure IPCC [76]	Digestate swine manure [59]	Digestate from codigestion 30-35% manure[124]
Location	–	Brazil	Germany
Climate zone	Temperate climate	Tropical	Warm temperate moist & Cool temperate moist
C loss [% C]	–	65	50
CH_4 [% C]	1.7	6	8.12
CO_2 [% C]	–	57	41.88
Water loss [% H_2O]	–	–	80
N loss [% TN]	–	60	–
NH_3 [%]	65% TN for NH_3 & NO_x	10% TN	20 % NH4-N
NO_x [%]		0	0
N_2O [% TN]	0.5	6	1.4
N_2 [% TN]	1.5	44 %	–
$N_{leaching}$ to soil [% TN inflow]	4	–	–

A.2. Ammonia concentrations during bioplastic production process

Table A.2: Ammonia nitrogen limitations for acidogenesis, PHA enrichment and production and micro algae cultivation

Inflow	Acidogenesis	PHA enrichment	PHA production	Micro algae cultivation
TAN condition [g/L]	< 1.2	0.5 - 1.0		< 0.3
TAN applied [g/L]	0.2 - 0.6	0.1	0.5 - 3.5	–
TAN activity	production	(slight) consumption		consumption
Source	[157]	[54]		[164]

A.3. Fate of chemicals

Polydimethylsiloxane (PDMS) is highly hydrophobic and chemically stable. Studies evaluating pilot-scale composting of municipal sludge amended with PDMS show that the compound does not biodegrade during the active composting phase and remains largely intact throughout the biological treatment process. The presence of PDMS does not negatively affect the composting process. No inhibition of microbial activity, temperature development, or carbon dioxide evolution has been observed. Furthermore, no adverse effects have been reported when the compost is applied to agricultural land. The degradation of PDMS primarily occurs after land application of the compost, under natural environmental conditions, the compound is subsequently biodegraded by soil microorganisms into carbon dioxide, water, and inorganic silicates. Based on these findings, the environmental impact of the silicone-based defoamer is expected to be limited [90]. Sulfuric acid is recognized as a potentially hazardous sub-

stance due to its strong acidity and its capacity to alter soil and groundwater chemistry, depending on environmental conditions such as soil composition and buffering capacity. However, within the system considered in this study, sulfuric acid is in very low concentrations , and as a result its potential environmental impact is expected to be low. Nevertheless, in the case of accidental release, local effects could be significant due to its high reactivity and dependence on site-specific conditions [136]. Life

cycle assessment studies using the same recycle ratio do not explicitly include these emissions in their inventories [158, 30]. As only 15% can be emitted as gas to the stratosphere and chloroform has a relatively low global warming potential, the environmental impact of chloroform emissions in this study is assumed to be negligible [102]. The fate of sodium hypochlorite should also be considered. NaClO

is a very toxic chemical, mostly to aquatic organisms. However, due to its highly reactive nature, it does not persist in the environment. When released, it rapidly reacts with organic matter and other substances, leading to its degradation before it can accumulate or cause long-term impacts. Therefore, despite its potential toxicity, the overall environmental impact of sodium hypochlorite is limited due to its rapid degradation and low persistence in natural systems. Given the rapid degradation, no NaOCl is expected to be measured in outflowing streams of the extraction step, only in forms of unharmful ions (Na^+, Cl^-). [7].

B

Appendix: Model background and outcomes

B.1. Electricity consumption

Table B.1: Energy consumption per process in both production scenarios per treatment of 1000 kg manure.

Biogas scenario	kWh	total [%]
AD	0.25	3
Decanter	3.98	51
Cultivation	3.07	40
Composting	0.45	6
Total	7.75	
Bioplastic scenario	kWh	total [%]
Pretreatment	2.30	0.87
Acidogenesis	53.5	20
Decanter	4.41	1.7
PHA enrichment and production	133	51
Second decanter	4.87	1.9
PHA extraction	58.6	22
Micro-algae cultivation	0.53	0.2
Composting	0.42	0.2
Total	262	

