

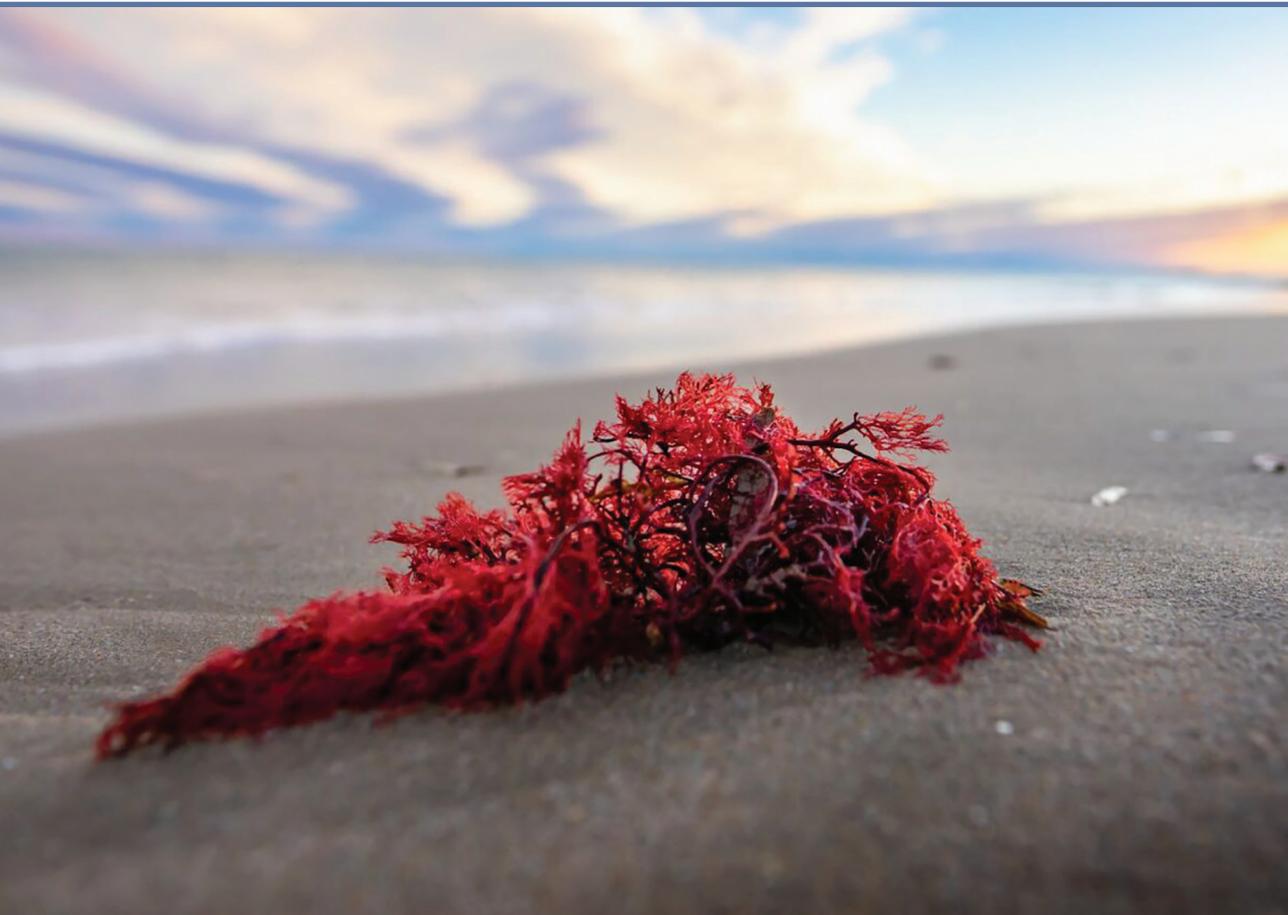


RIGA TECHNICAL
UNIVERSITY

Riccardo Paoli

**NOVEL CASCADE BIOREFINERY CONCEPT FOR
FURCELLARIA LUMBRICALIS: A LIFE CYCLE
SUSTAINABILITY ASSESSMENT-BASED STUDY**

Doctoral Thesis



RIGA TECHNICAL UNIVERSITY

Faculty of Natural Sciences and Technology
Institute of Energy Systems and Environment

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BASED STUDY**

Doctoral Thesis

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ANNOTATION

Climate change and resource scarcity have intensified the global pursuit of sustainable, bio-based solutions, with macroalgae emerging as a promising yet underutilized resource. Within this context, this Doctoral Thesis explores the development of a novel cascade biorefinery framework for *Furcellaria lumbricalis*, a red macroalga abundantly available in the Baltic Sea region. The research is strictly linked to the principles of circular bioeconomy and aligns with the European Green Deal, EU Algae Strategy, and global sustainability agendas, addressing the need for innovative, low-impact, and resource-efficient biomass valorization pathways.

The core objective of the thesis is to define and assess a life cycle sustainability assessment (LCSA) framework for an *F. lumbricalis* cascade biorefinery that integrates three key pillars of sustainability: environmental life cycle assessment (LCA), life cycle costing (LCC), and social life cycle assessment (S-LCA). This multidisciplinary and systems-based approach provides a holistic evaluation of the environmental performance, economic feasibility, and social impacts of multiple biorefinery design options. The novel cascade concept is designed to sequentially extract multiple high-value compounds, namely pigments, proteins, and carrageenan, while also valorizing residual biomass through energy recovery pathways or possible horticultural applications.

Beyond addressing technical, environmental, and economic bottlenecks, the thesis emphasizes the underexplored social dimension of macroalgae-based biorefineries by implementing an original S-LCA framework tailored to the sector. This includes value chain hotspot analysis and community-level assessments of employment, stakeholder engagement, and regional development. The integration of these dimensions culminates in the development of a composite sustainability index using a multi-criteria decision-making method (TOPSIS), enabling the comparison of alternative biorefinery scenarios and the identification of the most sustainable configuration.

The research methodology spans a comprehensive literature review compliant with PRISMA guidelines, detailed case study analysis in the Baltic Sea Region context, original data collection through the TACO ALGAE project, and model validation through peer-reviewed scientific publications and international conferences. The findings underscore the scientific innovation of combining LCSA with macroalgal biorefinery design and highlight the practical significance of such systems for informing policy, guiding industry investments, and supporting sustainable marine bioresources management.

By bridging technological development, environmental evaluation, and social implications, this Thesis provides a blueprint for scalable and replicable cascade biorefinery systems in Europe and beyond. It contributes a novel, transferable methodology for sustainability assessment in the marine bioeconomy and offers actionable insights for advancing *F. lumbricalis* as a cornerstone species in the emerging algae-based bio-industrial sector.

ANOTĀCIJA

Klimata pārmaiņas un resursu trūkums ir pastiprinājuši globālo virzību uz ilgtspējīgiem, uz bioloģiskiem resursiem balstītiem risinājumiem, kur makroaļģes izceļas kā daudzsološs, taču vēl nepietiekami izmantots resurss. Šajā kontekstā šajā promocijas darbā tiek pētīta jauna kaskādes biorafinēšanas koncepcija, kas balstīta uz sarkano makroaļģi *Furcellaria lumbricalis*, kas plaši sastopama Baltijas jūras reģionā. Pētījums cieši saistīts ar aprites bioekonomikas principiem un ir saskaņots ar Eiropas zaļo kursu, ES Aļģu stratēģiju un globālajiem ilgtspējas mērķiem, risinot nepieciešamību pēc inovatīviem, videi draudzīgiem un resursu ziņā efektīviem biomasas valorizācijas ceļiem.

Promocijas darba galvenais mērķis ir izstrādāt un novērtēt Dzīves cikla ilgtspējas novērtējuma (LCSA) sistēmu *F. lumbricalis* kaskādes biorafinēšanai, integrējot trīs galvenos ilgtspējas aspektus: Vides dzīves cikla novērtējumu (LCA), Dzīves cikla izmaksu aprēķinu (LCC) un Sociālo dzīves cikla novērtējumu (S-LCA). Šī starpdisciplinārā un uz sistēmām balstītā pieeja nodrošina visaptverošu vides veiktspējas, ekonomiskās dzīvotspējas un sociālo ietekmju novērtējumu dažādiem biorafinēšanas dizaina variantiem. Inovatīvā kaskādes koncepcija ir izstrādāta tā, lai pakāpeniski iegūtu vairākas augstas pievienotās vērtības vielas — pigmentus, proteīnus un karagīnu — vienlaikus valorizējot atlikušo biomasu enerģijas atgūšanas procesos vai potenciāliem izmantošanas veidiem dārzkopībā.

Papildus tehnisko un ekonomisko šķēršļu risināšanai darbs uzsver līdz šim maz pētīto makroaļģu biorafinēšanas sociālo dimensiju, ieviešot pielāgotu S-LCA novērtējuma sistēmu šai nozarei. Tā ietver vērtību ķēdes karsto punktu analīzi un sabiedrības līmeņa novērtējumu par nodarbinātību, iesaistīto pušu līdzdalību un reģionālo attīstību. Šo dimensiju integrācija noslēdzas ar saliktā ilgtspējas indeksa izstrādi, izmantojot daudzkritēriju lēmumu pieņemšanas metodi (TOPSIS), kas ļauj salīdzināt dažādus biorafinēšanas scenārijus un identificēt ilgtspējīgāko konfigurāciju.

Pētījuma metodoloģija ietver visaptverošu literatūras analīzi, ievērojot PRISMA vadlīnijas, detalizētu gadījumu izpēti Baltijas kontekstā, oriģinālu datu vākšanu projekta TACO ALGAE ietvaros un modeļa validāciju, pamatojoties uz recenzētiem zinātniskiem rakstiem un prezentācijām starptautiskās konferencēs. Iegūtie rezultāti uzsver zinātnisko inovāciju, apvienojot LCSA ar makroaļģu biorafinēšanas dizainu, un izceļ šādu sistēmu praktisko nozīmi politikas veidošanai, investīciju vadīšanai un ilgtspējīgai jūras bioresursu pārvaldībai.

Apvienojot tehnoloģisko attīstību, vides novērtējumu un sociālās ietekmes analīzi, šis promocijas darbs sniedz ceļvedi mērogojamu un atkārtojamu kaskādes biorafinēšanas sistēmu izstrādei Eiropā un ārpus tās. Tas piedāvā jaunu, pārnesamu metodoloģiju ilgtspējas novērtēšanai jūras bioekonomikā un sniedz praktiskas atziņas, lai virzītu *F. lumbricalis* kā atslēgas sugu jaunajā aļģu bāzētajā bioindustriālajā sektorā.

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And, of course, to Riga, you've become a huge part of me. I'll carry you with me always.

As this amazing chapter comes to a close, I hope it's just the beginning of something even greater. Whatever comes next, I plan to carry the same mindset and spirit that defined these years.

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ABBREVIATIONS

ABE	Acetone-Butanol-Ethanol Fermentation
AMC	Acid-Mediated Carbonization
AP	Acidification Potential
APT	Alkaline Pre-Treatment
AS	Alternative Scenarios
ATPE	Aqueous Two-Phase Extraction
BCS	Best-Case Scenario
BMP	Biochemical Methane Potential
CAGR	Compound Annual Growth Rate
CC	Climate Change
CB	Cascade biorefinery
CTUe	Comparative Toxic Unit for aquatic ecotoxicity
DW	Dry Weight
e-LCI	Economic Life Cycle Inventory
EAE	Enzyme-Assisted Extraction
ECF	Ecotoxicity Freshwater
EF	Eutrophication Freshwater
EM	Eutrophication Marine
FRS	Fossil Resources
FU	Functional Unit
GAE	Gallic Acid Equivalents
GHG	Greenhouse Gases
GWP	Global Warming Potential
HAE	Hydrothermal-Assisted Extraction
HCT	Human Toxicity (cancer)
HNCT	Human Toxicity (non-cancer)
HDC	Hydrodynamic Cavitation
HHV	Higher Heating Value
HPH	High-Pressure Homogenization
HS	Health & Safety of the worker
HSH	High Shear Homogenization
HTC	Hydrothermal Carbonization
HTL	Hydrothermal Liquefaction
HWE	Hot Water Extraction
IL	Ionic Liquid
IPA	Impact Pathway Approach
IR	Ionizing Radiation
IRR	Internal Rate of Return
LCA	Environmental Life Cycle Assessment
LCC	Life Cycle Costing

LCI	Life Cycle Inventory
LCIA	Life Cycle Impact Assessment
LCSA	Life Cycle Sustainability Assessment
LCT	Life Cycle Thinking
LED	Local Economy Development
LU	Land Use
MAE	Microwave-Assisted Extraction
MCDA	Multi-Criteria Analysis
ME	Marine Eutrophication
MEc	Marine Ecotoxicity
MHT	Microwave Hydrothermal Treatment
mrheq.	Medium Risk-Hour equivalent
MRS	Mineral Resources
MUFAs	Monounsaturated Fatty Acid
NaDES	Natural Deep Eutectic Solvents
NPV	Net Present Value
OFC	Off-Shore Cultivation
OFhh	Tropospheric Ozone Formation (human)
OFt	Tropospheric Ozone Formation (ecosystem)
ONC	On-Shore Cultivation
PEF	Pulsed Electric Field
PI	Profitability Index
PLE	Pressurized Liquid Extraction
PM	Particulate Matter
PRISMA	Preferred Reporting Items for Systematic Reviews and Meta-Analyses
PRP	Performance Reference Points
PSSF	Pre-Saccharification and Simultaneous Fermentation
Pt	Eco-point
R-PE	R-Phycoerythrin
REC	Renewable Energy Certificate System
RF	Resource Use Fossil
RM&M	Resource Use Minerals & Metals
ROI	Return of Investment
RS	Reference Scale
RSA	Reference Scale Approach
S-LCA	Social Life Cycle Assessment
S-Pt	Annual Global per capita Units
scCO ₂	Supercritical CO ₂
SFA	Saturated Fatty Acid
SFPS	Sulfated Fucooidan Polysaccharides
SHDB	Social Hotspot Database
SOD	Stratospheric Ozone Depletion

SPE	Single Product Extraction
SPEc	Single Product Extraction Carrageenan
SPEp	Single Product Extraction Pigments
SPEpr	Single Product Extraction Proteins
SWE	Subcritical Water Extraction
TA	Terrestrial Acidification
TE	Terrestrial Ecotoxicity
TLE	Three-Line Extraction
TPC	Total Phenolic Content
TRL	Technological Readiness Level
UAE	Ultrasound-Assisted Extraction
VS	Volatile Solids
WACC	Weighted Average Cost of Capital
WCS	Worse-Case Scenario
WD	Wealth Distribution among the supply chain
WH	Wild Harvesting
WHS	Working hours of a worker
WU	Water Use
WW	Wet Weight

INTRODUCTION

Macroalgae are increasingly recognized as a versatile and sustainable resource within the bio-based economy [1], owing to their unique characteristics and rapid growth potential. These marine organisms represent the fastest-growing biomass [2] and play crucial ecological roles in marine ecosystems [3], including the absorption of dissolved nutrients [4] and their function as habitat-structuring species [5]. Furthermore, macroalgae contribute to coastal protection by mitigating the impact of hazardous waves [6] and play a significant role in carbon sequestration [7]. The biochemical composition of macroalgae is particularly diverse, encompassing a wide array of bioactive compounds such as phytopigments (e.g., carotenoids) [8], polyunsaturated fatty acids [9], phenolic compounds [10], tannins [11], peptides [12], lipids [13], enzymes [14], vitamins [15], carbohydrates [16], and terpenoids [17]. Notably, these compounds are often more readily accessible than those found in terrestrial biomass, primarily due to the lower lignin content of macroalgae [18].

These distinctive properties have facilitated the integration of macroalgae into various commercial applications. In Europe, in particular, these applications have been further supported by policies aimed at promoting a sustainable economy, particularly within the framework of the European Commission's Green Deal. The Deal's objectives, achieving climate neutrality [19], preserving biodiversity [20], and advancing sustainable food production [21] by 2050, highlight the critical role of macroalgae-based biorefineries. A key milestone in this context is the adoption of the EU Algae Initiative [22] in 2022, which sets out a strategic framework of 23 targeted actions aimed at enhancing algae production, streamlining regulatory processes, and increasing public awareness of its benefits. Complementing this initiative, the EU4Algae Platform [23] serves as an interactive hub, fostering collaboration and knowledge exchange within the European algae industry. Further contributions have been made by the Joint Research Centre, which has established guidelines for sustainable European seaweed aquaculture [24], aiming to meet the growing global demand for biomass while ensuring environmental sustainability.

Despite the regulatory progress, fully unlocking the potential of macroalgae necessitates the development of standardized processes across all operational stages, from biomass cultivation or harvesting to chemical extraction, within the biorefinery framework [25]. Several challenges must be addressed before macroalgae biorefineries can achieve full-scale implementation. Currently, research and commercial initiatives remain in the developmental phase, predominantly focusing on single-product extractions while lacking comprehensive studies that facilitate the transition to large-scale applications [26]. The majority of studies have primarily examined the energy recovery potential of macroalgae, rather than an integrated valorization approach. The unique composition of macroalgae requires tailored processing techniques distinct from those applied to traditional lignocellulosic biomass [27].

Presently, research efforts are primarily centered on refining extraction methodologies rather than addressing practical market implementation and sustainability quantifications. While environmental and economic analyses have begun to raise awareness in recent years,

social implications remain largely unexplored. This highlights the need for a truly sustainable approach that integrates, with a balanced approach, all three pillars of sustainability: environmental, economic, and social. The life cycle thinking (LCT) approach provides a comprehensive framework for assessing sustainability by considering the entire life cycle of a product or process, from raw material extraction to end-of-life disposal [28].

This Thesis, therefore, focuses on defining a life cycle sustainability assessment (LCSA) for a novel cascade biorefinery based on the red macroalga *Furcellaria lumbricalis*, which is abundantly available in the Baltic Sea region [29]. The LCSA integrates environmental life cycle assessment (LCA), life cycle costing (LCC), and social life cycle assessment (S-LCA) to provide a complete evaluation of the sustainability of macroalgae-based biorefinery systems.

Objectives and tasks of the Doctoral Thesis

The objective of this Thesis is to evaluate the sustainability of a cascade biorefinery utilizing the red macroalga *F. lumbricalis* through a quantitative LCSA. The defined system aims to recover valuable compounds such as pigments, proteins, and carrageenan, while also valorizing the residual biomass. To achieve this overarching goal, the research is structured around the following key questions:

- What is the current state of macroalgae biorefineries?

This objective seeks to assess and define the latest advancements in macroalgae-based biorefineries, highlighting key challenges and opportunities in the field.

- What are the possible designs for an *F. lumbricalis* biorefinery?

This involves developing a tailored cascade biorefinery model for *F. lumbricalis*, with the aim of maximizing biomass valorization from a sustainability perspective. Additionally, this model will be compared to alternative benchmark biorefinery designs identified in the state-of-the-art analysis.

- What is the environmental footprint of the *F. lumbricalis* biorefinery system?

To quantify the environmental impact of the designed biorefinery, an LCA will be conducted, identifying critical processes and parameters that must be addressed to ensure environmental sustainability.

- What is the economic feasibility of the *F. lumbricalis* biorefinery?

A LCC analysis will be performed to assess the financial viability of the system, evaluating its economic sustainability.

- What are the social implications of implementing an *F. lumbricalis* biorefinery?

An S-LCA will be conducted to analyze the social impacts associated with the biorefinery, considering different scales of implementation within the macroalgae value chain.

- Is the cascade biorefinery the most sustainable approach for *F. lumbricalis* valorization?

The final step involves integrating LCA, LCC, and S-LCA results into a comprehensive sustainability index to determine the most sustainable biorefinery design for *F. lumbricalis*.

Hypothesis of the Thesis

The hypothesis underlying this Thesis is as follows:

*“The development of a cascade biorefinery approach specifically tailored for the red macroalgae *F. lumbricalis* enables a sustainable, circular, and efficient utilization of biomass, ensuring environmental benefits, economic viability, and increased social awareness when compared with other designs.”*

This hypothesis will be tested through a comprehensive evaluation of the proposed cascade biorefinery model, assessing its performance in comparison to alternative biorefinery designs. The benchmarking process will incorporate environmental, economic, and social sustainability criteria, ensuring a holistic assessment of its advantages and limitations. By integrating insights from LCA, LCC, and S-LCA, this research aims to provide a quantitative and multidisciplinary perspective on the feasibility and impact of macroalgae-based biorefineries in the bioeconomy.

Scientific significance of the Doctoral Thesis

Marine biomass, particularly macroalgae, presents a promising yet fully underutilized resource for sustainable biorefinery applications. However, challenges remain in ensuring that such systems are environmentally sound, economically viable, and socially responsible.

This doctoral research positions itself at the intersection of bioresource valorization, LCSA, and circular bioeconomy principles, offering a comprehensive evaluation of a cascade biorefinery approach for the red macroalga *F. lumbricalis* in the Baltic Sea Region. By integrating a full sustainability assessment, this study contributes to advancing macroalgae-based biorefineries as a key component of future sustainable bioeconomic strategies.

The urgent need for standardized sustainability assessments in the biorefinery sector underscores the scientific relevance of this research. While macroalgae-based biorefineries have been explored primarily from a technological perspective, there remains a critical gap in assessing their environmental, economic, and social sustainability through a quantitative, life-cycle-based framework. Moreover, European policies emphasize the necessity of establishing sustainable and scalable macroalgae-based industries. However, current methodologies lack a unified approach to evaluating sustainability comprehensively.

This doctoral study contributes to the scientific advancement of macroalgae biorefineries in three innovative ways:

1. **Integrated sustainability.** This research integrates the three sustainability pillars (i.e., environmental, economic, and social) through a quantitative LCSA tailored to a Baltic Sea Region scenario. By combining LCA, LCC, and S-LCA, it provides a robust framework for evaluating macroalgae biorefineries beyond conventional environmental impact analyses.
2. **Innovation in S-LCA.** The study advances S-LCA methodologies, particularly in the macroalgae sector, where social impacts remain largely unexplored. By investigating

the socio-economic implications of macroalgae-based biorefineries at different scales, this research enhances understanding of their potential role in coastal community development, job creation, and stakeholder engagement, for example.

3. Collaboration impact. This Thesis, as part of the European project TACO ALGAE [30]strengthens multidisciplinary collaborations between research institutions, universities, and industry partners, facilitating knowledge exchange and practical implementation of sustainable biorefinery concepts. Through industry partnerships, it fosters the development of real-world applications that align with European sustainability policies and bioeconomic strategies.

The concepts and outcomes developed and applied in the Thesis have been discussed in peer-reviewed scientific papers and presented at international conferences, highlighting their scientific innovation and significance.

Practical significance of the Doctoral Thesis

The findings of this doctoral research are of critical importance to stakeholders in the macroalgae sector, including industry professionals, policymakers, and researchers working towards sustainable bioeconomic solutions. As the global demand for renewable biomass resources grows, macroalgae biorefineries have the potential to offer environmentally friendly, economically feasible, and socially responsible alternatives to traditional biomass processing. However, their large-scale implementation remains hindered by technical, economic, and regulatory challenges.

This research provides a structured approach to overcoming these challenges by integrating LCSA into cascade biorefinery designs. By applying a holistic sustainability framework, the study delivers quantitative insights that inform decision-making across the macroalgae industry, policy landscape, and academia.

The practical relevance of this research is particularly reflected in three key areas:

1. Industry application. The study delivers technical insights that are directly applicable to macroalgae-sector stakeholders, including biorefinery operators, investors, and policymakers. By evaluating various biorefinery designs, this research provides a benchmark for best practices, guiding stakeholders in selecting the most efficient, sustainable, and scalable approaches for *F. lumbricalis* valorization.
2. Feasibility analysis. The research identifies the key challenges affecting the environmental and economic viability of macroalgae biorefineries. Through comparative analysis and sustainability benchmarking, this study pinpoints bottlenecks in processing, supply chain limitations, and financial constraints that must be addressed to ensure long-term feasibility. Additionally, the integration of LCC highlights cost-effective strategies for reducing economic risks in commercial implementation.
3. Awareness & LCT. By applying LCT, this research promotes a sustainability-driven perspective on macroalgae utilization, emphasizing the importance of evaluating biorefinery systems beyond mere economic performance. The incorporation of S-LCA raises awareness about the socioeconomic implications of macroalgae biorefineries,

fostering discussions on equitable resource management, local employment opportunities, and industry-wide collaboration.

Thus, this Thesis bridges the gap between scientific advancements and industrial applications, offering a data-driven framework that can be directly integrated into macroalgae valorization strategies at regional, national, and European levels. The methodologies developed in this study are transferable to broader bioeconomy sectors, supporting policy development, investment strategies, and sustainable innovation in marine biotechnology.

By addressing technical, economic, and social considerations, this thesis provides a practical roadmap for guiding the transition toward sustainable macroalgae-based biorefineries, contributing to a circular bioeconomy and the broader goals of the EU Green Deal.

Research methodology

The structure of the applied research framework is illustrated in Fig. 1. The methodology integrates both qualitative and quantitative research techniques, including literature review, data collection and analysis, experimental data acquisition, case study definition, software modeling, and statistical analysis. The research follows a structured, stepwise approach to ensure a comprehensive and systematic evaluation of macroalgae biorefinery systems.

1. Step 1: Literature Review and Biorefinery System Definition.

The first phase involves an extensive literature review to analyze the current landscape of macroalgae biorefinery systems, identifying key limitations and opportunities within the sector. This step aims to establish a comprehensive knowledge base for designing an optimal biorefinery system. Based on insights from literature and input from project partners within the TACO ALGAE project, a cascade biorefinery model for *F. lumbricalis* will be formulated. Additionally, alternative benchmark biorefinery designs will be outlined to enable comparative analysis.

2. Step 2: Data Collection and Environmental & Economic Assessment.

In this phase, a direct data collection strategy is implemented, incorporating primary data from the TACO ALGAE project. In cases of data gaps, sectoral and literature data will be utilized to ensure completeness. Once the inventory analysis is established, covering key parameters such as material inputs, energy consumption, waste generation, and emissions, the LCA will be conducted. This analysis will initially focus on evaluating the environmental impact of different macroalgae harvesting and cultivation techniques, which are critical in determining the sustainability of biomass preparation operations. Subsequently, the environmental impact of various biorefinery designs proposed in Step 1 will be assessed.

Following the LCA, the LCC analysis will be conducted by assigning monetary values to inventory data, allowing for an economic comparison of the different biorefinery configurations.

3. Step 3: S-LCA Development and Analysis.

Since S-LCA methodologies are significantly underdeveloped for macroalgae systems, this step begins with the definition of a tailored S-LCA framework. Two levels of social analysis will be performed. i) Value Chain-Level Analysis, identifying social hotspots across the macroalgae production system, ii) Community & Company-Level Analysis, assessing the potential social impacts at a localized scale, focusing on small communities and businesses involved in macroalgae biorefining.

4. Step 4: LCSA and Final Sustainability Index.

In the final phase, a comprehensive LCSA will be performed by integrating the results from Step 2 (LCA & LCC) and Step 3 (S-LCA). A multicriteria decision analysis (TOPSIS methodology) will be applied to calculate a final sustainability index, enabling a quantitative comparison of the proposed biorefinery designs. This approach will determine the most sustainable configuration for an *F. lumbricalis* biorefinery based on environmental, economic, and social sustainability indicators.

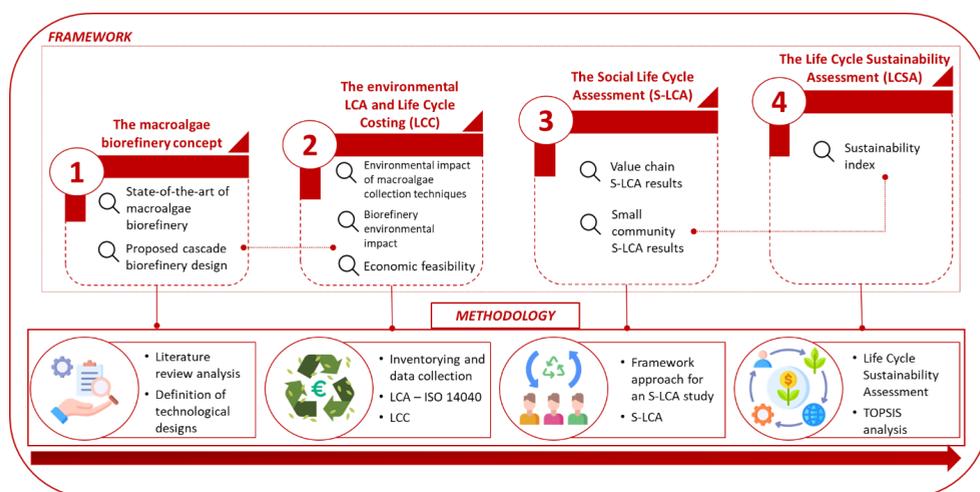


Fig. 1. The research methodology of the Doctoral Thesis.

Approbation of the research results

The results of the author's research have been presented and discussed in 4 scientific conferences and published in 5 peer-reviewed scientific journals.

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Structure of the Doctoral Thesis

This Doctoral Thesis is written in English and consists of an introduction, three main chapters, conclusions and recommendations, references, and two annexes.

The Introduction presents the overall context and relevance of the research, outlines the objectives and methodology, and highlights the significance of the study's findings.

Chapter one provides a state-of-the-art review of the macroalgae biorefinery sector, identifying current opportunities and challenges. It also includes a literature review on the application of LCA to macroalgae systems, with a focus on sustainability aspects and end-use applications of macroalgae-derived products.

Chapter two introduces the technological design of the proposed *F. lumbricalis* cascade biorefinery, alongside two alternative system configurations: the single-product extraction and the three-line extraction designs. The chapter also details the methodological frameworks used to assess the environmental, economic, and social sustainability dimensions through the application of LCA, LCC, and S-LCA. It concludes with the application of the TOPSIS multi-criteria decision-making method to calculate a composite sustainability index within the LCSA framework.

Chapter three presents the results of the sustainability assessments (LCA, LCC, and S-LCA) across the different biorefinery designs and evaluates them through sensitivity

analyses. It also discusses the outcomes of the TOPSIS analysis, identifying the most sustainable design among the proposed scenarios.

The Conclusions and Recommendations chapter summarizes the key findings of the research and offers strategic guidance for future developments in the sector.

The full Doctoral Thesis comprises 185 pages, including 50 tables, 57 figures, 2 annexes, and 235 references.

1. STATE-OF-THE-ART ANALYSIS

1.1. Framework for the literature review

To define the state-of-the-art in macroalgae biorefinery, a systematic literature review was conducted using the Scopus database to assess developments in macroalgae-based biorefineries and their associated LCA studies from the period of time 2012-2024. To ensure a transparent and reproducible methodology, the review followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines [31].

The research framework, illustrated in Fig.1.1, is structured into two primary investigative pathways, with searches performed within article titles, abstracts, and keywords. The first pathway focused on identifying publications specifically related to macroalgae biorefineries, using the search string:

“(biorefinery AND seaweed OR macroalgae OR macroalga) AND NOT (life AND cycle AND assessment) AND NOT (micro)”

This search was intentionally designed to exclude LCA-related studies while filtering out microalgae-based research to maintain a focus on macroalgae applications.

The second pathway examined the integration of LCA within macroalgae biorefineries, employing the search string:

“(biorefinery AND life AND cycle AND assessment) AND (seaweed OR macroalgae OR macroalga) AND NOT (micro)”

As in the previous case, microalgae-focused publications were removed to refine the dataset.

A PRISMA flowchart (Fig. 1.1) details the selection process, including the application of inclusion and exclusion criteria. To enhance methodological transparency, an annex provides a comprehensive list of selected studies, along with a breakdown of exclusion criteria applied during data refinement.

The selected studies were analyzed to highlight key aspects of macroalgal biorefinery systems. The research categorized macroalgae species into brown, green, and red groups and explored the range of products extracted through different biorefinery strategies. Additionally, the processing technologies employed were assessed based on their efficiency, limitations, and yield potential.

A crucial component of this review involved evaluating the Technological Readiness Level (TRL) of various biorefinery configurations, providing insight into their development stage and potential for commercial scalability. The integration of LCA findings enabled a techno-environmental evaluation, allowing for the identification of major trends, bottlenecks, and research gaps within the field. This holistic perspective is essential for optimizing macroalgal biorefinery processes, ultimately enhancing their sustainability and economic viability.

It is important to note that two studies classified under the general macroalgae biorefinery category also incorporated LCA analyses. However, these studies were not initially captured within the LCA-specific search category and, therefore, were not included

in the total count of 16 LCA studies explicitly reported. Despite this classification distinction, the LCA results from these two studies have been fully considered and integrated into the discussion, ensuring that their environmental insights contribute to the overall assessment of macroalgae biorefinery sustainability.

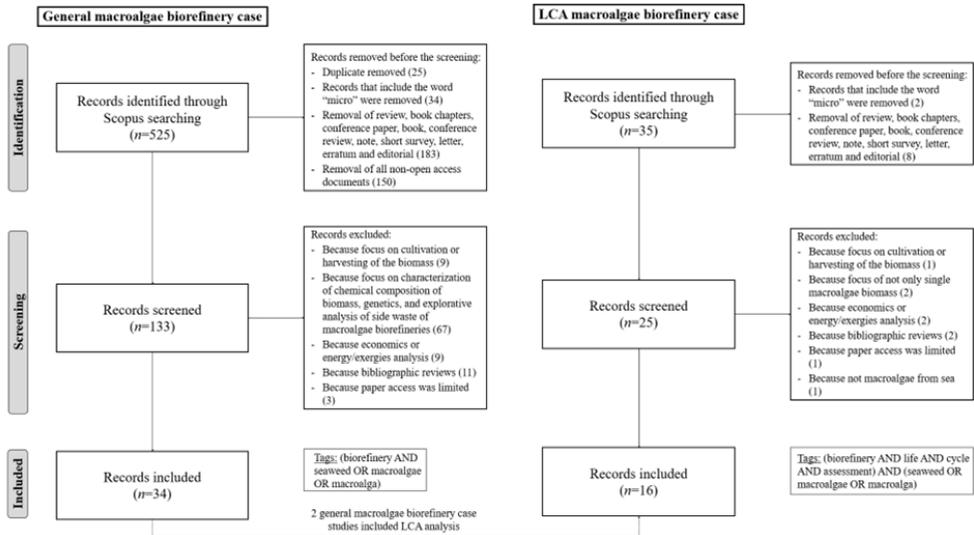


Fig. 1.1. Methodological framework. Flow information through different phases of the systematic literature search. Diagram generated according to the PRISMA statement.

1.2. The macroalgae biorefinery system

This section provides a comprehensive overview of the macroalgae biorefinery system, detailing its structure, processes, and potential applications. It begins by introducing the fundamental concept of a macroalgae biorefinery, emphasizing its role in transforming macroalgal biomass into a diverse range of high-value products. This broad perspective lays the foundation for understanding the various biorefinery pathways and their significance in sustainable resource utilization.

The biorefinery concept is formally defined as *"the sustainable processing of biomass into a spectrum of marketable products and energy"* [32]. Similar to conventional petroleum refineries, which produce various fuels and byproducts from crude oil, biorefineries operate using renewable biological feedstocks such as macroalgae, agricultural residues, forestry byproducts, and aquaculture waste [33]. These systems incorporate multiple processing technologies, ranging from biomass collection to product extraction and refinement.

Developing a macroalgae biorefinery requires careful attention to several critical factors, including site selection, process integration, and environmental compatibility. The efficiency and sustainability of both upstream (biomass cultivation and harvesting) and downstream (processing and product development) operations depend on a well-planned system that aligns with natural ecological conditions. These considerations are essential to

minimize environmental impacts and maximize the economic and social benefits of macroalgae-based biorefinery projects [34].

A key aspect of sustainable biorefinery implementation is evaluating its impact on local ecosystems, biodiversity, and socio-economic conditions. A thorough assessment of these factors ensures that macroalgae-based industries contribute positively to both the environment and communities engaged in the biorefinery value chain.

Fig. 1.2 illustrates a conceptual model of a macroalgae biorefinery, showcasing the main cultivation and harvesting techniques applied in the upstream phase. It should be noted that not all possible downstream extraction pathways are included in the figure, as these vary significantly depending on parameters such as the targeted products and the macroalgae species used. To maintain a clear and accessible overview of a general biorefinery system, the diagram has been intentionally simplified. A more detailed explanation of the downstream extraction processes is provided in Chapter 1.4.