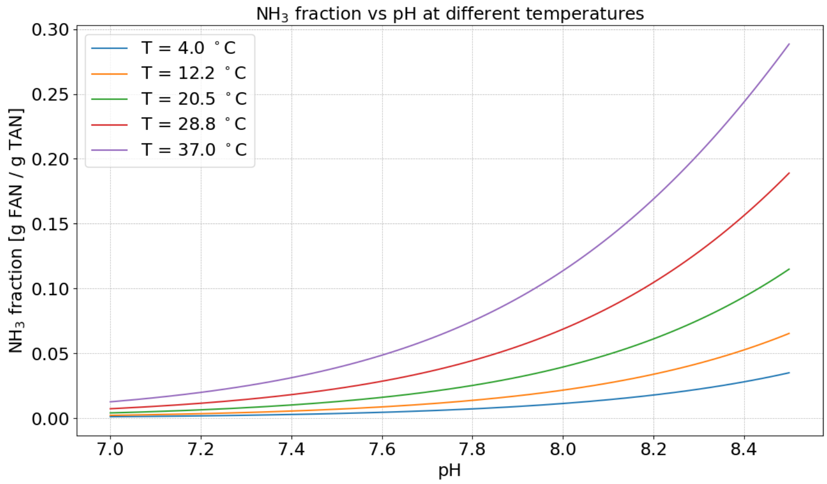
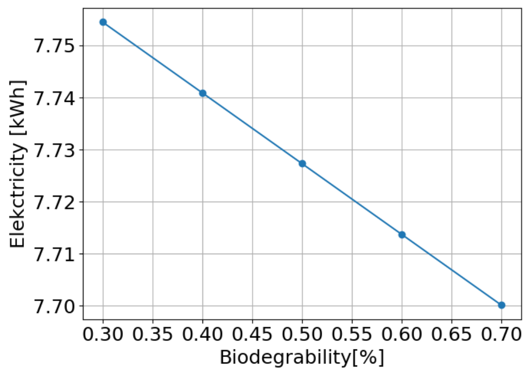


Figure B.1: Relation between FAN and TAN over different temperatures and pH

B.2. FAN TAN relation



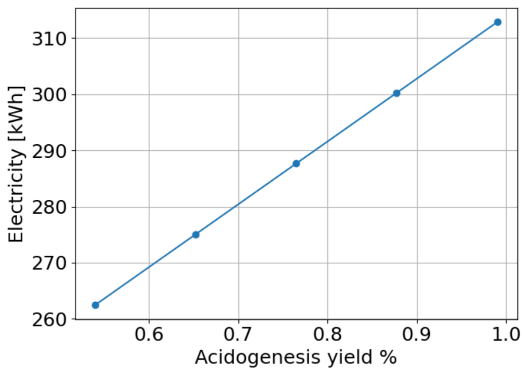


Figure B.2: Electricity demand over yield improvement biogas (left) and bioplastic (right).

Table B.2: Total mass flow though swine manure treatment system per component per operator step.

A) Biogas production scenario

	Manure [kg]	Digestate [kg]	Influent decanter [kg]	SF decanter [kg]	Compost [kg]	LF decanter [kg]	Outflow cultivation [kg]	Algae [kg]
Mass [kg]	1.00E+03	9.96E+02	9.96E+02	9.00E+01	2.48E+01	9.04E+02	5.38E+03	7.17E+00
TS [kg]	2.47E+01	1.85E+01	1.85E+01	1.20E+01	9.17E+00	6.47E+00	6.47E+00	6.97E+00
Water [kg]	9.75E+02	9.75E+02	9.75E+02	7.80E+01	1.56E+01	8.97E+02	5.37E+03	1.99E-01
VS [kg]	2.28E+01	1.66E+01	1.66E+01	9.61E+00	8.16E+00	6.96E+00	9.74E-01	5.78E+00
TAN [kg]	2.00E+00	2.00E+00	1.88E+00	2.44E-01	1.95E-01	1.62E+00	1.45E-01	1.90E-02
Ash [kg]	1.90E+00	1.90E+00	1.90E+00	1.01E+00	1.01E+00	8.93E-01	–	–
TN [kg]	2.60E+00	2.47E+00	2.04E+00	4.90E-01	3.94E-01	1.55E+00	2.48E-01	5.93E-01
P [kg]	6.00E-01	6.00E-01	6.00E-01	4.26E-01	4.26E-01	1.68E-01	1.50E-02	1.31E-01
K [kg]	3.50E+00	3.50E+00	3.50E+00	3.50E-01	3.50E-01	3.15E+00	3.15E+00	–
C [kg]	1.08E+01	8.13E+00	8.13E+00	5.69E+00	2.85E+00	2.44E+00	2.13E+00	2.04E+00