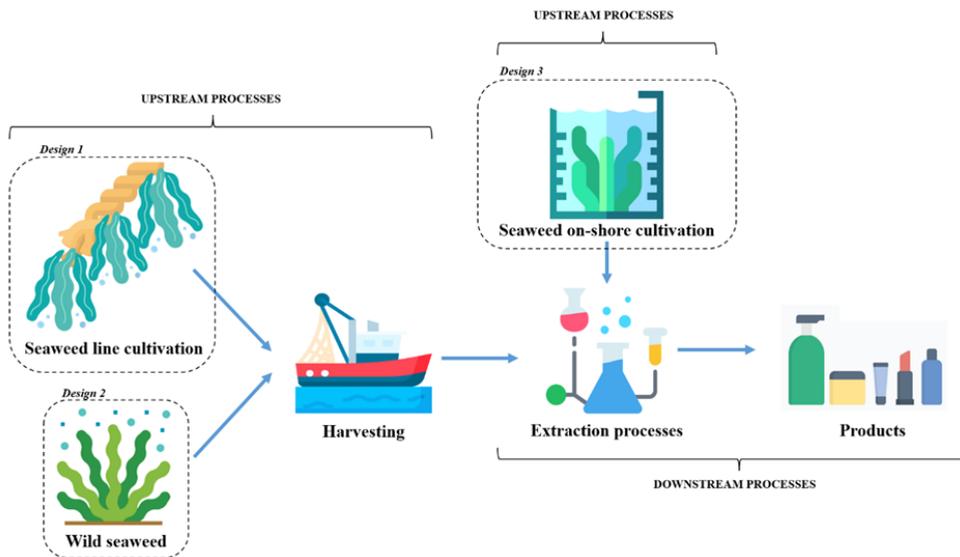


Fig. 1.2. Schematic representation of a hypothetical macroalgae biorefinery.

Three primary methods can be adopted in the macroalgae biomass gathering phase:

1. Cultivation on an artificial substrate (Off-shore systems)

In this case, macroalgae grow on artificial substrates, such as seeded lines made from ropes or nylon nets [35], which are deployed in open marine environments [36]. This technique has been widely used for species such as *Laminaria digitata* and *Saccharina latissima* [37]. Cultivation may involve direct seeding onto these substrates or transplanting juvenile macroalgae from controlled indoor facilities [34]. Integrating these macroalgae farms with offshore wind energy infrastructure can provide logistical and economic advantages, reducing resource costs and improving marine space utilization [38].

2. Wild macroalgae harvesting

This approach is based on wild specimen harvesting, which involves the direct collection of naturally occurring macroalgae stocks, particularly in the Atlantic region of Europe [5]. Biomass is gathered using mechanized vessels or through manual harvesting, depending on species type and geographic location [39]. The sustainability of this approach depends on managing seasonal biomass fluctuations and complying with national regulations on harvesting limits and permits [5], [40].

3. On-shore cultivation (land-based systems)

It consists of on-shore cultivation systems, similar to the more well-developed microalgae ponds, which are grown in controlled environments such as open raceways or land-based seawater tanks [41]. This approach is commonly used for green algae species such as *Ulva spp.* and enables precise control over growth conditions and productivity [42]. While this method allows optimization of biomass yield, it may compete with land availability for other agricultural or industrial applications.

The downstream phase of a macroalgae biorefinery involves converting harvested biomass into value-added products. This requires targeted scientific research and technological development to optimize extraction efficiency and sustainability. The choice of processing methods is influenced by several factors [34], including:

- Macroalgae species and composition;
- Extraction technologies and product yield;
- TRL for industrial scalability;
- Environmental, economic, and social considerations;

Currently, macroalgae valorization has been largely dominated by single-product extraction processes, particularly for biofuel production. These include biomethane production [43], bioethanol generation [44], and butanol extraction [45].

However, such approaches often result in the underutilization of valuable macroalgal components, making them economically and environmentally unsustainable. To address this limitation, cascade biorefinery models have been proposed as a more holistic and efficient strategy.

The cascade biorefinery approach involves [46]:

1. Fractionating macroalgal biomass into its primary macromolecular components (e.g., carbohydrates, proteins, minerals).
2. Converting these components into multiple high-value products rather than focusing on a single output.
3. Enhancing resource efficiency and economic viability through integrated product recovery.

This multi-product extraction strategy is increasingly recognized as the most promising and sustainable method for macroalgae biorefineries [47]. By maximizing biomass utilization and integrating advanced extraction techniques, cascade biorefineries can significantly improve the economic profitability and environmental sustainability of macroalgae-based industries.

1.3. Applicative sectors for macroalgae-based products

Valuable compounds extracted from macroalgae have demonstrated promising applications across various industries, contributing to advancements in biotechnology, pharmaceuticals, food, agriculture, and sustainable materials. Several successful examples highlight the diverse potential of macroalgae-derived products in commercial and industrial sectors.

Food

The incorporation of macroalgae into the food industry is driven by their direct edibility and high nutritional value [48]. Their increasing popularity, particularly in Europe, is linked to their distinctive flavors and perceived uniqueness, making them especially appealing to consumers interested in healthy, plant-based, and minimally processed diets [49]. Macroalgae offer remarkable culinary flexibility, as they can be consumed fresh, incorporated as powdered supplements, or undergo fermentation processes [50]. They are also used in the production of snacks, such as seaweed-based crackers, and are integrated into beverages, including soft drinks and beers [51]. Additionally, their nutritional properties make them a valuable ingredient when combined with traditional cereal-based products, enhancing their overall dietary benefits [52].

Horticulture and crop application

Throughout history, macroalgae have been recognized for their effectiveness as natural fertilizers, primarily due to their high mineral content, which supplies essential nutrients, and their fibrous composition, which enhances soil moisture retention and overall soil structure. The application of macroalgae-based fertilizers varies, with forms ranging from granular and powdered formulations to liquid extracts [53]. One widely used method involves mechanically pressing fresh algae to obtain a nutrient-rich liquid fertilizer, commonly referred to as "sap" [51]. This extract contains a high concentration of micronutrients, amino acids, vitamins, and plant growth-stimulating compounds [54].

Beyond their fertilizing properties, macroalgae-based formulations have demonstrated biofungicidal activity, contributing to plant protection and disease resistance [55]. For instance, foliar application of *Kappaphycus alvarezii* sap at a 5% concentration has been shown to increase tomato productivity by 61 % [56]. Similarly, the application of liquid extracts from *Kappaphycus* and *Gracilaria* on green bean plants (*Vigna radiata*) significantly enhances protein and mineral content, improving the overall quality of the produce [57]. Macroalgae-derived fertilizers have also been reported to positively influence aromatic plant cultivation, increasing the concentrations of sesquiterpenes and monoterpenes in rosemary, basil, and mint, thereby improving the composition of their essential oils [58], [59].

The integration of macroalgae-derived fertilizers within cascade biorefinery systems represents a sustainable approach to waste valorization, ensuring that residual biomass from biorefinery operations is effectively repurposed in agriculture. However, careful monitoring of toxic metal accumulation is essential, as macroalgae-derived fertilizers may undergo

chemical changes over time, influenced by factors such as oxidation, which could alter their physical and chemical properties [60].

Feed

The use of macroalgae as animal feed has a long history, primarily due to their high concentrations of essential minerals, vitamins, and fibers, which contribute to the growth and overall health of livestock [61]. Incorporating *Ulva* species into animal diets has demonstrated significant benefits, including enhanced poultry health, improved eggshell strength, and more intense yolk pigmentation [62]. Additionally, *Ulva* has exhibited antimicrobial properties, effectively inhibiting the proliferation of bacteria, fungi, and viruses, including the Newcastle disease virus [63], [64].

Brown macroalgae have also been linked to better weight gain and lower mortality rates in poultry exposed to *Salmonella enteritidis* infections [65]. Similarly, red macroalgae have been shown to increase both egg production and quality in quails, further reinforcing their nutritional potential in poultry farming [66]. In cattle breeding, supplementing the diet of pregnant cows with 10 % *Undaria pinnatifida* (brown macroalgae) for two months has resulted in the birth of healthier and heavier calves [67]. Additionally, a combination of brown macroalgae, including kelp and fucus, has significantly improved the digestibility of dry matter, organic matter, and crude protein, optimizing nutrient absorption in livestock [51].

Beyond ruminant nutrition, macroalgae-derived compounds have also shown promise in swine diets. Extracts from brown macroalgae, particularly alginate oligosaccharides, have been used as antibacterial agents against pathogens such as *Escherichia coli* [68] and as prebiotics, enhancing gut health and immune response [69]. These findings underscore the multifaceted benefits of macroalgae as a feed supplement, demonstrating their potential to enhance animal health, improve productivity, and reduce the need for synthetic additives across different livestock species.

Bioactive compounds

To withstand challenging environmental conditions, macroalgae have developed a diverse array of bioactive compounds that hold substantial value for the food, cosmetic, and pharmaceutical industries [70]. These compounds exhibit various biological activities, including antioxidant, antimicrobial, anti-inflammatory, and antiviral properties, making them highly sought after for commercial applications. A comprehensive overview of these bioactive substances, categorized by macroalgae family and their specific functional properties, is provided in Table 1.1.

Table 1.1

List of some of the macroalgae bioactive compounds.

Types of macroalgae	Bioactive compounds	Bioactive effects	References
<i>Laminaria</i> (brown)	Sulfate ester	Tumor-inhibiting and anticoagulant agent	[71]
	Laminarin	Antibacterial and a boost for the immune system	[51]

Types of macroalgae	Bioactive compounds	Bioactive effects	References
<i>M. nitidum</i> (green), <i>L. pinnatifida</i> (brown) <i>P. yezoensis</i> (red)	Polysaccharides	Antihypertensive and antihyperlipidemic effects	[72]
<i>Ulva spp.</i> (green)	Fatty acids	Anti-inflammatory action	[73]
<i>H banksia</i> (brown)	Polyphenols	Antioxidant and anti-inflammatory effects	[74]
Brown macroalgae (general)	Phloroglucinol, fucoxanthin, and fucoidan	Antitumoral for breast cancer	[75]
	Polyphenols	Protection against cardiovascular diseases and diabetes	[76]
Red macroalgae (general)	Phycobiliproteins	Natural colorant for food and cosmetics	[77]
		Antimicrobial, antioxidant, anti-inflammatory, and neuroprotective action	[78]
<i>Ulva spp.</i>	Acidic	Used in cosmetic formulation	[79]
<i>E. cottonii</i> (red)	Phlorotannins	Natural tyrosinase inhibitor for skin whitening	[80]
<i>E. cava</i> (brown)	Phlorotannins	Healing skin inflammation	[81]

Biofuels and bioenergy

Biofuels, available in solid, liquid, and gaseous forms, represent a promising alternative for automotive and industrial energy applications, utilizing biological feedstocks. Among these, macroalgae serve as a particularly versatile raw material for biofuel production through a range of biochemical and thermochemical conversion pathways, including anaerobic digestion, fermentation, gasification, purification, transesterification, hydroprocessing, and pyrolysis/liquefaction [82].

One of the most immediate and viable applications of macroalgae in biofuel production is biogas generation, largely due to their favorable carbon-to-nitrogen ratio, low lipid content, and absence of lignin, which facilitates their integration into biorefinery processes [83]. In the context of bioethanol production, macroalgae are considered a promising feedstock due to their relatively low lignocellulosic content. However, the high moisture content of macroalgae presents a major challenge, as it significantly impacts production efficiency. The need to reduce moisture levels from approximately 90 WW% to 5 WW% before processing remains a key limitation for large-scale thermochemical applications, directly affecting overall conversion yields [84]. While bioethanol yields from macroalgae can be comparable to those of sugarcane (29.6 kg/t raw material), they remain lower than those obtained from barley, wheat, and rice [85].

Further research is required to evaluate the industrial feasibility of biobutanol production, as large-scale deployment remains limited by both technical and economic constraints [83]. Additionally, several compositional challenges hinder the viability of macroalgae as a feedstock for bio-oil and biodiesel production, including high moisture, nitrogen, and ash content, as well as the presence of metals and relatively low lipid levels [83]. These factors significantly impact the efficiency and cost-effectiveness of biofuel extraction and processing, making macroalgae less competitive for certain biofuel applications.

Beyond technological challenges, economic barriers remain a major obstacle to scaling up macroalgae-based biofuel production, particularly in relation to the cost of large-scale cultivation, harvesting, and transportation. Addressing these limitations through process optimization and supply chain improvements is essential to ensuring a consistent, low-cost biomass supply, making macroalgae a more viable and sustainable resource for biofuel production [84].

1.4. Technological exploitations of macroalgae's valuable products

The initial findings from the literature review (first pathway) emphasize the technological advancements in macroalgae biorefineries. Consequently, this chapter examines the most relevant technological development in macroalgae-based biorefineries, as identified through the literature review analysis. To ensure a structured and coherent presentation, the discussion is organized based on macroalgae families (i.e., brown, green, and red), allowing for a clearer comparison of their biorefinery applications and potential exploitations.

Brown macroalgae

In Fig. 1.3 are reported the possible valuable products obtainable from brown macroalgae.

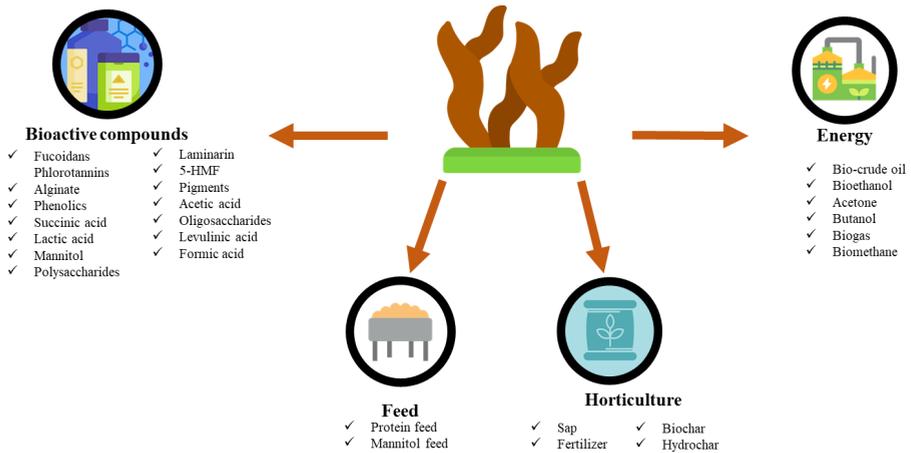


Fig. 1.3. List of possible recoverable products from brown macroalgae.

Considering the macro-families of products:

- Bioactive compounds

Among the most extensively studied bioactive compounds, fucoidans have gained significant attention for their biological properties and wide-ranging applications. Enzyme-assisted extraction (EAE) has proven to be the most effective method, yielding up to 18 % DW in *Ecklonia maxima* [86]. Hydrothermal-assisted extraction (HAE) also demonstrates high efficiency, with optimal conditions (120 °C, 80.9 minutes, 12.02 mL/g solvent)

resulting in 2,782.3 mg fucoidan per 100 g DW [87]. In *Sargassum muticum*, autohydrolysis significantly enhances recovery, increasing fucoidan yield from 0.20 g/g in raw biomass to 0.32 g/g in pressed samples [88].

Alginate, another widely studied compound, is traditionally extracted through sequential acid-alkaline treatment. The highest purity (88.98 %) and yield (65.13 % DW) have been achieved using hydrodynamic cavitation (HDC) in *Alaria esculenta* [89]. Ultrasound-assisted extraction (UAE) has been found to enhance efficiency, while autohydrolysis, despite being a solvent-free alternative, results in lower yields, ranging from 0.01 to 0.03 g/g in *S. muticum* [88].

Phenolic compounds are efficiently extracted using microwave-assisted extraction (MAE), which offers both high yield and energy efficiency. In *S. muticum*, MAE using water at 180 °C for 11 minutes yields between 1.59 and 2.85 g gallic acid equivalents (GAE) per liter [90]. Ethanol-based extraction has demonstrated the highest total phenolic content (TPC), reaching 22 g GAE per kg volatile solids (VS) in *Rugulopteryx okamurae* [91].

Laminarin, a polysaccharide of commercial interest, is best extracted through HCl-assisted extraction, with *L. digitata* yielding up to 55.8 % DW [87]. Similarly, *S. latissima* achieves a yield of 44.6 % using the same method, while UAE, in combination with acid treatment, further improves efficiency. Other polysaccharides, such as sulfated fucoidan polysaccharides (SFPS), benefit from UAE, which enhances release while minimizing degradation. Optimal conditions in *Sargassum fusiforme* yield 15.30 wt% SFPS [92].

Oligosaccharides, another valuable class of bioactive molecules, are effectively recovered through MAE and HAE. In *Undaria pinnatifida*, distilled water extraction at 120–220 °C for five minutes has been found to maximize yield [93], while in *S. muticum*, HAE at 160 °C achieves a peak fuco-oligosaccharide concentration of 4.17 g/L [90].

The extraction of pigments from macroalgae requires specialized methods to optimize purity and recovery. In *S. latissima*, a combination of solid-liquid extraction and liquid-liquid separation yields 4.93 mg chlorophyll per g DW and 1,956 µg fucoxanthin per g DW [94]. Ethanol extraction is a suitable alternative for chlorophyll-rich species, such as *Fucus spiralis*, where the yield reaches 13.03 mg/g DW [95].

Hydrothermal carbonization (HTC) is the preferred method for obtaining organic acids from macroalgae. In *L. digitata*, treatment at 200 °C with 4 % sulfuric acid results in 12.5 % levulinic acid and 6.9–17.7 % formic acid [96]. Bio-succinic acid is produced via fermentation, with a maximum yield of 0.73 g/g sugars, though *S. japonica* is not considered a viable feedstock due to high production costs and hydrolysis inefficiencies [97].

Mannitol extraction is optimized using MAE applied to *S. latissima* solid residues. Under optimal conditions (190–210 °C), recovery reaches 3.62–4.15 % DW [98]. Similarly, phlorotannin extraction employs sequential liquid extraction techniques, with the highest concentration detected in *S. latissima* extracts, reaching 0.48 mg phloroglucinol equivalents per mg extract [99].