B) Bioplastic production scenario

	Raw PM [kg]	Pretreat [kg]	Digestate [kg]	SF decant. [kg]	LF decant. [kg]	PHA liq. [kg]	LF decant 2 [kg]	LF out cult. [kg]	Algae [kg] v	SF decant 2 [kg]	Bio-waste [kg]	Input comp. [kg]	Compost [kg]
Water [kg]	9.75E+02	1.09E+03	1.09E+03	6.51E+01	1.02E+03	1.19E+03	1.12E+03	1.12E+03	3.40E-02	7.16E+01	7.16E+01	1.37E+02	2.73E+01
TAN [kg]	2.00E+00	3.10E-01	6.50E-01	5.20E-02	5.98E-01	5.98E-01	5.51E-01	5.00E-02	3.00E-03	4.80E-02	4.80E-02	1.00E-01	8.00E-02
FAAN [kg]	4.00E-02	1.00E-01	4.40E-01	3.50E-02	4.05E-01	4.05E-01	3.72E-01	0.00E+00	–	3.20E-02	0.00E+00	3.50E-02	–
TN [kg]	2.60E+00	7.03E-01	7.03E-01	1.76E-01	5.27E-01	5.27E-01	3.95E-01	6.30E-02	1.02E-01	1.32E-01	1.32E-01	3.08E-01	2.58E-01
TS [kg]	2.47E+01	2.47E+01	2.08E+01	1.79E+01	2.92E+00	8.55E+00	1.20E+00	1.17E+00	1.21E+00	7.35E+00	7.35E+00	2.53E+01	8.84E+00
VS [kg]	2.28E+01	2.28E+01	1.05E+01	6.82E+00	3.67E+00	3.67E+00	1.29E+00	1.80E-01	9.99E-01	2.39E+00	2.39E+00	9.20E+00	7.36E+00
VFA [kg]	–	–	8.43E+00	0.00E+00	8.43E+00	0.00E+00	–	–	–	–	–	–	–
Ash [kg]	1.90E+00	1.90E+00	1.90E+00	1.01E+00	8.93E-01	8.93E-01	4.20E-01	–	–	4.73E-01	4.73E-01	1.48E+00	1.48E+00
K [kg]	3.50E+00	1.12E+01	1.12E+01	7.82E-01	1.04E+01	1.04E+01	9.66E+00	9.66E+00	–	7.27E-01	7.27E-01	1.51E+00	1.51E+00
P [kg]	6.00E-01	6.00E-01	6.00E-01	4.68E-01	1.32E-01	1.32E-01	2.90E-02	3.00E-03	2.30E-02	1.03E-01	1.03E-01	5.71E-01	5.71E-01
C [kg]	1.08E+01	1.08E+01	9.64E+00	6.74E+00	2.89E+00	2.89E+00	8.67E-01	8.39E-01	3.52E-01	2.02E+00	5.12E-01	7.26E+00	3.63E+00
PHA [kg]	–	0.00E+00	0.00E+00	0.00E+00	0.00E+00	3.99E+00	5.58E-01	–	–	3.43E+00	5.14E-01	5.14E-01	5.14E-01

C

Appendix: Environmental impact

C.1. Background information LCA

An example of the four steps of LCA is shown in Figure C.1 for manure storage, to visually explain the process.

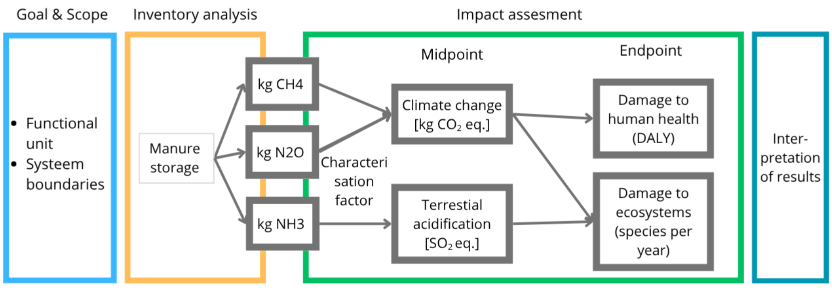


Figure C.1: Example of a Life Cycle Assessment for manure storage.

C.1.1. Midpoint or endpoint

Midpoint indicators describe specific environmental problems along the impact pathway (such as climate change or acidification), linking emissions to environmental mechanisms. This means, that for example all air emissions that lead to climate change are evaluated in one value in amount of equivalents of CO_2. Endpoint indicators translate these impacts into damage to the three areas of protection: human health (DALY's), ecosystem quality (species loss over time), and resource scarcity (extra costs in dollars). To approach the systems in this study for evaluation between both scenarios and with literature, the Midpoint method is chosen to be more appropriate [63].

Time period

In the Midpoint method, different time frames are applied, as the characterized effect differs over time, certain impact have different effects over 20, 100 and 1000 years. The time frames that are considered in the ReCiPe method are:

1. The individualistic perspective: this emphasizes short-term interests, focuses on undisputed impact types, and exhibits technological optimism regarding human adaptation. A time frame of 20 years is considered.
2. The hierarchic perspective: this relies on scientific consensus, considering both the time frame and the plausibility of impact mechanisms. This perspective goes until 100 years ahead.
3. The egalitarian perspective: this time frame is the most precautionary, accounting for the longest time frame (1000 years) and all available impact pathways. This perspective is the most pessimistic regarding future social-economic developments.

For this LCA, a hierarchic perspective is chosen, as it relies on scientific consensus, focusing on well-established impact mechanisms with predictable outcomes. Pathways such as nutrient leaching and ammonia emissions are adopted in this frame. Additionally, it considers manageable time frame, which allows for a realistic and actionable assessment of environmental impacts. This approach helps avoid the uncertainty associated with long-term predictions while ensuring relevant, evidence-based comparison of processes [63].