- Feed and horticulture

Brown Macroalgae-derived proteins are increasingly being explored as sustainable feed supplements. The most effective extraction methods include MAE, which increases the protein content of *U. pinnatifida* solid residues to 28.7 % at 200 °C [93]. Microwave hydrothermal treatment (MHT) in *S. muticum* achieves protein recovery rates of 69–93 %, preserving the integrity of most proteins [90]. HDC in *A. esculenta* also enhances protein yield, increasing the final concentration to 17.19 % after the removal of laminarin and alginate [89].

Fertilizer production from brown macroalgae follows various pathways, with liquid fertilizers derived from *S. muticum* containing 199 g/L of dissolved compounds [88]. Hydrothermal liquefaction (HTL) of wastewater from *S. latissima* provides a nitrogen recovery rate of 73 kg NH₃ per 1,000 kg DW [36]. Hydrochar production through HTC has also been optimized, with *L. digitata* processed at 200 °C with 4% sulfuric acid, yielding a carbon content of 65.4 % and a higher heating value (HHV) of 25.5 MJ/kg [96].

- Energy applications

Bioethanol production from brown macroalgae is a widely studied application, with efficiency largely dependent on pretreatment and fermentation strategies. MHT has been identified as an energy-efficient approach, operating at 160–180 °C to maximize sugar release while minimizing energy consumption [90], [100]. Pre-saccharification and simultaneous fermentation (PSSF) achieves a conversion efficiency of 76.23 %, yielding 18.14 g/L ethanol in 12 hours [101]. Following MHT, ethanol yields can reach 96 %, with a final concentration of 20.5 g/L [90]. In *S. muticum*, autohydrolysis results in approximately 80 % ethanol yield with reduced processing times [100].

Biocrude production is effectively achieved through HTL, with *S. fusiforme* processed at 280 °C for 45 minutes in 80 % ethanol, achieving a biocrude yield of 27.98 DW% [92]. The use of ethanol significantly enhances carbon content (69.83 %) and HHV (33.42 MJ/kg), improving fuel properties compared to water-based extractions.

Acetone-butanol-ethanol (ABE) fermentation has also been explored, with *Clostridium acetobutylicum* utilized to process hydrolysates from *S. latissima*. Demineralization significantly improves performance, reducing the fermentation lag phase from 72–144 hours to 24–48 hours, increasing ABE yield from 0.07 to 0.17 g ABE/g sugar [102].

Green macroalgae

In Fig. 1.4 are reported the possible valuable products obtainable from green macroalgae.

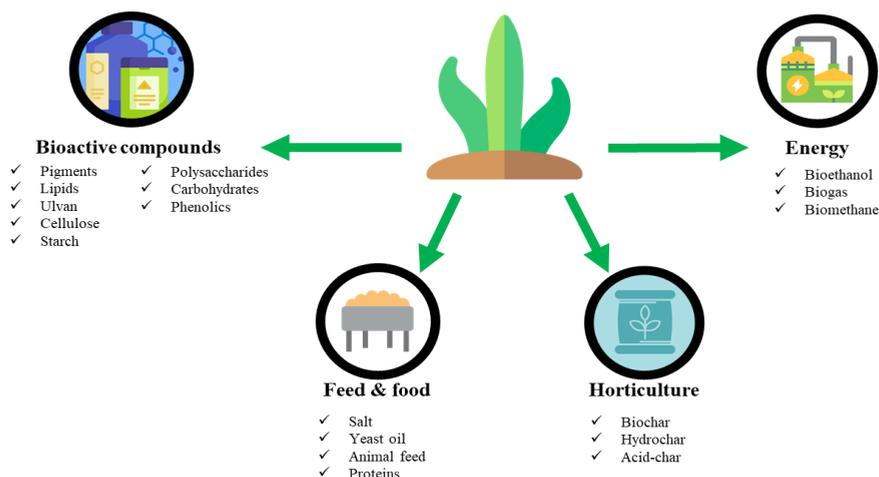


Fig. 1.4. List of possible recoverable products from green macroalgae.

Considering the macro-families of valuable products:

- Bioactive compounds

Ulvan, a key bioactive polysaccharide in *Ulva spp.*, has been extracted using various techniques, with pretreatment significantly improving recovery. Aqueous salt extraction at 40 °C for 30 minutes removes salts and enhances subsequent HCl extraction yields [103]. Pigment removal with ethanol prior to ulvan recovery has also been reported to improve extract quality [104]. Multiple extraction methods have been applied for ulvan recovery. Autohydrolysis, a non-isothermal process, heats wet biomass without chemical reagents but is less effective than HCl extraction, which suspends biomass in 0.05 M HCl, followed by heating, filtration, and freeze-drying, yielding 3.7–8.2 % DW in *Ulva ohnoi* [103]. Hydrothermal extraction followed by ethanol precipitation achieves a 23 % ulvan yield, whereas choline chloride precipitation results in a 17.8 % yield. Conventional acidic extraction (0.01 M HCl, 92 °C, 4 h) followed by ethanol precipitation provides the highest recovery at 30.4 % [104]. Different techniques influence the molecular weight of the extracted ulvan; HCl extraction leads to depolymerization, producing lower molecular weights (10.5–16.3 kDa), while Na₂C₂O₄ extraction preserves higher molecular weights (219–312 kDa) [104]. A cascading biorefinery approach has been proposed for *Ulva fasciata*, integrating ulvan extraction with multiple co-product recoveries. Ethanol extraction (1:20 w/v) recovers chlorophyll a (24.59 mg/g), chlorophyll b (1.55 mg/g), carotenoids (4.05 mg/g), total chlorophyll (2.61 % DW), and total carotenoids (0.41 % DW). The biomass residue, after homogenization and centrifugation, yields a mineral-rich extract (34.89 % DW). Starch is recovered at 12.55 % DW via ethanol extraction, while Soxhlet extraction (chloroform/methanol, 50 °C) yields 3.27 % lipid. Ulvan extraction via chilled isopropanol precipitation (1:2.5 v/v) results in 22.24 % DW, followed by protein recovery (13.37 % DW) through NaOH treatment (60 °C, 10 h). Final acid hydrolysis (HCl, 100 °C) enables cellulose recovery (10.66 % DW) [105].

Carbohydrates in *Ulva spp.* have been successfully extracted using hot water treatment, enzymatic hydrolysis, and ionic liquid (IL)-assisted techniques. Hot water extraction combined with enzymatic hydrolysis yields a sugar-rich hydrolysate with 38.8 g L⁻¹ of sugars, primarily glucose, rhamnose, and xylose. Aqueous extraction at 150 °C for 10 min solubilizes 59.6 % of total sugars, with enzymatic hydrolysis further increasing recovery to 78.4 % for glucose, 53.7 % for rhamnose, and 66.7 % for xylose, resulting in final hydrolysate concentrations of 22 g L⁻¹ glucose, 12 g L⁻¹ rhamnose, and 4.8 g L⁻¹ xylose [106]. IL-assisted extraction has been applied to *Ulva lactuca* using mechanical shear and two-phase partitioning, demonstrating an alternative fractionation method that allows for IL recovery. Using [Emim] [DBP] as the extraction medium, carbohydrate yields reach 30.7 %, proving ILs to be a viable approach for the selective separation of proteins and carbohydrates [107]. Physical extraction methods, including osmotic shock, pulsed electric field (PEF), and high shear homogenization (HSH), have also been explored. Osmotic shock at 30 °C for 24 hours achieves the highest carbohydrate yield (44.7 %), while PEF treatment (0.05 ms pulse, 7.5 kV cm⁻¹ field strength) improves cell membrane permeabilization, enhancing sugar release. HSH extraction, applied at 21 m/s rotor speed, results in a 68 % carbohydrate yield, though excessive rotor speeds reduce efficiency at lower biomass concentrations [108].

Phenolic compounds exhibit antioxidant, anti-inflammatory, and cytotoxic properties, making their extraction an essential process in biorefinery applications. Solvent-based extraction using n-hexane, ethyl acetate, ethanol, and hydro-ethanol (75:25) has been evaluated. Hydro-ethanol and ethanol extracts exhibit anti-inflammatory properties, reducing TNF- α levels, while ethyl acetate extracts (Cd4) display the highest antioxidant activity (ORAC assay) [109]. Phenolic content remains stable up to 180 °C but increases to 1.2 % at 200 °C following hydrothermal treatment. Supercritical CO₂ (scCO₂) extraction initially yields <0.5 % DW, but the addition of 10 % ethanol as a modifier increases the phenolic yield to 1.7 % DW with 4 % purity. Further purification through non-ionic polymeric resin adsorption and ethanol desorption results in a phenolic extract of 42.86 % purity, significantly improving antioxidant and antiradical properties [104].

- Feed and horticulture

Protein extraction from *U. lactuca* has been optimized using various techniques. Alkaline solubilization provides the highest yield (36 %), as NaOH facilitates protein release above its isoelectric point. Aqueous two-phase extraction (ATPE) selectively partitions proteins into a PEG-rich phase, separating them from carbohydrates. IL-assisted extraction using [Emim] [DBP] demonstrates high selectivity, yielding 80.4 % protein, while ultrafiltration achieves 64.6 % recovery [107]. Alternative mechanical techniques include osmotic shock (19.5 % yield), enzymatic disintegration (26.1 %), PEF (15.1 %), and HSH (39.0 %), with HSH being the most efficient [108].

Feed applications of *U. lactuca* involve hot water pretreatment and enzymatic hydrolysis, producing a solid fraction suitable for monogastric nutrition [106]. Selective enzymatic liquefaction achieves 59.3 % fermentable sugars in *U. lactuca*, respectively.

Yeast biomass production using unprocessed hydrolysate supernatant yields 44.8 g L⁻¹ with 37.1 % lipid content, providing a high-performance feed additive [110].

Biochar and hydrochar have been produced from *Codium sp.* through pyrolysis, HTC, and acid-mediated carbonization (AMC). Pyrochar, produced via slow pyrolysis at 400 °C for 1 hour, achieves a 44 % yield, while hydrochar produced via HTC at 170 °C for 24 hours yields 28 %. AMC, using H₂SO₄ digestion (50 °C, 15 min) followed by reflux heating (90 °C, 6 hours), results in 18 % acid-char yield. Carbon content varies across methods, with pyrochar containing 56–75 % carbon, hydrochar 22–30 %, and acid-char 10 %. Pyrochar has the highest ash content (51 %), whereas hydrochar (14 %) and acid-char (10 %) contain significantly less ash, offering distinct material properties for various applications [109].

- Energy applications

Biogas production from *Ulva spp.* is enhanced through thermal pretreatment, which increases biochemical methane potential (BMP) by 70–112 %. Methane yield reaches 60 mL CH₄/g VS for raw biomass, with Ulva-derived biogas containing 68 % methane, compared to 62 % in cattle manure-derived biogas, leading to a better energy density of 22 MJ/m³ [111].

Bioethanol production from *U. rigida* has been evaluated using commercial hydrolytic enzymes and yeast strains. Among these, *Saccharomyces cerevisiae* exhibited the highest ethanol yield, reaching 9.35 g L⁻¹ with an 80.78 % conversion efficiency in 24 hours. The highest ethanol productivity (0.40 g L⁻¹ h⁻¹) was achieved with a 1 % inoculum, demonstrating feasibility for industrial-scale production [112].

Red macroalgae

In Fig. 1.5 are reported the possible valuable products obtainable from red macroalgae.

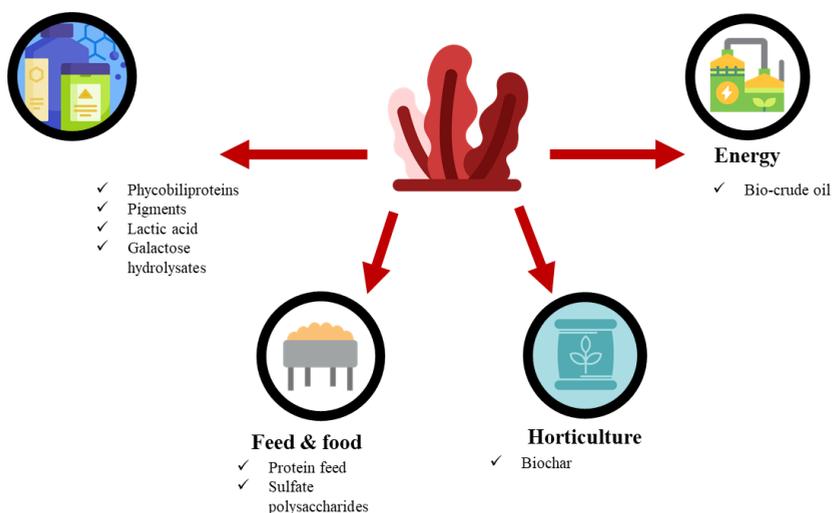


Fig. 1.5. List of possible recoverable products from red macroalgae.

Considering the macro-families of valuable products:

- Bioactive compounds

Carrageenan, a major polysaccharide from red macroalgae, is extracted through EAE, MAE, and conventional alkaline treatments. EAE, tested on *Sarcodiotheca filiformis*, employs Protamex® protease at 50 °C for 3 hours, followed by filtration and phase separation. The enzyme is either denatured at 85 °C (condition a) or left active (condition b), yielding two different extract fractions [113]. MAE applied to *S. filiformis* after lipid removal, involves rehydration overnight, microwave processing at 159 kPa, and precipitation with Cetavlon, achieving yields of 17.1 %–29.7 %, comparable to direct MAE extraction [114].

Phycobiliproteins, including R-phycoerythrin (R-PE), allophycocyanin, and phycocyanin, are extracted using buffer-based extraction, UAE, high-pressure homogenization (HPH), and cold water extraction. Buffer extraction from *Gracilaria gracilis* using sodium acetate-acetic acid (pH 5.5) and homogenization achieves 7 mg/g DW of R-PE, 3.5 mg/g DW of allophycocyanin, and 2 mg/g DW of phycocyanin [115]. UAE applied to *Sarconema skottsbergii* at 30–90 % amplitude for 10–30 min, producing 0.6–0.8 mg PE/g DW in phosphate buffer and 1.7–2.6 mg PE/g DW in distilled water. HPH, tested at 100–500 MPa with 1–3 passes, achieves 3.1–5.1 mg PE/g DW in phosphate buffer and 4.6–5.7 mg PE/g DW in distilled water, with optimal results at 400 MPa and two passes [116]. HPH consistently outperforms UAE in *Ulva palmata*, while natural deep eutectic solvents (NaDES) composed of glycerol:glucose (2:1) with 50 % water have proven the most effective solvent mixture [117]. Cold water extraction from *Porphyra dioica*, *Gracilaria vermiculophylla*, and *Gracilaria corneum* at 20–25 °C for 16 hours under agitation results in higher protein content (26 %) in *P. dioica* compared to *G. vermiculophylla* and *G. corneum* (15 %) [118].

Phenolic compounds from *Mastocarpus stellatus* are effectively extracted using scCO₂ and pressurized liquid extraction (PLE) with NaDES. scCO₂ extraction at 30 MPa achieves a lipid yield of 1.1 g per 100 g biomass, while ethanol-modified CO₂ (10 % ethanol, 30 MPa) enhances total phenolic content from 1.5 to 2.5 g GAE per 100 g extract. A subsequent MAE at 45 °C further increases the phenolic yield to 1.9 mg per 100 g [114]. PLE using ChCl:Gly (1:2) with 60 % water at 150 °C for 25 min results in higher TPC values (5–13 mg GAE/g extract) than conventional ethanol/water (70:30 v/v) PLE, demonstrating its effectiveness [117].

Agar and porphyrans, extracted from *G. corneum*, *G. vermiculophylla*, and *P. dioica*, are recovered using hot water extraction with or without alkaline pretreatment (APT). The APT method involves 6 % NaOH treatment at 85 °C for 3.5 hours, followed by neutralization with 0.5 % acetic acid. Species-specific extraction conditions include 85 °C for 2 hours in *G. vermiculophylla*, 95 °C for 3 hours in *G. corneum*, and 80 °C for 3 hours in *P. dioica*. Sequential processing improves total polysaccharide solubilization, though it reduces the yield of the main soluble polysaccharides [118].

Lipids extracted from *S. filiformis* using a DCM/MeOH (7:3 v/v) mixture under mechanical agitation for 24 hours result in a high saturated fatty acid (SFA) content (69.2–

73.6 %), dominated by palmitic acid (C16:0, 43.9–55.8 %). Monounsaturated fatty acids (MUFAs) range from 12.47 % to 16.57 %, while polyunsaturated fatty acids (PUFAs) reach ~14 %, including eicosapentaenoic acid (EPA, C20:5 ω 3) and arachidonic acid (AA, C20:4 ω 6) [113].

Lactic acid and galactose hydrolysates from *Kappaphycus alvarezii* have been recovered through hydrolysis, purification, and fermentation using *Lactobacillus pentosus* ATCC 8,041. Hydrolysis at 110 °C for 45 minutes in 1 % H₂SO₄ yields 40 g/L galactose with minimal HMF. Fermentation under pH 6, 37 °C, 150 rpm achieves a lactic acid yield of 1.37 g/g in batch mode, while pulse-fed batch fermentation improves productivity to 3.57 g/(L·h) [119].

- Feed and horticulture

Protein recovery for feed and food applications is influenced by extraction methods and solvent types. Alkaline extraction at 60 °C for 120 min with 0.5 M NaOH achieves 29 % protein recovery from *G. corneum*, significantly higher than the initial 14.52 % crude protein content [118].

MAE and subcritical water extraction (SWE) of *M. stellatus* in a microwave reactor at 190 °C for 3 minutes allow for protein extraction without degradation [114].

For horticultural application, biochar is obtained through fast pyrolysis of *G. gracilis* and its residues after phycobiliprotein extraction. Conducted at 400, 500, and 600 °C in an inert atmosphere, biochar yield decreases with increasing temperature, from 33 WW% at 400 °C to 26.5 WW% at 600 °C. Despite retaining P, K, Ca, Fe, and Mg, the mineral content of post-extraction residues varies, potentially reducing their fertilizer value [115].