C.2. Primary data from Simapro

Table C.1: SimaPro 10.3.0.1 analysis 16-03-2026. Results for impact assessment of 1000 kg swine manure treated (30% yield) using the ReCiPe 2016 Midpoint (H) V1.12 method for a Biogas production scenario.

Impact category	Unit	Total	Manure	Biogas	Energy	Compost	Algae	Digestate	Sub N	Sub P	Sub Heat	Sub Elec
Impact category	Unit	Total	Manure	Biogas	Energy	Compost	Algae	Digestate	Sub N	Sub P	Sub Heat	Sub Elec
Global warming	kg CO_2 eq	22.749	0	3.089	0.039	23.688	0.061	0.0003	-2.192	-0.986	-0.125	-0.826
Strat. ozone depletion	kg CFC11 eq	7.58E-05	0	0	1.29E-06	8.02E-05	2.27E-08	1.05E-08	-3.53E-06	-1.35E-06	-3.08E-07	-4.84E-07
Ionizing radiation	kBq Co-60 eq	-0.947	0	0	0	0	0.021	0	-0.112	-0.073	-0.002	-0.780
Ozone human health	kg NO_x eq	0.0106	0	0.0107	0.0076	0	0.00014	0	-0.00379	-0.00185	-0.00024	-0.00196
Fine particulate matter	kg $PM_{2.5}$ eq	0.0248	0	0.0216	0.00062	0.00747	9.66E-05	0	-0.00237	-0.00135	-0.00021	-0.00106
Ozone ecosystems	kg NO_x eq	0.0447	0	0.0310	0.0220	0	0.00014	0	-0.00409	-0.00197	-0.00025	-0.00207
Terrestrial acidification	kg SO_2 eq	0.388	0	0.296	0.00262	0.105	0.00022	0	-0.00876	-0.00474	-0.00062	-0.00294
Freshwater eutrophication	kg P eq	-0.000833	0	0	0	0	0.000241	0	-0.000476	-0.000310	-3.96E-05	-0.000249
Marine eutrophication	kg N eq	0.0194	0	0	0	0.00262	0.0171	6.63E-08	-0.000228	-4.19E-05	-2.08E-06	-2.65E-05
Terrestrial ecotoxicity	kg 1,4-DCB	-18.606	0	0	0	0	0.168	0	-11.102	-3.948	-0.150	-3.574
Freshwater ecotoxicity	kg 1,4-DCB	-0.381	0	0	0	0	0.00538	0	-0.149	-0.0708	-0.0030	-0.164
Marine ecotoxicity	kg 1,4-DCB	-0.479	0	0	0	0	0.00756	0	-0.187	-0.0921	-0.00400	-0.203
Human carcinogenic toxicity	kg 1,4-DCB	-0.595	0	0	0	0	0.195	0	-0.414	-0.196	-0.0177	-0.162
Human non-carc. toxicity	kg 1,4-DCB	-5.937	0	0	0	0	0.0929	0	-2.758	-1.383	-0.100	-1.788
Land use	m^2a crop eq	-0.199	0	0	0	0	0.00201	0	-0.0820	-0.0696	-0.00730	-0.0417
Mineral resource scarcity	kg Cu eq	-0.0584	0	0	0	0	0.000753	0	-0.0274	-0.0254	-0.000265	-0.00609
Fossil resource scarcity	kg oil eq	-1.342	0	0	0	0	0.0163	0	-0.792	-0.302	-0.0186	-0.246
Water consumption	m^3	0.121	0	0	0	0	0.202	0	-0.0552	-0.0134	-0.000204	-0.0125

Table C.2: SimaPro 10.3 analysis conducted on 15-04-2026. Results for impact assessment of 1000 kg treated swine manure to bioplastic (yield 54%) using the ReCiPe 2016 Midpoint (H) V1.12 method for Bioplastic production scenario.