- Energy applications

Bio-crude oil, a co-product of fast pyrolysis, is produced from *G. gracilis* at 400, 500, and 600 °C under inert conditions. The highest bio-oil yield (68 WW%) occurs at 500 °C, with an organic phase yield of 27.1 WW% for fresh biomass and 34.5 WW% for the residue. Similar yields between biomass and its residue suggest that pyrolysis is a viable strategy for valorizing macroalgae biorefinery waste streams [115].

1.5. LCA applications in the macroalgae biorefinery

The second key outcome of the literature review focuses on the LCA implications related to macroalgae biorefinery systems. Given the challenges in distinguishing economic analyses from technical assessments in LCC studies and the lack of available studies on S-LCA for macroalgae biorefineries, the review prioritizes LCA as the primary sustainability assessment tool.

Applying LCA to macroalgae-based biorefineries is crucial for evaluating environmental performance and identifying areas for process optimization. A critical factor in LCA studies is the choice of the functional unit (FU), which varies depending on the study objectives. Some studies define FU based on bio-based material production, such as 1 kg of polymeric packaging material from *S. latissima* [120], 1 kg of bioplastic [121], or 1 kg of biocrude oil from HTL [121]. Others focus on energy-related functional units,

including 1 MJ of energy from seaweed processing [122] or 1 GJ of biogas per year [111]. For broader biorefinery system evaluations, FU selection shifts to 1 tonne of dried seaweed [123] or 1 hectare of cultivated macroalgae sea [36]. These FU variations affect result interpretation, making cross-study comparisons challenging, particularly due to differences in system boundaries and allocation methods. Product-based functional units assess environmental impacts per unit of the final product, whereas energy-based functional units evaluate impacts relative to net energy yield, leading to diverging conclusions on sustainability.

LCIA methodology varies across studies, affecting both scope and comparability. The ReCiPe 2016 Midpoint method, particularly Hierarchist (H) and Egalitarian (E) perspectives, is one of the most commonly applied frameworks [92], [121], [124], [125]. Other studies adopt CML-IA, which emphasizes global warming potential (GWP), acidification, eutrophication, and photochemical ozone formation [97], [123]. Additional methodologies include Eco-indicator 99 [111] and IMPACT World+ [122]. The choice of software further influences LCA results, with SimaPro [121], [122], [123], [125] and OpenLCA [92], [120] being the most frequently used platforms. Furthermore, database selection, including Ecoinvent (versions 3.6, 3.8) and USLCI, impacts life cycle inventory (LCI) data quality and comparability across studies.

Environmental performance in macroalgae biorefineries is highly dependent on system configurations, allocation approaches, and energy sources. The allocation method significantly influences GWP values, with economic allocation yielding 1.2–4.52 kg CO₂ eq/kg, whereas system expansion increases emissions to 14.19–41.52 kg CO₂ eq/kg [120]. The energy intensity of key biorefinery processes, particularly biomass drying and extraction, substantially contributes to the climate impact [124]. Regional energy profiles also play a crucial role; for example, in coal-dependent regions such as South Africa, SWE and hot water extraction (HWE) exhibit higher carbon footprints compared to biorefineries powered by renewable energy [86]. Conversely, adopting a green energy mix reduces global warming impact, though it may introduce trade-offs in acidification, human toxicity, and land use [86].

Beyond energy consumption, process optimization is essential for enhancing biorefinery sustainability. Extraction processes have been identified as major environmental hotspots [125], particularly when solvent use and membrane-based separations increase both economic and environmental burdens. Strategies that minimize solvent consumption and lower energy demand are critical for improving sustainability. Additionally, by-product valorization enhances environmental performance, as failure to utilize process residues negatively affects overall sustainability [86], [125], [126]. Zero-waste strategies, such as wastewater reuse and biochar production, contribute both environmental and economic benefits. The water footprint of macroalgae biorefineries is another relevant factor, particularly in water-scarce regions, where SWE and HWE offer advantages over conventional extraction methods [86].

One of the most significant challenges in LCA studies is data uncertainty and scalability [124]. Many assessments rely on laboratory- or pilot-scale data, making industrial-scale

extrapolation difficult. The transition from small-scale trials to full-scale commercial operations introduces complexities in energy efficiency, process design, and waste management [121]. Focusing on single impact categories without considering the entire life cycle may result in burden shifting, where reducing GWP inadvertently increases eutrophication or resource depletion [86]. A holistic LCA approach is therefore required to assess multiple environmental trade-offs and ensure balanced decision-making.

Another crucial factor influencing macroalgae biorefinery feasibility is biomass cost and logistics. Harvesting, transportation, and processing costs contribute significantly to both economic and environmental burdens [126], [127]. The distance between cultivation sites and processing facilities impacts fuel consumption, GHG emissions, and supply chain costs, making logistical optimization essential. Strategies such as localized processing, decentralized biorefineries, and improved biomass handling can reduce transportation-related impacts and enhance resource efficiency. Additionally, on-site or nearshore pre-processing, which involves reducing biomass water content before transport, lowers transportation costs and improves supply chain sustainability.

1.6. Limitations of common macroalgae biorefinery systems

At the conclusion of the literature review, several critical challenges in macroalgae biorefinery development have been identified. These gaps highlight key areas where further research, such as that undertaken in this Thesis, should aim to contribute. A preliminary discussion on these critical aspects is already presented in Fig. 1.6, which illustrates the publication trends over time, categorizing studies by the macroalgae family. While the figure also reflects an overall increase in publication volume over time, the primary aim is not to emphasize the temporal trend but rather to highlight the significant disparity in research focus between different macroalgae species.

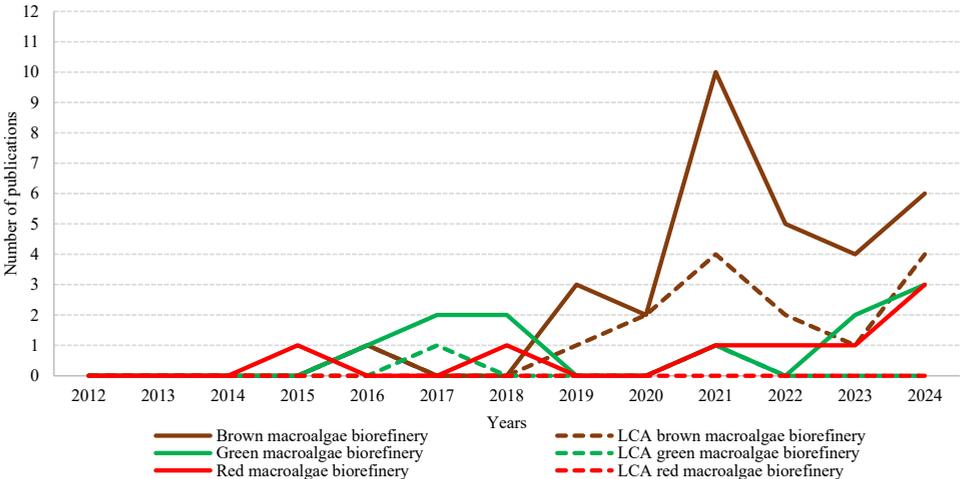


Fig. 1.6. Selected publications trends in the years 2012-2024.

The analysis reveals that brown macroalgae have been the primary focus of research, with 31 publications, significantly outnumbering studies on green macroalgae (11) and red macroalgae (9). This disparity is also reflected in LCA applications, where 15 studies have been conducted on brown macroalgae, while only 2 studies focus on green macroalgae and 0 studies on red macroalgae. These findings highlight a considerable gap in the integration of LCA within macroalgae biorefinery research, particularly for green and red macroalgae. Even within the most extensively studied category, brown macroalgae, less than half of the publications incorporate LCA, indicating that the application of environmental assessment methodologies in macroalgae biorefineries remains underdeveloped.

The literature review has then identified three major challenges that must be addressed to advance macroalgae biorefinery systems:

- Technological maturity.

Most of the macroalgae biorefineries reported remain largely at an experimental stage, lacking the technological maturity required for widespread industrial-scale deployment. The absence of optimized, scalable processes capable of efficiently handling the heterogeneous composition of macroalgae biomass represents a significant bottleneck. Extraction methods for bioactive peptides, polysaccharides, and other valuable compounds often require highly specific and tailored techniques, which may not be directly transferable or scalable across different macroalgae species. Furthermore, conversion processes must be improved in terms of efficiency and energy consumption to ensure the economic viability of large-scale production.

A key disconnect in current research is the gap between technological advancements and their environmental assessment. While significant efforts have been directed toward improving extraction and conversion processes, these innovations are often not immediately evaluated using LCA. As a result, LCAs tend to lag behind technological progress, preventing timely insights into environmental trade-offs and sustainability impacts. This misalignment between technological development and environmental assessment hinders the ability to effectively integrate sustainability considerations into biorefinery optimization.

- Biomass availability and quality.

The inconsistency of macroalgae biomass quality and availability poses a major challenge for biorefinery operations. Macroalgae growth is strongly influenced by environmental conditions, including water temperature, nutrient availability, and light intensity, which vary seasonally and regionally. These fluctuations impact both biomass yield and biochemical composition, affecting the efficiency and design of biorefinery processes. For example, lipid content, a crucial factor for biodiesel production, varies significantly between different seasons and environmental conditions. This variation affects conversion efficiency, process optimization, and economic feasibility, requiring adaptive processing strategies to maintain consistent product quality and yield. Developing robust feedstock management strategies and biorefinery designs that accommodate biomass variability is essential for improving the reliability and scalability of macroalgae-based biorefineries.

- Environmental impact and sustainability gaps.

While macroalgae cultivation is often promoted for its low land use requirements and carbon sequestration potential, the full ecological consequences of large-scale production remain insufficiently understood. Several key environmental concerns must be thoroughly assessed, including: potential for invasive species proliferation due to large-scale cultivation, alteration of local marine ecosystems, particularly in terms of habitat disruption and changes in nutrient cycles, and nutrient runoff and eutrophication risks, which could counteract environmental benefits if not properly managed.

To ensure that macroalgae biorefineries provide genuine environmental advantages, comprehensive LCAs must be conducted across the entire value chain, preventing unintended burden shifting where GHG reductions are achieved at the cost of other environmental impacts.

The current imbalance in LCA research across macroalgae groups further limits holistic sustainability assessments. Brown macroalgae are the most studied, with 15 LCA-focused studies, whereas only 2 studies have been conducted on green macroalgae and 0 on red macroalgae. This disparity highlights a critical research gap, underscoring the need to expand LCA applications to green and red macroalgae to achieve a more comprehensive and representative sustainability evaluation of macroalgae biorefineries.

1.7. Needs and opportunities for macroalgae biorefinery systems

At the conclusion of this chapter, several recommendations and key solutions can be outlined, some of which this Thesis aims to address.

- Development of integrated biorefinery processes.

A fundamental approach to overcoming economic barriers in macroalgae biorefinery development is the implementation of integrated processing strategies that maximize biomass valorization. By converting different macroalgal components into high-value chemicals, fuels, and materials, biorefineries can achieve greater economic resilience and operational efficiency. As highlighted in several studies within this chapter, residual biomass from biofuel production can be repurposed for biogas generation or used in agricultural applications, enhancing resource efficiency and economic returns. This circular approach not only improves financial viability but also contributes to waste minimization and sustainable resource utilization.

- Standardization of cultivation and harvesting techniques.

Despite the extensive focus on biomass valorization and extraction processes, standardized cultivation and harvesting methods remain underexplored in macroalgae biorefinery research. Ensuring consistent biomass quality is critical for process stability, particularly given the seasonal variability and environmental dependencies of macroalgae growth. Controlled-environment aquaculture techniques, such as land-based cultivation systems, could help mitigate seasonal fluctuations and improve yield predictability.

Moreover, advancements in harvesting technologies that minimize ecosystem disruption while enhancing biomass collection efficiency are essential for stabilizing the supply chain. Innovations such as automated harvesting systems, precision aquaculture techniques, and

selective harvesting strategies could improve both the sustainability and economic viability of large-scale macroalgae production.

- Optimization of LCA for environmental impact reduction.

Expanding the role of LCA as a process optimization tool can significantly enhance the environmental sustainability of macroalgae biorefineries. Comprehensive LCAs help identify critical environmental hotspots and guide process improvements to reduce carbon footprints, energy consumption, and resource depletion.

One key area of improvement is the regional adaptation of LCA studies to account for differences in biomass production, processing methods, and local ecological conditions. The integration of region-specific LCAs would enable the development of site-adapted biorefinery models, improving both efficiency and sustainability. Additionally, while some research highlights green technologies, precise quantification of their environmental benefits remains limited. Future efforts must focus on accurate impact assessment of eco-friendly extraction methods, energy-efficient processing technologies, and circular economy principles within macroalgae biorefineries.

Noticing these aspects, to further advance sustainable practices in the macroalgae biorefinery sector, different key future opportunities should be pursued.

- Technological innovation and scalability.

Addressing scalability and efficiency constraints is essential for the widespread commercialization of macroalgae biorefineries. Emerging biotechnological advancements, such as genetic engineering and selective breeding, hold significant potential for enhancing biomass yields and biochemical composition by developing macroalgae strains optimized for growth rates, nutrient uptake, and environmental resilience.

Additionally, the integration of automation, artificial intelligence, and bioprocess optimization techniques can streamline macroalgae cultivation, harvesting, and processing, significantly improving operational efficiency and economic feasibility. Implementing data-driven precision aquaculture would allow for real-time monitoring and optimization, reducing resource input while maximizing biomass productivity.

- Policy and regulatory Support.

A strong policy framework is crucial to accelerate the development and adoption of macroalgae-based biorefineries. Governments can foster growth through policy-driven incentives such as subsidies, tax credits, and funding programs, mitigating the financial risks associated with emerging bio-based industries.

Moreover, regulations supporting carbon emission reductions and promoting renewable energy integration can establish a favorable market environment for macroalgae-derived products. Clear legislative guidelines on sustainable aquaculture, marine spatial planning, and environmental management will be essential to facilitate the scaling-up of macroalgae production while ensuring minimal ecological impact.

- Market development for macroalgae-based products.

Expanding market demand is fundamental for the long-term economic viability of macroalgae biorefineries. Key industries such as bioplastics, pharmaceuticals, nutraceuticals, and bio-based chemicals present significant growth opportunities for

macroalgae-derived products. However, regulatory challenges, consumer awareness gaps, and cost competitiveness with fossil-based alternatives continue to limit market penetration.

To enhance market uptake, public awareness campaigns, collaborations between industry and research institutions, and corporate partnerships should be strengthened. Additionally, sustainability certification schemes and eco-labeling initiatives for macroalgae-based products can help build consumer trust and promote the transition toward bio-based alternatives.

- Implementation of S-LCA for community impact evaluation.

The integration of social sustainability considerations is increasingly recognized as a critical aspect of macroalgae biorefinery development. Several project initiatives [128], [129] have demonstrated the socio-economic potential of macroalgae-based industries, particularly in coastal and rural communities. These initiatives highlight how macroalgae biorefineries can generate employment opportunities, strengthen food security, and stimulate local economies through sustainable aquaculture and bio-based value chains.

Future research and development efforts should incorporate comprehensive S-LCA to evaluate community-level impacts, including: employment generation and skills development in coastal regions, effects on small-scale fisheries and traditional livelihoods, fair trade and ethical sourcing of macroalgae raw materials, and social equity and stakeholder engagement in biorefinery planning.

To maximize socio-economic benefits, it is essential to ensure that local communities are actively involved in decision-making processes throughout the planning and implementation of macroalgae biorefinery projects. Strategies such as community co-management models, participatory governance structures, and knowledge-sharing platforms can help equitably distribute benefits while addressing potential socio-environmental challenges.

2. METHODOLOGY

The methodology employed in this Thesis will be thoroughly detailed in the following sub-chapters. In summary, the process begins with the technological definition of the cascade biorefinery design, along with the identification of alternative designs. Subsequently, the methodology applied to assess the three pillars of sustainability, LCA, LCC, and S-LCA, will be outlined. Finally, the TOPSIS multicriteria analysis, adapted for LCSA, will be explained in detail.

2.1. The technological designs for the *F. lumbricalis* biorefinery

As extensively discussed in the previous chapter, a macroalgae biorefinery is a complex system that encompasses multiple operational stages. The upstream phase involves biomass acquisition, which can include wild harvesting or controlled cultivation, for example. Following this, the downstream phase focuses on extraction processes aimed at recovering valuable compounds from the biomass. This same structural approach is adopted in the following sub-chapters to provide a detailed explanation of the *F. lumbricalis* biorefinery system, outlining both its upstream and downstream processes.

The *F. lumbricalis* macroalgae

F. lumbricalis (Fig. 2.1) is a red macroalgae [130] commonly found in the North Atlantic and Arctic Ocean waters. It is one of the most dominant rhodophyte species in the Baltic Sea Region [131]. This macroalga primarily grows on hard substrates, including rocks, boulders, and stones, but it is also capable of thriving in open environments with sandy or muddy seabeds, where it forms dense monotypic meadows [130].



Fig. 2.1. The *F. lumbricalis* macroalgae [132].

As a perennial species, *F. lumbricalis* has a lifespan of 5 to 10 years and can grow at depths of up to 30 meters, although its highest occurrence is recorded between 8 and 12 meters [133]. Notably, this species exhibits high tolerance to low salinity environments, making it well-adapted to Baltic Sea conditions [130].

The collection site is located within Estonia, specifically in the Kassari Bay area (58.805°N, 22.786°E), at a depth of 7 meters (Fig. 2.2). This region contains an estimated biomass stock of up to 140,000 tons, the majority of which remains unutilized, presenting a significant resource opportunity for biorefinery applications.

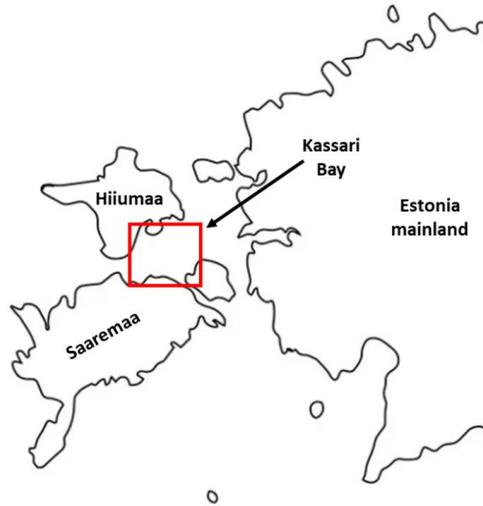


Fig. 2.2. The geographical location of Kassari Bay (Estonia). From approbation publication No. 2 [134].