Impact category	Unit	Total	Elec	Pre	PHA prod	PHA ext	Comp	Pond	Sub N	Sub P	Sub am	Sub PETT
Global warming	kg CO2 eq	117.677	52.648	25.022	6.075	13.101	31.321	0.210	-0.806	-1.050	-8.486	-0.358
Strat. ozone depletion	kg CFC11 eq	0.0029	3.08E-05	8.11E-06	0.0008	0.0020	6.19E-05	1.23E-07	-1.29E-06	-1.43E-06	-1.32E-06	-1.36E-06
Ionizing radiation	kBq Co-60 eq	56.531	49.715	5.118	0.316	1.839	0.023	0.198	-0.041	-0.078	-0.538	-0.021
Ozone human health	kg NOx eq	0.212	0.125	0.064	0.010	0.015	0.014	0.0004	-0.001	-0.001	-0.013	-0.001
Fine particulate	kg PM2.5 eq	0.112	0.067	0.048	0.007	0.009	0.004	0.0002	-0.0008	-0.001	-0.023	-0.0003
Ozone ecosystems	kg NOx eq	0.221	0.131	0.066	0.010	0.015	0.015	0.0005	-0.001	-0.002	-0.014	-0.001
Terrestrial acid.	kg SO2 eq	0.303	0.187	0.139	0.017	0.022	0.031	0.0007	-0.003	-0.005	-0.085	-0.0007
Freshwater eutro.	kg P eq	0.033	0.015	0.012	0.001	0.005	0.0002	0.0001	-0.0001	-0.0003	-0.001	-0.0002
Marine eutro.	kg N eq	0.014	0.001	0.0008	0.0001	0.0005	0.002	0.018	-8.39E-05	-4.46E-05	-0.008	-5.33E-05
Terrestrial ecotox.	kg 1,4-DCB	144.396	227.783	138.415	5.676	21.540	22.320	0.909	-4.084	-4.205	-260.890	-3.067
Freshwater ecotox.	kg 1,4-DCB	12.315	10.430	1.983	0.123	0.686	0.060	0.042	-0.055	-0.075	-0.858	-0.020
Marine ecotox.	kg 1,4-DCB	15.805	12.946	2.616	0.165	0.894	0.100	0.051	-0.068	-0.098	-0.773	-0.028
Human carc. tox.	kg 1,4-DCB	17.880	10.308	5.953	0.681	2.586	0.305	0.041	-0.152	-0.209	-1.571	-0.062
Human non carc. tox.	kg 1,4-DCB	172.562	113.946	44.135	5.519	22.405	1.040	0.454	-1.014	-1.473	-11.953	-0.496
Land use	m2a crop eq	4.037	2.659	1.109	0.256	0.264	0.028	0.010	-0.030	-0.074	-0.168	-0.018
Mineral scarcity	kg Cu eq	0.506	0.388	0.135	0.013	0.041	0.006	0.001	-0.010	-0.027	-0.042	-0.0009
Fossil scarcity	kg oil eq	24.987	15.657	7.593	1.582	2.698	1.035	0.062	-0.291	-0.322	-2.953	-0.075
Water consumption	m3	1.015	0.795	0.365	0.048	0.080	0.003	0.003	-0.020	-0.014	-0.245	-0.001

C.2.1. Yield improvements

Biogas 70% yield production

Table C.3: Inventory data for Biogas 70% yield production

Input process	Value	Unit	Output process	Value	Unit
Anaerobic digestion					
Swine manure	1000	kg	Digestate	989.2	kg
Electricity	0.251	kWh	Biogas (66% CH4)	12.58	kg
			CH4 (biogenic)	0.0926	kg
			NH3	0.121	kg
			NOx	0.0091	kg
Digestate storage					
Digestate	989.2	kg	Digestate	988.9	kg
			N leaching (soil)	0.052	kg
			CH4 (biogenic)	0.00372	kg
			N2O	0.0944	kg
Combined heat power					
Biogas (66% CH4)	3.58	kg	Electricity	7.7	kWh
			CO2 (biogenic)	6.46	kg
			NOx	0.00639	kg
Solid liquid separation					
Digestate	988.9	kg	Solid fraction	85.32	kg
Electricity	3.96	kWh	Liquid fraction	902.1	kg
Composting site					
Solid fraction	85.32	kg	Compost	21.06	kg
Electricity	0.427	kWh	CH4 (biogenic)	0.386	kg
Diesel	0.206	kg	CO2 (biogenic)	5.461	kg
			N leaching (soil)	0.0208	kg
Micro algae cultivation					
Liquid fraction	902.1	kg	Algae (dry)	7.17	kg
CO2 from CHP	13.12	kg	Outflowing water	1093	kg
Electricity	3.07	kWh	Nitrogen (to water)	0.0608	kg
Tap water	192.1	kg	Phosphate (to water)	0.0035	kg

Table C.4: Substitution inventory for the biogas production scenario in the anaerobic digestion system industrial plant based on project Arsal specifications.

Substitution category	Value	Unit
Nitrogen fertilizer	0.425	kg
Phosphorus fertilizer	0.383	kg
Biogas	9.000	kg
Electricity	19.65	kWh
Heat	23.18	kWh

Table C.5: SimaPro 10.3 analysis conducted on 05-03-2026. Results for impact assessment of 1000 kg swine manure treated (70% yield AD) using the ReCiPe 2016 Midpoint (H) V1.12 method for biogas production plant

Impact category	Unit	Total	Biogas	CHP	Composting	Algae pond	Dig. storage	Sub N	Sub P	Sub Elec	Sub Heat
Global warming	kg CO2 eq	11.7211	3.0893	0.0384	16.2107	0.0585	0.0002	-2.1967	-0.7696	-4.0890	-0.6207
Strat. ozone depletion	kg CFC11 eq	7.32E-05	0	1.27E-06	8.04E-05	2.16E-08	1.05E-08	-3.53E-06	-1.05E-06	-2.39E-06	-1.52E-06
Ionizing radiation	kBq Co-60 eq	-4.0109	0	0	0.0120	0.0199	0	-0.1122	-0.0573	-3.8612	-0.0121
Ozone human health	kg NOx eq	0.0098	0.0107	0.0075	0.0075	0.0001	0	-0.0037	-0.0014	-0.0097	-0.0011
Fine particulate	kg PM2.5 eq	0.0211	0.0216	0.0006	0.0085	9.21E-05	0	-0.0023	-0.0010	-0.0052	-0.0010
Ozone ecosystems	kg NOx eq	0.0438	0.0310	0.0217	0.0079	0.0001	0	-0.0040	-0.0015	-0.0102	-0.0012
Terrestrial acid.	kg SO2 eq	0.3706	0.2964	0.0026	0.1015	0.0002	0	-0.0087	-0.0037	-0.0145	-0.0030
Freshwater eutro.	kg P eq	-0.0018	0	0	0.0001	0.0001	0	-0.0004	-0.0002	-0.0012	-0.0001
Marine eutro.	kg N eq	0.0202	0	0	0.0026	0.0180	6.63E-08	-0.0002	-3.26E-05	-0.0001	-1.03E-05
Terrestrial ecotox.	kg 1,4-DCB	-20.8315	0	0	11.6513	0.1598	0	-11.1282	-3.0820	-17.6914	-0.7409
Freshwater ecotox.	kg 1,4-DCB	-0.9928	0	0	0.0313	0.0051	0	-0.1491	-0.0553	-0.8100	-0.0147
Marine ecotox.	kg 1,4-DCB	-1.2254	0	0	0.0522	0.0072	0	-0.1876	-0.0718	-1.0055	-0.0198
Human carc. tox.	kg 1,4-DCB	-1.1111	0	0	0.1593	0.1860	0	-0.4150	-0.1532	-0.8006	-0.0875
Human non carc. tox.	kg 1,4-DCB	-12.5596	0	0	0.5429	0.0886	0	-2.7643	-1.0800	-8.8499	-0.4968
Land use	m2a crop eq	-0.3626	0	0	0.0147	0.0019	0	-0.0821	-0.0543	-0.2065	-0.0361
Mineral scarcity	kg Cu eq	-0.0746	0	0	0.0034	0.0007	0	-0.0275	-0.0198	-0.0301	-0.0013
Fossil scarcity	kg oil eq	-1.7822	0	0	0.5403	0.0155	0	-0.7938	-0.2360	-1.2160	-0.0921
Water consumption	m3	0.0656	0	0	0.0017	0.1924	0	-0.0552	-0.0104	-0.0617	-0.0010

Bioplastic 99% improvement

Table C.6: Inventory table for the bioplastic production scenario with 99% efficiency of the bioplastic production scenario. Data based on the treatment of 1000 kg of swine manure.

Input process	Value	Unit	Output process	Value	Unit
Pretreatment					
Swine manure	1000	kg	Pretreated swine manure	1122.7	kg
Electricity	2.3	kWh	Ammonium sulfate	8.934	kg N
KOH / H2SO4 / Tapwater	11 / 5.91 / 15.9	kg			
Acidogenesis					
Pretreated swine manure	1122.7	kg	Digestate	1101.8	kg
Electricity	53.5	kWh	CO2 / H2 (biogenic)	8.4 / 0.084	kg
Solid liquid separation					
Digestate	1101.8	kg	Solid fraction	80.49	kg
Electricity	4.41	kWh	Liquid fraction	1022.5	kg
Pha production					
Liquid fraction	1022.5	kg	Pha containing liquid	1605.7	kg
Electricity	132.8	kWh	CO2 (biogenic)	2.12	kg
H2SO4 / Defoamer	0.637 / 1.27	kg			
Second solid liquid separation					
Pha containing liquid	1605.7	kg	Pha containing solid fraction	109.1	kg
Electricity	6.42	kWh	Liquid fraction	1496.6	kg
Pha extraction					
Pha containing solid fraction	109.1	kg	Pha product (99% pure)	5.48	kg
Electricity	107.4	kWh	Biowaste (Biomass without Pha)	103.6	kg
NaOCl / Chloroform	5.31 / 4.09	kg			
Composting site					
Solid fraction / Biowaste	80.49 / 103.6	kg	Compost	32.19	kg
Electricity / Diesel	0.402 / 0.445	kWh/kg	CH4 / CO2 / N2O / NH3	0.669 / 9.47 / 0.0056 / 0.0115	kg
Micro algae cultivation					
Liquid fraction	1496.6	kg	Algae (dry)	1.24	kg
CO2 from Chp / Electricity	2.27 / 0.533	kg/kWh	N / P / COD (to water)	0.0461 / 0.00147 / 0.00309	kg

Table C.7: Substitution inventory for the bioplastic production scenario in bioplastic production scenario with 99% acidogenesis yield.