Table 2.1 presents the average mineral composition and heavy metal concentrations of *F. lumbricalis*. This data provides essential insights into the elemental profile of the macroalga, which is crucial for evaluating its nutritional potential, industrial applications, and environmental safety considerations.

Table 2.1

Composition of the *F. lumbricalis* macroalgae. From approbation publication No. 4 [135]

Nutrients	Value	Unit
Phosphorus (P)	4,500	mg/kg
Potassium (K)	39,000	mg/kg
Calcium (Ca)	30,600	mg/kg
Magnesium (Mg)	9,630	mg/kg
Sodium (Na)	11,800	mg/kg
Sulphur (S)	44,000	mg/kg
Manganese (Mn)	1,597	mg/kg
Copper (Cu)	20.7	mg/kg
Boron (B)	182	mg/kg
Iron (Fe)	2,137	mg/kg
Zinc (Zn)	25	mg/kg
Cobalt (Co)	0.52	mg/kg
Molybdenum (Mo)	<0.7	mg/kg
Selenium (Se)	0.67	mg/kg
Arsenic (As)	9.7	mg/kg

Nutrients	Value	Unit
Mercury (Hg)	0.012	mg/kg
Cadmium (Cd)	0.332	mg/kg
Lead (Pb)	2.44	mg/kg
Chrome (Cr)	2.72	mg/kg
Nickel (Ni)	18.8	mg/kg
Total nitrogen	38	kg/t
Soluble nitrogen	0	kg/t
Nitrate nitrogen	4.6	kg/t

The macroelements composition of *F. lumbricalis* is distributed as follows: carbohydrates constitute 39.82 %, lipids account for 30 %, proteins make up 28.32 %, and ash content is 1.86 %.

Upstream processes (*F. lumbricalis* harvesting)

For the recovery of *F. lumbricalis*, wild harvesting (WH) has been selected as the preferred method. This choice aligns with the current industry practice, as wild collection remains the primary means of obtaining this species. While there is one documented study exploring the artificial cultivation of *F. lumbricalis*, it remains at an experimental stage and has not yet progressed beyond exploratory research [136]. Additionally, WH is the established method used by Vetik OÜ [132], the industrial partner of the TACO ALGAE project, which holds annual licenses permitting the harvest of up to 1,000 tons of *F. lumbricalis* per year for a single license. Consequently, the following process description is based on data collected from Vetik OÜ, making it specific to this case study.

The WH scenario analyzed in this research focuses on the collection of *F. lumbricalis* in Kassari Bay, Estonia. A process flowchart illustrating the different stages is presented in Fig. 2.3. The harvesting operations begin with the deployment of a trawling boat equipped with a diesel engine, which travels approximately 40 minutes from the harbor to reach the designated harvesting area. The collection method involves a custom-designed trawling apparatus, consisting of a rectangular metal frame covered with a net, which is towed at low speed behind the boat to efficiently gather the target species. This technique has been shown to achieve an extraction efficiency of 92.9 % for *F. lumbricalis*. However, it also results in the unintentional capture of 5% clam shells and 2.1 % clay and mud, which are categorized as solid waste.

Following the harvesting phase, the trawling boat returns to the harbor, where the collected biomass is unloaded using an electrically powered unloading system. The harvested *F. lumbricalis* is then transported by road (53.2 km) to a processing facility, where it undergoes further refinement and value-added processing.

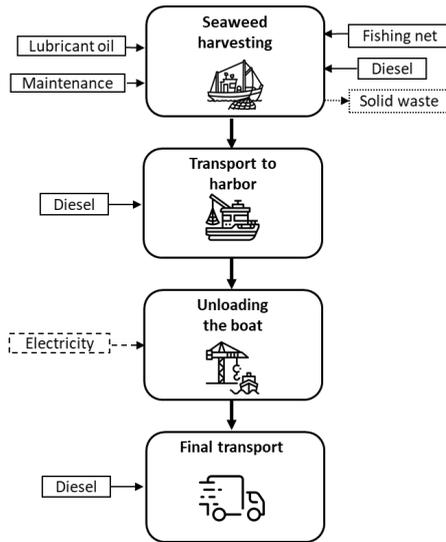


Fig. 2.3. Chart flow for the WH process. From approbation publication No. 2 [134].

Proposed cascade biorefinery design (downstream processes)

To achieve the full valorization of *F. lumbricalis*, a multiproduct cascading biorefinery approach has been developed. This strategy, designed in collaboration with Vetik OÜ and Nofima AS (i.e., research institute partner of the TACO ALGAE project), enables the circular and sustainable utilization of the macroalgae by maximizing the recovery of valuable compounds while minimizing waste.

The proposed biorefinery process allows for the extraction of multiple high-value products, including pigments, proteins, and carrageenan. Following these extraction stages, the remaining biomass residue is further repurposed as a fertilizer amendment or used for biogas production, ensuring that all fractions of the biomass contribute to value-added applications.

Fig. 2.4 presents a technical flow diagram illustrating the cascading biorefinery process, detailing each step involved in the efficient and sustainable processing of *F. lumbricalis*.

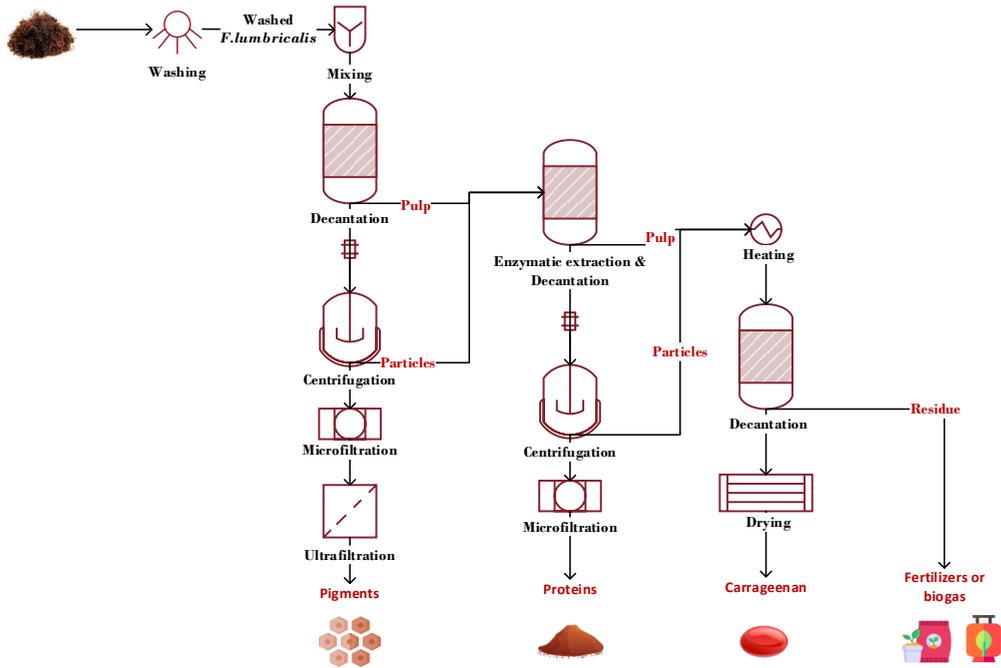


Fig. 2.4. System diagram for the cascade biorefinery design. Adapted from the approbation publication No. 4 [135].

The technological design can be summarized in 5 stages:

1. Pre-treatment: biomass washing

The pre-treatment phase of *F. lumbricalis* is crucial for ensuring biomass purity before further processing. This stage primarily involves removing impurities, including sand, marine organisms (such as clams, shrimp, juvenile fish, and insects), and other contaminants. The washing process requires approximately 10 m³ of water per ton of fresh macroalgae, effectively eliminating unwanted materials and ensuring the biomass meets the required quality standards for subsequent extraction steps.

2. Pigment extraction (First Processing Line)

Pigments, recognized as the highest-value component within *F. lumbricalis* biorefineries [137], are extracted as the first product in the cascading process. The extraction begins with an aqueous-based method, where the biomass is mixed with water in a 1:1 ratio. Phase separation is then conducted using specialized machinery, including Flottweg Z3E and Alfa Laval Separator AFPX 810 centrifuges, to recover the solid and liquid fractions.

To further purify the extracted pigments, microfiltration (utilizing the Alfa Laval 6.3" UF/MF membrane spiral system) and ultrafiltration (<5 kDa) techniques are applied. This multi-stage filtration approach enhances pigment recovery efficiency, ensuring a high-purity extract suitable for commercial applications. The remaining biomass, after pigment extraction, is redirected for subsequent processing.

3. Protein extraction (Second Processing Line)

Following pigment recovery, the remaining biomass (pulp and particles) undergoes protein extraction. The process begins with heating the biomass to 40 °C, followed by the addition of water (1:2 ratio), alcalase enzyme (0.006 L per kg of dry matter), and KOH (0.006 kg per kg of dry matter). The enzymatic hydrolysis occurs over two hours under continuous stirring, facilitating protein release.

After the enzymatic treatment, phase separation is conducted using the same centrifugation system as in pigment extraction. The soluble protein fraction is further purified via microfiltration, while the remaining solid fraction is directed to the carrageenan extraction stage.

4. Carrageenan extraction (Third Processing Line)

The input for this stage consists of the residual pulp and particles from protein extraction. To recover carrageenan, the biomass undergoes thermal processing at 95 °C with the addition of water (1:2 ratio) and KOH (0.006 kg per kg of dry matter). The extraction process is followed by phase separation using a tricanter, ensuring that carrageenan is isolated while the biomass is still warm.

The final step in this process involves drying and evaporation, utilizing the Alfa Laval AlfaVap1 effect with TVR technology to concentrate and recover carrageenan. The residual biomass, now considered seaweed waste, is subsequently repurposed as a biostimulant for agricultural applications.

5. Biostimulant application - option 1

The residual biomass from the cascade biorefinery process is repurposed as a fertilizer additive, taking advantage of its biostimulant properties attributed to its rich mineral composition (as outlined in Table 2.1). Extensive examples of its horticultural applications are discussed in Chapter 1.3, highlighting its effectiveness in enhancing crop productivity, soil health, and plant resilience.

6. Biogas production - option 2

This scenario focuses on the potential integration of biogas production and electricity cogeneration by utilizing residues from carrageenan extraction, rather than repurposing them as biostimulants. The technological framework for this approach is based on the study conducted by Fasahati et al. (2022) [123], which provides a detailed process description and inventory for biogas generation from macroalgae.

The process begins with anaerobic digestion, where the organic residues undergo microbial breakdown to produce biogas. The resulting biogas is then converted into electricity through a boiler/turbogenerator system, allowing for on-site energy generation. This alternative valorization pathway enhances the circularity and energy efficiency of the macroalgae biorefinery, reducing waste while contributing to renewable energy production.

The described biorefinery system is theoretically designed to extract 20 kg of pigments, 100 kg of proteins, and 200 kg of carrageenan from 1,000 kg of DW *F. lumbricalis*. However, it is important to emphasize that these values are theoretical estimations derived

from hypothetical process modeling and laboratory analyses and do not yet have industrial-scale validation.

A mass balance analysis of the process, considering 1 ton of DW *F. lumbricalis*, is illustrated in Fig. 2.5. For reference, 1 ton of DW corresponds to 7.6 tons of WW biomass, highlighting the significant water content in the raw feedstock.

Pre-treatment		Pigments extraction		Proteins extraction		Carrageenan extraction	
Washing (kg)		1st line (kg)		2nd line (kg)		3rd line (kg)	
INPUT		INPUT		INPUT		INPUT	
<i>Flumbricalis</i> harvested	7,692	<i>Flumbricalis</i> biomass for the biorefinery	7,692	Pulp + particles 1st line	7,672	Pulp + particles 2nd line	7,572
Water	76,920	Water	7,692	KOH	5,88	KOH	5,28
OUTPUT		OUTPUT		OUTPUT		OUTPUT	
Wastewater	76,920	Wastewater	7,692	Wastewater	15,355	Water to air	15,144
<i>Flumbricalis</i> biomass for the biorefinery	7,692	Pulp 1st line	7,288.40	Pulp 2nd line	6,814.80	Residue	7,377.28
		Particles 1st line	383.6	Particles 2nd line	757.20	Carrageenan	200
		Pigments	20	Proteins	100		

Fig. 2.5. Mass balance for the cascade biorefinery design. From the approbation publication No. 4 [135].

It is important to acknowledge that the proposed biorefinery configuration represents just one of several possible design pathways. For instance, alternative techniques for pigment extraction, such as ultrasound-assisted or enzymatic extraction, were also considered during the design phase [138]. Despite the potential of these methods to reduce energy consumption and improve efficiency, the water-based extraction route was ultimately selected due to the availability of reliable primary data, which ensures greater accuracy and consistency in the sustainability assessment. Nevertheless, the potential environmental and economic advantages of these alternative extraction techniques are further explored and critically discussed in the results chapter.

Alternative biorefinery designs (downstream processes)

The alternative biorefinery designs developed in this research represent two conventional approaches that have been previously documented in the literature, contrasting with the cascade biorefinery model described earlier. Up to the pre-treatment, all designs follow identical technological layouts, meaning that *F. lumbricalis* is harvested and pre-treated using the same procedures in all three cases. However, a major divergence occurs in the biorefinery extraction stages, specifically in how biomass is processed, how waste is managed, and the conceptualization of final products. The two designs are:

1. Single-product extraction

The single-product extraction represents the most direct scenario in biorefinery operations. It is widely applied in LCA studies in the literature, particularly concerning the use of macroalgae for energy production, like bioethanol [42], [139], and biogas [111]. For biocompounds, single extraction methods have been reported for red seaweed-derived carrageenan (*Soliera chordalis* and *Hypnea musciformis*), proteins (*Gracilaria lemaneiformis*), agar (*G. vermiculophylla*), and peptides (*P. yezoensis*) [140].

These studies are valuable for understanding the phenomenon and identifying the benefits of various extraction technologies, helping to determine the critical parameters and

optimal processes for maximizing product yield. However, this approach does not align with the circularity concept, as it does not involve the systematic reuse of extraction residues. As a result, the biomass is utilized for a single product extraction, leaving other valuable compounds in the residues. Consequently, more initial biomass is required to obtain equivalent quantities of valuable compounds compared to a cascade approach. The system modeled in this study includes separate lines for pigment, protein, and carrageenan extractions. For comparability, it is assumed that seaweed waste continues to be used to produce an equivalent amount of fertilizers or biogas. In Fig. 2.6, the technological scheme for the single-product extraction design is reported.

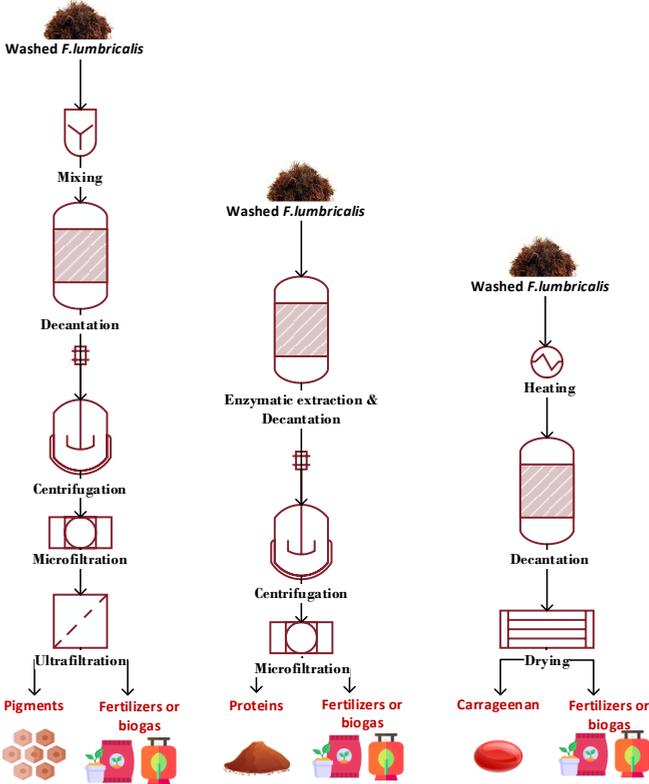


Fig. 2.6. System diagram for the single-product extraction design. From the approbation publication No. 4 [135].

The mass balance for the single-product design is reported in Fig. 2.7. It considers 1 ton of DW *F. lumbricalis*.

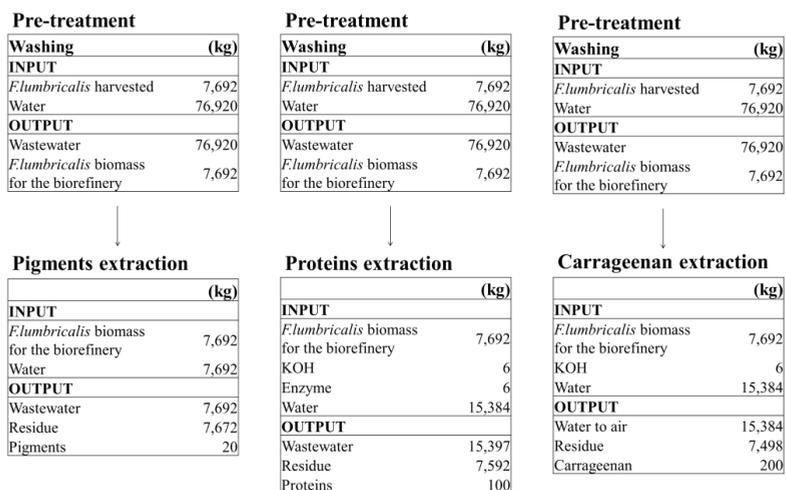


Fig. 2.7. Mass balance for the single-product extraction design. From the approbation publication No. 4 [135].

2. Three-line extraction

This configuration distributes the initial biomass equally across three extraction lines dedicated to recovering pigments, proteins, and carrageenan, as illustrated in Fig. 2.9.

Although the feedstock input remains consistent with that of the cascade biorefinery model, the efficiency in obtaining valuable compounds is significantly lower, making this approach less effective for utilizing *F. lumbricalis* as a feedstock.

A major limitation of this model is that valuable bioactive compounds remain within the residual biomass, which is subsequently directed toward biogas and fertilizer production rather than being fully valorized through integrated extraction processes.