Substitution category	Value	Unit
Nitrogen fertilizer	0.156	kg
Phosphorus fertilizer	0.408	kg
Conventional plastic (Pet)	4.028	kg
Ammonium sulfate	8.93	kg

Table C.8: SimaPro 10.3 analysis conducted on 15-04-2026 at 10:27. Results for impact assessment of 1000 kg treated swine manure to bioplastic 99% using the ReCiPe 2016 Midpoint (H) V1.12 method for bioplastic production plant.

Impact category	Unit	Total	Compost	Elec	Liquid	PHA prod	Pre	Sub N	Sub P	Sub am	Sub PETT
Global warming	kg CO2 eq	139.639	26.482	64.051	11.116	23.971	25.022	-0.806	-1.050	-8.490	-0.658
Strat. ozone depletion	kg CFC11 eq	0.0053	6.20E-05	3.75E-05	0.0015	0.0036	8.11E-06	-1.29E-06	-1.43E-06	-1.32E-06	-2.51E-06
Ionizing radiation	kBq Co-60 eq	68.877	0.026	60.483	0.580	3.366	5.118	-0.041	-0.078	-0.538	-0.039
Ozone human health	kg NOx eq	0.260	0.016	0.152	0.018	0.027	0.064	-0.001	-0.001	-0.013	-0.002
Fine particulate	kg PM2.5 eq	0.141	0.005	0.082	0.014	0.016	0.048	-0.0008	-0.001	-0.023	-0.0005
Ozone ecosystems	kg NOx eq	0.272	0.017	0.160	0.019	0.028	0.066	-0.001	-0.002	-0.014	-0.002
Terrestrial acid.	kg SO2 eq	0.379	0.034	0.227	0.032	0.040	0.139	-0.003	-0.005	-0.085	-0.001
Freshwater eutro.	kg P eq	0.042	0.0002	0.019	0.003	0.010	0.012	-0.0001	-0.0003	-0.001	-0.0003
Marine eutro.	kg N eq	0.011	0.002	0.002	0.0002	0.0009	0.0008	-8.39E-05	-4.46E-05	-0.008	-9.81E-05
Terrestrial ecotox.	kg 1,4-DCB	213.590	25.143	277.121	8.425	39.424	138.415	-4.084	-4.205	-261.007	-5.642
Freshwater ecotox.	kg 1,4-DCB	15.193	0.067	12.688	0.224	1.256	1.982	-0.054	-0.075	-0.858	-0.037
Marine ecotox.	kg 1,4-DCB	19.423	0.112	15.751	0.300	1.636	2.616	-0.068	-0.098	-0.773	-0.052
Human carc. tox.	kg 1,4-DCB	22.773	0.343	12.541	1.250	4.732	5.953	-0.152	-0.209	-1.571	-0.115
Human non carc. tox.	kg 1,4-DCB	219.538	1.171	138.626	9.965	40.999	44.135	-1.014	-1.473	-11.958	-0.913
Land use	m2a crop eq	5.023	0.031	3.236	0.469	0.483	1.109	-0.030	-0.074	-0.168	-0.034
Mineral scarcity	kg Cu eq	0.634	0.007	0.472	0.023	0.076	0.135	-0.010	-0.027	-0.042	-0.001
Fossil scarcity	kg oil eq	31.933	1.166	19.048	2.893	4.937	7.593	-0.291	-0.322	-2.954	-0.138
Water consumption	m3	1.297	0.003	0.967	0.094	0.147	0.365	-0.020	-0.014	-0.245	-0.002

D

Appendix: Economic evaluation

D.1. Assumptions

Table D.1: Investment site factor

Country	Investment site factor	Source
France	1,13	[120]
Spain	1,14	[103]
Brazil	1,14	[120]
US	1,03	[120]
Italy	1,14	[120]
Slovenia	1,14	assumed
India	1,02	[120]

D.2. Equipment costs

Biogas scenario	Flow rate [m^3/h]	HRT	V_R [m^3]	Other	Price	Source
Anaerobic digestion	1.07	32	989		€ 259,434.56	[32]
CHP	–			12 kWe	€ 55,567.90	[149]
Digestate storage tank	1.07	70	1800		–	[132]
Decanter	1.07		582		€ 49,573.44	[132]
Composting site	0.10	154	18		€ 47,388.13	[32]& [132]
Micro-algae cultivation			139		€ 17,654.18	[116]
Total					€ 429,618.22	
Bioplastic scenario	**Flow rate [m^3/h]**	**HRT [days]**	**V_R [m^3]**	**Other**	**Price**	**Source**
Ammonia stripping column	1.07		0.0		€ 83,117.90	[132]
Acidogenesis	1.20	1.5	52.0		€ 217,638.52	[88]
Decanter	1.19				€ 52,789.38	[132]
Storage tank	1.32	5.0	190.1		€ 23,528.99	[110]
PHA enrichment	0.55	4.0	63.3		€ 244,820.58	[88]
PHA accumulation	1.51	0.4	15.4		€ 104,980.84	[88]
Decanter 2	1.30				€ 55,852.07	[132]
PHA extraction	1.30				€ 58.425,17	[110]
Composting site	0.15	154.0	24.9		€ 61,087.48	[32]
Micro-algae cultivation			24.0		€ 6,160.12	[116]
Total					€ 908.401,06	