In this scenario, the system processes 1 ton of *F. lumbricalis* biomass, allocating equal portions of 333.33 kg per extraction line. Consequently, the final product yields are substantially reduced compared to the cascade biorefinery: pigment recovery amounts to 13.33 kg, protein extraction yields 33.33 kg, and carrageenan output reaches 66.67 kg.

Fig. 2.8 reports the mass balance of the three-line extraction design. It considers 1 ton of DW *F. lumbricalis*.

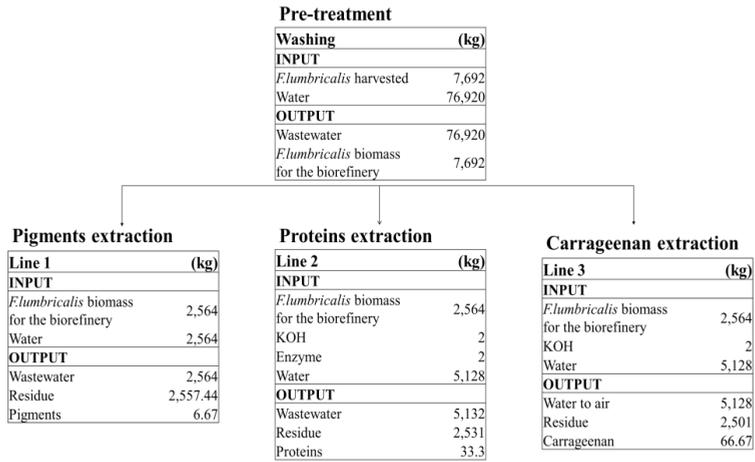


Fig. 2.8. Mass balance for the three-line extraction design. From the approbation publication No. 4 [135].

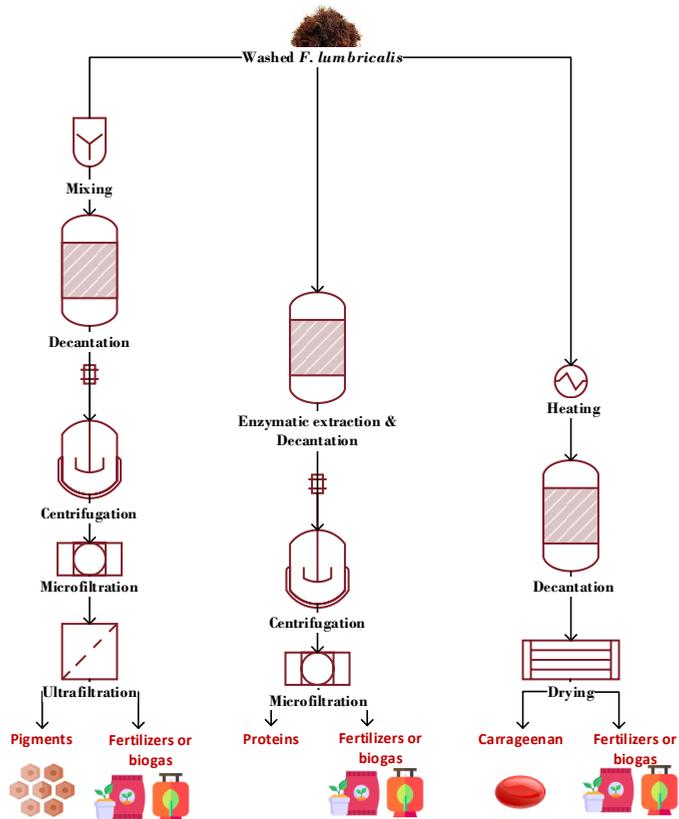


Fig. 2.9. System diagram for the three-line extraction design. From the approbation publication No. 4 [135].

2.2. Environmental life cycle assessment

This chapter outlines the implementation of the LCA to evaluate the environmental sustainability of the proposed biorefinery systems. Initially, a concise introduction to the LCA framework methodology will be provided, establishing the basis for its application. The assessment will then be conducted on the upstream biorefinery phases, aiming to identify the environmental impacts associated with different harvesting and cultivation techniques. Following this, a comprehensive LCA of the full cascade biorefinery system will be performed to determine the environmental hotspots within the process. Finally, the environmental performance of the cascade biorefinery will be compared against alternative scenarios, enabling a holistic evaluation of sustainability across different biorefinery configurations.

The LCA framework

LCA is a methodology used to analyze, evaluate, and estimate the potential environmental impact associated with a material, product, service, or process throughout its entire life cycle, from raw material extraction to transportation, utilization, and final disposal [141]. Following the guidelines set by ISO 14040 and ISO 14044, LCA is structured into four key phases: (1) goal and scope definition, (2) inventory analysis, (3) impact assessment, and (4) interpretation, as illustrated in Fig. 2.10.

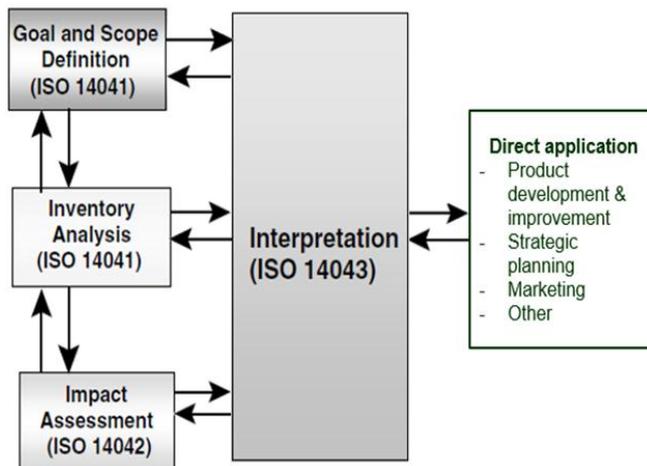


Fig. 2.10. Stages of an LCA study [141].

The goal and scope definition phase establishes the system boundaries and the level of detail required based on the study's objectives. The precision and depth of an LCA vary depending on its intended purpose. The LCI analysis involves the collection of data necessary for modeling the system under study. In the impact assessment phase, the results from the LCI are further analyzed to evaluate their environmental significance, providing a clearer understanding of the environmental performance of the system. The interpretation

phase consolidates the findings, ensuring that the results align with the initial goal and scope, enabling a comprehensive discussion of the system's environmental implications.

Each system assessed through LCA can serve multiple functions, each corresponding to a specific characteristic of the system itself. The FU plays a critical role in quantifying these functions, ensuring consistency in input and output assessments, and facilitating comparability between LCA studies. The system boundary defines the unit processes included in the study, ensuring that all inputs and outputs within the boundary are accounted for as elementary flows. The selection of system elements depends on the predefined research objectives, intended application, target audience, data availability, cost constraints, and cut-off criteria. Properly defining system boundaries is essential to determine the reliability of study outcomes and the feasibility of achieving the research objectives.

The inventory analysis phase consists of data collection and computational procedures required to quantify system inputs and outputs. This is an iterative process, as continuous data refinement improves system representation and enhances study accuracy. Inputs and outputs can be classified into several categories, including: (1) energy consumption, raw materials, and physical inputs; (2) products, co-products, and waste; and (3) emissions to air, discharges to water, and soil contamination. Additionally, special consideration must be given to allocation methods, particularly in systems involving multiple products and recycling loops.

Finally, different LCIA methodologies can be applied in order to evaluate the environmental performance of the selected systems.

To validate and contextualize the LCA methodology used in this Thesis, it was first applied to systems sharing key characteristics with the proposed macroalgae biorefinery.

The first case involved the assessment of a microalgae cultivation system [142], which provided valuable insight into parameter selection, particularly regarding energy use, waste, and wastewater flows, and biogas valorization. In this context, results from the microalgae study, especially those concerning the CHP unit, were reused and adapted for the *F. lumbricalis* system. Given the similar geographical location within the Baltic region, this case also informed regional modeling parameters and energy dynamics relevant to medium-scale biogas facilities.

A second reference case focused on the LCA of a novel food waste treatment system based on Black Soldier Fly larvae [143]. This system was selected due to its closed-loop design, which closely aligns with the waste valorization concept underpinning the cascade biorefinery. Specifically, this case study offered useful insights into inventory modeling for biomass-centric systems and the development of alternative scenarios. These methodological approaches were subsequently adapted and refined for use in the assessment of the *F. lumbricalis* cascade biorefinery.

LCA on the different techniques of harvesting and cultivation for the upstream stage

The primary objective of this initial LCA analysis is to identify the environmental hotspots associated with the WH of *F. lumbricalis* in Kassari Bay, Estonia, which represents the predominant method for this biomass collection.

To establish a benchmark comparison, alternative macroalgae cultivation techniques have also been evaluated. Given that no other documented *F. lumbricalis* cultivation methods exist beyond WH, the comparative analysis has been conducted using cultivation techniques applied to other macroalgal species. For the off-shore cultivation (OFC) system, the methodology outlined by Seghetta et al. (2016) [36] was adopted since it provides a detailed and quantified inventory.

The on-shore cultivation (ONC) system was assessed based on the approach developed and tested by HYNDLA [144] (another industrial partner of the TACO ALGAE project) on the red macroalgae *Schizymenia valentinae*. Also, in this case, primary data were prioritized to enhance the reliability and accuracy of the assessment.

The FU for this study is defined as “1 ton of fresh macroalgae harvested or produced, assuming 100 % species purity”. This choice ensures methodological consistency and facilitates comparability with similar LCA studies on macroalgae systems reported in the literature [145], [146].

The LCA study was conducted using the SimaPro 9.4 software, developed by Pré Consultants [147]. This software was selected due to its comprehensive database integration, robust modeling capabilities, and wide acceptance in scientific and industrial LCA studies. It enables detailed environmental impact assessments, ensuring the accurate evaluation of the proposed scenarios within the study. The database used is Ecoinvent 3.8. The Ecoinvent database was selected for this Thesis’ LCA analysis due to its status as one of the most comprehensive, transparent, and widely recognized LCI databases available. It provides high-quality, peer-reviewed data covering a broad spectrum of industrial, agricultural, transport, and energy processes across multiple geographical contexts. This ensures both data reliability and methodological consistency across system boundaries. Ecoinvent supports multiple system models, allowing flexibility in aligning the LCA with the intended goal and scope, whether attributional or consequential. Its datasets are structured according to ISO 14040/44 standards, facilitating standardized and reproducible results, which are essential for academic rigor and transparency [148].

Below are discussed the specific LCA features of the research.

- System boundaries

For the WH, the system described in Chapter 2.1 was adopted.

The scenario of the ONC is summarized in Fig. 2.11.

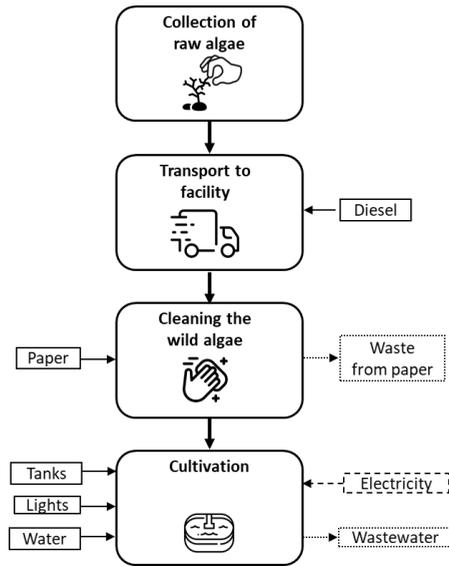


Fig. 2.11. Chart flow for the ONC process. From approbation publication No. 2 [134].

The process begins with the harvesting of *S. valentinae* specimens directly from their natural habitat in the Reykjavik area, Iceland. This collection takes place twice a year, in spring and autumn, with a maximum yield of 8 kg per harvest. Following collection, the harvested seaweed undergoes road transportation over a distance of 15 km to the designated cultivation facility. Upon arrival, the biomass is manually cleaned using paper towels, weighed, and prepared for the cultivation phase.

The cultivation process takes place in fiberglass tanks, where the seaweed undergoes a growth phase lasting six weeks. During this period, seawater is the only input, which is directly pumped from drill holes at depths of 20–40 m. Due to the natural nutrient richness of this water, no additional nutrients are required. By the end of the six-week growth cycle, the seaweed achieves an average growth rate of approximately 270 % [144]. Once matured, the newly grown specimens are collected and manually cut into 36 mm fragments to promote continuous regrowth. This cyclical growing-harvesting process is sustained throughout the year.

In terms of energy consumption, the two primary energy-intensive operations during cultivation are seawater pumping and tank lighting, which is used to simulate natural sunlight. However, all the electricity required for these operations is sourced from geothermal energy.

The OFC is detailed in Fig. 2.12.

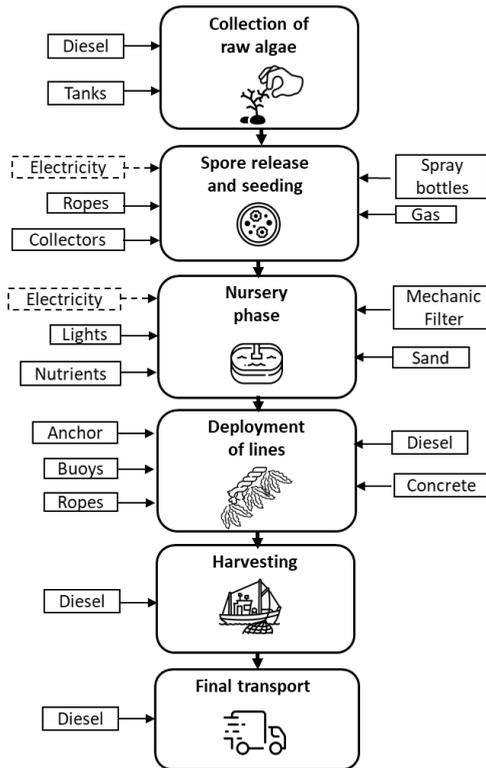


Fig. 2.12. Chart flow for the OFC process. From approbation publication No. 2 [134].

The OFC system follows the design outlined by Seghetta et al. (2016) [36] for *L. digitata*, selected for its integration into a Baltic Sea Region context, specifically in Denmark. Although not explicitly tailored for *F. lumbricalis* or *S. valentinae*, this system provides a comprehensive inventory and aligns with other OFC designs documented in the literature for red seaweed species [149], [150].

The first phase for the OFC is creating a bioengineered cultivation system. Some macroalgae specimens are taken from their natural environment and will serve as an initial inoculum for the community's growth on the artificial substrate. It consists of Kuralon twine lines on which seaweed spores are deposited. Once the seeded lines have been prepared, they are deployed at sea. The operation is carried out with vessels. They are also used to maintain the lines and collect the algae produced. A final road transport is considered to standardize this technique for the WH scenario.

- Alternative scenarios

This study explores the potential for modifying specific parameters from the baseline scenario, introducing alternative scenarios (AS) to assess possible improvements in the environmental performance of the system. The analysis of these alternative configurations enables a better understanding of how adjustments in key factors can enhance sustainability. The proposed alternative scenarios include:

1. AS1 - Implementing solar energy to supply 50 % of the electricity demand in both ONC and OFC systems. This adjustment is motivated by the observation that electricity consumption in these systems plays a crucial role in determining environmental impact.
2. AS2 - Modifying the ONC system by replacing artificial LED lighting with natural sunlight to reduce energy consumption and improve sustainability.
3. AS3 - Introducing an environmentally friendly antifouling agent, replacing the copper oxide-based solution currently used in the WH scenario. The new antifouling agent is modeled from the publication of Lin and Usino (2014) [151].
4. AS4 - Reducing diesel consumption by 10 % in both the WH and OFC scenarios, focusing on vessel fuel efficiency improvements to lower emissions and resource use.

The alternative scenarios, AS1 and AS2, were also aligned with the approach defined in the Black Soldier Fly case study [143], ensuring methodological consistency in the development and application of scenario-based LCA modeling.

- LCIA selected

The Environmental Footprint 3.0 impact assessment method [152] was selected to evaluate the environmental performance of the three scenarios. A set of key impact indicators was chosen to ensure a comprehensive assessment of the potential environmental burdens associated with macroalgae biorefinery operations. These indicators include:

- Climate change (CC), expressed in kg CO₂ eq., is a crucial metric for assessing the GWP of materials and processes. This indicator is particularly relevant for evaluating the emissions linked to harvesting, transportation, and processing activities.
- Particulate matter (PM), measured in disease incidence, is particularly useful for quantifying the health-related impacts of emissions, especially those arising from the extensive use of boats in WH and OFC scenarios.
- Acidification potential (AP), expressed in mol H⁺ eq., represents atmospheric pollution resulting from anthropogenic emissions of sulfur, nitrogen oxides (NO_x), and ammonia [153], [154]. This indicator is crucial for assessing the emissions generated by vessel operations and combustion processes throughout the system.
- Eutrophication freshwater (EF) and eutrophication marine (EM), quantified in kg P eq. and kg N eq., respectively, are particularly relevant due to the aquatic nature of the operations. Given that macroalgae cultivation and harvesting occur in marine environments, it is essential to measure their potential contribution to nutrient enrichment and ecosystem imbalances.
- Ecotoxicity freshwater (ECF), expressed in CTUe (Comparative Toxic Unit for aquatic ecotoxicity), evaluates the toxic effects of emissions on aquatic ecosystems. This metric is particularly relevant as both WH and OFC systems directly interact with marine habitats, making ecotoxicity a key consideration in assessing their sustainability.
- Land use (LU) and water use (WU), measured in impact points and m³ depriv., respectively, are essential for ONC assessments. The intensive land occupation and

freshwater consumption required for ONC necessitate an evaluation of their environmental implications.

- Resource use fossil (RF) and resource use minerals & metals (RM&M), expressed in MJ and kg Sb eq., respectively, quantify the impact of fossil fuel consumption and the depletion of critical mineral and metal resources. The high dependency on fossil energy inputs, particularly in marine-based operations, makes this indicator crucial for evaluating the overall sustainability of macroalgae-based biorefinery systems.
 - LCI

The dataset utilized for the WH scenario is detailed in Table 2.2, where all values are normalized according to the FU and adjusted based on the lifetime of equipment and processes (when applicable). The scaling of these values is directly correlated with the total biomass of seaweed harvested in 2021 by VETIK OÜ, ensuring that the data accurately reflects real-world operations.