Table D.2: Total equipment costs

D.3. Operational costs

Table D.3: Combined operational expenditure

Operational expenditure category	Scenario 1 [€]	Scenario 2 [€]
Total labor cost (TLC)	36.675,20	1.281.240,58
Insurance	19.900,10	36.868,31
Local taxes	39.800,21	73.736,62
Factory expense	99.500,52	184.341,54
Maintenance	43.242,30	88.349,65
Laboratory control	1.833,76	192.186,09
Electricity demand operators	0,00	275.861,76
Raw materials	2.216,95	98.383,18
Waste treatment/ disposal	2.140,62	1.808,50
Total	245.309,66	2.546.995,29

D.4. Materials

Table D.4: Annual revenue breakdown for the biomethane and biofertilizer plant.

Raw materials	Biogas production scenario		Fraction	Bioplastic production scenario		Fraction
	Mass [kg/yr]	Cost [EUR/kg]	[%]	Mass [kg/yr]	Cost [EUR/kg]	[%]
Pig manure	8500000	0.00	0%	8500000	0.00	0%
Tap water	1640500	0.00194	100%	125800	0.00124	0%
KOH	0	0.00	0%	93500	0.67	58%
H2SO4	0	0.00	0%	53219	0.153	7%
NaOCl	0	0.00	0%	24565	0.383	9%
Chloroform	0	0.00	0%	18955	0.826	14%
Defoamer PDMS	0	0.00	0%	5891	1.19	6%
Diesel	0	0.00	0%	3315	1.72	5%
Total		6374	100%		108702	100%
Products	Biogas production scenario		Fraction	Bioplastic production scenario		Fraction
	Mass [kg/yr]	Cost [EUR/kg]	[%]	Mass [kg/yr]	Cost [EUR/kg]	[%]
Compost	209950	0.0975	29%	273615	0.11	82%
Algae	60945	0.609	53%	10532	0.605	18%
Electricity sold	33730	0.189	9%	0	0.00	0%
Heat sold	33730	0.15	7%	0	0.00	0%
PHA powder	0	0.00	0%	39916	1.64	180%
Ammonium sulfate	0	0.00	0%	75939	0.234	49%
CO2 avoided	18134	0.0848	2%	0	0.00	0%
Total		70541	100%		36358	100%
Waste	Biogas production scenario		Fraction	Bioplastic production scenario		Fraction
	Mass [kg/yr]	Cost [EUR/kg]	[%]	Mass [kg/yr]	Cost [EUR/kg]	[%]
Processed water	9290000	2140.62	0%	7210000	1808.50	0%

D.5. Acetic acid as pure product

Table D.5: Economic parameters for the anaerobic digestion system industrial plant including capital investment and annual operating costs. Process based on price of ammonia stripping column, bioreactor for acidogenesis, storage tank, decanter and composting.

Parameter	Value	Unit
Capital investment (Capex)	EUR	
Listed equipment cost	4.33E+05	same size assumptions as bioplastic production
Unlisted equipment cost	4.33E+04	10 % of listed
Total purchased cost (Pc)	4.76E+05	lang factor of 3.5
Direct fixed capital (Dfc)	1.67E+06 EUR	
Annual operating costs (Opex)	EUR/yr	
Maintenance	4.33E+04	10% of listed PC
Depreciation	4.06E+04	9.38% of DFC, based on interest rate of 3,24% and 15 depreciable years
Insurance	1.67E+04	1% of DFC
Local taxes	3.33E+04	2% of DFC
Factory expense	8.34E+04	5% of DFC
Labor cost	3.67E+04	same labor hours aas biogas production, same labor rate
Laboratory control cost	5.50E+03	15 % of labor cost (same as bioplastic)
Electricity cost	5.48E+04	Electricity demand 3.66E+05 kWh/yr
Raw material cost	4.96E+04	only water
Waste management cost	0.00E+00	none assumed
Total annual operating cost	3.64E+05	
Revenue and production		
Price of acetic acid	0.79	EUR/kg, pure acetic acid [65]
Annual revenue	9.17E+04	EUR/yr
Main product yield	7.35E+04	kg/yr
Main product price	4.95E+00	EUR/kg