Table 2.2

LCI for the WH scenario. Adapted from approbation publication No. 2 [134]

	Category	Material/ component	Input on SimaPro software	Quantity	Unit	Data source
Seaweed harvesting						
<i>Inputs</i>	Raw materials	Fishing net	Nylon 6-6 {RER} market for nylon 6-6 APOS, U	1.67	kg	Primary data
		Diesel for harvesting	Diesel, burned in fishing vessel {EU} diesel, burned in fishing vessel APOS, U	190	MJ	Primary data
		Lubricant oil	Lubricating oil {RER} market for lubricating oil APOS, U	2.75	kg	Primary data
		Epoxy paint	Epoxy resin, liquid {RER} production APOS, U	1.25	kg	Primary data
		Antifouling agent	Specific model from [151]	0.42	kg	Primary data
		Steel for the boat	Long liner, steel {RER} long liner construction, steel APOS, U	4.00	kg	[155]
<i>Outputs</i>	Raw materials, waste	Sand, clay, and mud waste	Refinery sludge {Europe without Switzerland} treatment of refinery sludge, sanitary landfill APOS, U	21	kg	Primary data
Transport to the harbor						
<i>Inputs</i>	Raw materials	Diesel for transport	Diesel, burned in fishing vessel {EU} diesel, burned in fishing vessel APOS, U	52.77	MJ	Primary data
Unloading the boat						
<i>Inputs</i>	Energy	Electricity for unloading the boat	Electricity, medium voltage {EE} market for APOS, U	0.69	kWh	Primary data
Final transport						
<i>Inputs</i>	Transport	Freight lorry	Transport, freight, lorry 7.5-16 metric ton, euro5 {RER} market for transport, freight, lorry 7.5-16 metric ton, EURO5 APOS, U	53.20	tkm	Primary data

Table 2.3 presents the inventory data for the ONC scenario, where all values are normalized based on the FU and adjusted according to the lifetime of equipment and processes (when applicable). The data are directly correlated with the total macroalgae biomass cultivated in 2021 by the HYNDLA.

Table 2.3

LCI for ONC scenario. Adapted from approbation publication No. 2 [134]

	Category	Material/ component	Input on SimaPro software	Quantity	Unit	Data source	
Transport to the facility							
<i>Inputs</i>	Transport	Passenger car	Transport, passenger car, EURO 4 {RER} market for APOS, U	15	km	Primary data	
Cleaning the wild algae							
<i>Inputs</i>	Raw materials	Paper tissues	Tissue paper {GLO} market for APOS, U	0.34	kg	Primary data	
<i>Outputs</i>	Waste	Paper waste	Waste paper, unsorted {Europe without Switzerland} market for APOS, U	0.34	kg	Primary data	
Cultivation							
<i>Inputs</i>	Raw materials	Tanks	Polystyrene, general purpose {GLO} market for APOS, U	1.40	kg	Primary data	
		Sea water	Water, Saline water consumption	32,640	kg	Primary data	
	Water cleaning	Water cleaning	Tap water {Europe without Switzerland} market for APOS, U	1,120	kg	Primary data	
		Soap cleaning	Modeled based on technical datasheet	0.446	l	Primary data	
		LED lights	Light emitting diode {GLO} market for APOS, U	0.1587	kg	Primary data	
	Water pump	Water pump	Cast iron {GLO} market for APOS, U	12	kg	Primary data	
		Energy	Electricity for lights	Electricity, medium voltage {IS} market for APOS, U	4,652	kWh	Primary data
			Electricity for pumps	Electricity, medium voltage {IS} market for APOS, U	2.22	kWh	Primary data
	Land use	Cultivation facility	Occupation, industrial area	333	m ² a	Primary data	
	<i>Outputs</i>	Wastewater	Wastewater	Wastewater, from residence {CH} market for wastewater, from residence APOS, U	1.12	m ³	Primary data
Cultivation with photovoltaic (added from the baseline scenario raw materials) - for ASI							
<i>Inputs</i>	Raw materials	Solar panel	Photovoltaic laminate, multi-Si wafer {UE} market for APOS, U	0.24	m ²	Primary data	
		Inverter	Inverter, 0.5kW {GLO} market for APOS, U	0.03	p	Primary data	

The electricity consumption for pumping water in the ONC scenario was modeled using the equation proposed by Piccino et al. (2016) [156]. This approach was necessary because the company shares its pumping system with another facility, making it challenging to isolate the specific energy consumption attributed solely to macroalgae cultivation.

Table 2.4 presents the inventory data for the OFC scenario, derived from the inventory reported by Seghetta et al. (2016) [36]. The values have been appropriately scaled to align with the FU of this study, ensuring consistency and comparability with the other cultivation and harvesting scenarios.

Table 2.4

LCI for OFC scenario. Adapted from approbation publication No. 2 [134]

	Category	Material/ component	Input on SimaPro software	Quantity	Unit	Data source	
Collection of raw algae							
<i>Inputs</i>	Raw materials	Diesel	Diesel {Europe without Switzerland} market for APOS, U	1.15	kg	[36]	
		Tank	Polyethylene terephthalate, granulate, amorphous {GLO} market for APOS, U	0.017	kg	[36]	
Spore release and seeding							
<i>Inputs</i>	Raw materials	Plastic jug	Polyethylene terephthalate, granulate, amorphous {GLO} market for APOS, U	0.035	kg	[36]	
		Block of collectors	Polyethylene, high density, granulate {GLO} market for APOS, U	1.382	kg	[36]	
		Kuralon	Modeled from [157]	3.71	kg	[36]	
		Twine					
		Spray bottles	Polyethylene terephthalate, granulate, amorphous {GLO} market for APOS, U	0.014	kg	[36]	
		Energy	Natural gas	Natural gas, high pressure {Europe without Switzerland} market group for APOS, U	0.40	m ³	[36]
			Electricity	Electricity, medium voltage {EE} market for APOS, U	5.18	kWh	[36]
		Electricity	Electricity, medium voltage {EE} market for APOS, U	1.62	kWh	[36]	
Spore and release with photovoltaic (added from the baseline scenario raw materials) - for ASI							
<i>Inputs</i>	Raw materials	Solar panel	Photovoltaic laminate, multi-Si wafer {UE} market for APOS, U	0.00042	m ²	[36]	
		Inverter	Inverter, 0.5kW {GLO} market for APOS, U	0.0307	p	[36]	
Nursery phase							
<i>Inputs</i>	Raw materials	Sand	Sand {CH} market for sand APOS, U	0.77	kg	[36]	
		Mechanical filter	Polypropylene, granulate {GLO} market for APOS, U	0.18	kg	[36]	
		UV lamp	Light emitting diode {GLO} market for APOS, U	0.0000105	kg	[36]	
		F2 medium	From the commercial formulation	0.83	l	[36]	
		Energy	Electricity	Electricity, medium voltage {EE} market for APOS, U	0.51	kWh	[36]
	Electricity		Electricity, medium voltage {EE} market for APOS, U	0.13	kWh	[36]	
	Electricity		Electricity, medium voltage {EE} market for APOS, U	0.08	kWh	[36]	
		Electricity	Electricity, medium voltage {EE} market for APOS, U	0.57	kWh	[36]	
Deployment of lines							
<i>Inputs</i>	Raw materials	Screw anchor	Cast iron {GLO} market for APOS, U	0.933	kg	[36]	

Category	Material/ component	Input on SimaPro software	Quantity	Unit	Data source	
Consumption	Black buoys	Polyethylene, high density, granulate {GLO} market for APOS, U	14.77	kg	[36]	
	Ropes	Nylon 6-6 {RER} market for nylon 6-6 APOS, U	7.98	kg	[36]	
	Concrete blocks	Concrete block {DE} market for concrete block APOS, U	138.23	kg	[36]	
	Ropes for buoys	Polypropylene, granulate {GLO} market for APOS, U	3.11	kg	[36]	
	Headline rope	Polypropylene, granulate {GLO} market for APOS, U	3.36	kg	[36]	
	Concrete block rope	Concrete block {DE} market for concrete block APOS, U	5.18	kg	[36]	
	Deployment of lines	Transport, freight, sea, Longliner {GLO} transport, freight, sea, ferry APOS, U	311	tkm	[36]	
Harvesting						
Inputs	Consumption	Boat use	Transport, freight, sea, Longliner {GLO} transport, freight, sea, ferry APOS, U	311	tkm	[36]
		Sea use	Occupation, sea and ocean	0.0003	ha	[36]
Final transport						
Inputs	Transport	Freight lorry	Transport, freight, lorry 7.5-16 metric ton, euro5 {RER} market for transport, freight, lorry 7.5-16 metric ton, EURO5 APOS, U	53.20	tkm	[36]

Note that the use of “electricity, medium voltage” in the modeling reflects the intention to simulate conditions representative of a potential industrial-scale operation.

It is important to underline that, as reported in Tables 2.2-2.4, the APOS system model was chosen. This decision is grounded in the fact that the harvesting stage generally lacks recycling processes or significant outputs involving secondary materials. Therefore, APOS does not materially affect the results. Additionally, certain upstream components, such as steel used in vessels or netting material, may contain recycled content. APOS accounts for this through partial burden allocation, offering a more accurate representation of upstream processes.

For the AS formulation. In AS1, the incorporation of solar panels and inverters for partial electricity generation from solar energy was considered. Multi-Si photovoltaic panels from the Ecoinvent database were selected as input materials. Each panel consists of 60 solar cells ($156 \times 156 \text{ cm}^2$) with a capacity of 210 Wp, while the inverters, also sourced from Ecoinvent, have a capacity of 0.5 kW. The photovoltaic system was assigned a 25-year lifetime [158]. The input values, scaled according to the FU and useful life and adjusted to provide 50 % of the electricity demand, resulted in 0.243 m² of solar panels and 0.031 inverter units for ONC and 0.00042 m² of solar panels and 0.031 inverter units for OFC.

AS2 represents a modification of the ONC scenario, where artificial lighting is completely replaced by natural sunlight. As a result, energy consumption for lighting was entirely eliminated in this scenario.

In AS3, the antifouling agent traditionally used in marine applications, which is copper oxide-based and recognized for its high environmental toxicity, was replaced with a more

environmentally friendly alternative. The new antifouling formulation was selected based on the research findings of Lin and Usino (2014) [151].

AS4 explores the potential reduction in fuel consumption for vessels operating in WH and OFC scenarios. In this alternative scenario, the diesel consumption per FU for WH was reduced to 4.5 liters, while for the OFC scenario, the fuel consumption was adjusted to 0.0224 kg/tkm of heavy fuel oil.

LCA on the full biorefinery system

The primary objective of this LCA analysis is to assess the environmental impact of the cascade biorefinery (CB) system proposed for the *F. lumbricalis* macroalgae, as extensively detailed in Chapter 2.1. Additionally, this study aims to compare the innovative cascade biorefinery model with two more conventional biorefinery configurations. The first alternative design (SPE) follows a single-product extraction approach, while the second (TLE) adopts a three-line extraction process, both of which are also described in Chapter 2.1.

To ensure a comprehensive assessment, two FUs have been established. The first functional unit (FU1) is defined to evaluate the environmental impact based on processing a specific amount of dry biomass. In this study, FU1 is set at “1 ton of DW biomass of *F. lumbricalis*”. The second functional unit (FU2) is based on the added-value products derived from the biorefinery process, corresponding to “20 kg of pigments, 100 kg of proteins, and 200 kg of carrageenan as final valuable outputs.” Notably, the significantly lower output yields in the TLE scenario require functional compensation to match the performance of the CB. To address this shortfall, commercially available substitutes were introduced to ensure functional equivalence. Specifically, disodium disulphite was selected as a proxy for the pigment fraction, replicating the functional properties of Allura Red AC [159], [160]; soybean-based protein feed was used to offset the protein deficit, and potato starch served as a thickening agent to substitute for carrageenan [161].

The adoption of dual functional units aligns with existing literature standards [162], [163], providing a more robust framework for evaluating different perspectives within the system. FU1 allows for an in-depth analysis of the critical environmental parameters and hot spots within the cascade biorefinery process, identifying the most impactful stages. Conversely, FU2 enables a comparative assessment of various system configurations, highlighting differences in efficiency and sustainability. This approach is also reflected in the study conducted by Nilsson et al. (2022) [163], 1 kg of sodium alginate produced from *S. latissima* was selected as the FU, demonstrating the applicability of FU2 in biorefinery system evaluations.

The calculations were carried out using SimaPro v9.5 software, incorporating data from the Ecoinvent 3.9.1 database.

- System boundaries and alternative scenarios

In general, the complete biorefinery system encompasses the WH scenario, which is thoroughly detailed in the previous subchapter as the upstream stage, followed by the

biomass processing phase, classified as the downstream stage, as described in the subsequent sections.

The system boundaries align with the configuration outlined in Fig.2.4, which defines the CB as the baseline case. To assess how modifications in key parameters influence the environmental performance of this system, several AS were developed. These variations were designed to identify the most effective technological solutions for minimizing the environmental footprint of the *F. lumbricalis* cascade biorefinery. The selection of these scenarios was driven by critical factors, such as energy consumption and end-of-life biomass utilization, both of which play a central role in shaping the overall sustainability of the process. Specifically:

1. AS1 - It evaluates the integration of biogas production as an end-of-life valorization strategy for residual biomass from carrageenan extraction, rather than its conventional use as a biostimulant. This approach follows the methodology outlined by Fasahati et al. (2022) [123], detailing a two-stage process where biomass undergoes anaerobic digestion to produce biogas, which is subsequently converted into electricity via a boiler/turbogenerator system.
2. AS2 - It introduces a drying stage in the biorefinery process to prevent biomass degradation and facilitate transportation when immediate processing is not feasible. In this scenario, drying is applied after pigment extraction, based on laboratory findings from Vetik OÜ, which confirm that pigment yield is highest when extracted from fresh *F. lumbricalis*. The drying process is designed to reduce the water content of biomass to 20%, ensuring optimal preservation while mitigating the risk of degradation. Due to the lack of primary data on drying technology, the modeling was conducted using a standard drying process from the Ecoinvent 3.9.1 database, a methodology also adopted by Seghetta et al. (2016) [36] for drying *S. latissima* biomass.
3. AS3 - It investigates the adoption of a renewable energy mix instead of relying on Estonia's conventional electricity grid, which is largely fossil-fuel-dependent. The Renewable Energy Certificate System (REC) [164] was used as a reference, simulating a scenario where the biorefinery's electricity demand is met entirely by renewable energy sources. The energy mix composition was adapted from Ecoinvent 3.9.1; in Fig. 2.13, the mix is reported.
4. AS4 - It explores the feasibility of installing an on-site solar energy system at the biorefinery plant to generate electricity internally, thereby reducing dependence on external power sources and lowering the environmental impact. The scenario incorporates photovoltaic panels and inverters, modeled using data from the Ecoinvent 3.9.1 database

The system boundaries for SPE and TLE are instead detailed in Fig. 2.5 and Fig. 2.6, respectively.

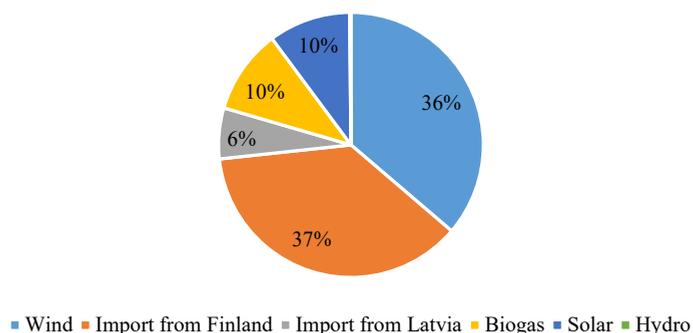


Fig. 2.13. Renewable energy mix adopted for the AS3.

- LCIA selected

For the LCA calculations, SimaPro v9.5 software [147] was employed, utilizing the ReCiPe2016 method (Hierarchist perspective, midpoint, and endpoint) [165]. This methodology encompasses 18 midpoint indicators, which are systematically grouped into three primary areas of protection: damage to human health, damage to ecosystems, and damage to resource availability. These categories are structured based on damage pathways, facilitating a more comprehensive interpretation when comparing different systems. The selection of the ReCiPe2016 method is justified by its broad scope, integrating multiple impact indicators derived from well-established methodologies. Table 2.5 provides a detailed overview of the damage categories included in this assessment.

Table 2.5

Explanation of the indicators included in the ReCiPe2016 method. From approbation publication No. 4 [135]

Indicator	Unit	Description	Area of protection
Particulate matter (PM)	Kg PM _{2.5} eq	It quantifies the impact of PM _{2.5} on health, including lung cancer and cardiovascular risks, using global and region-specific factors.	Human health
Tropospheric ozone formation (hum) (OFhh)	kg NO _x eq	It assesses the impact of ground-level ozone on human health, particularly respiratory issues, using global and region-specific characterization factors.	Human health
Ionizing radiation (IR)	kBq Co-60 eq	It evaluates the impact of radiation exposure on human health, considering different types of cancers and using time-horizon-specific characterization factors.	Human health
Stratospheric ozone depletion (SOD)	kg CFC11 eq	It measures the impact of ozone layer depletion on human health, particularly focusing on risks like skin cancer and cataracts across various time horizons.	Human health
Human toxicity (cancer) (HCT)	kg 1,4-DCB	It assesses the impact of chemical exposure on cancer risk in humans, using specific characterization factors for different chemicals.	Human health

Indicator	Unit	Description	Area of protection
Human toxicity (non-cancer) (HNCT)	kg 1,4-DCB	It evaluates the impact of chemical exposure on non-cancer health effects, using specific characterization factors for various substances.	Human health
Global warming (GWP)	kg CO ₂ eq	It measures the impact of greenhouse gas emissions on climate change, expressed as CO ₂ equivalents over various time horizons.	Human health and ecosystems
Water use (WU)	m ³	It evaluates the impact of water consumption on human health and ecosystems using region-specific characterization factors.	Human health and ecosystems
Freshwater ecotoxicity (ECF)	kg 1,4-DCB	It assesses the impact of toxic substances on freshwater ecosystems, using characterization factors that reflect the potential harm to aquatic life.	Ecosystems
Freshwater eutrophication (EF)	kg P eq	It measures the impact of nutrient enrichment, particularly phosphorus, on freshwater ecosystems, leading to issues like algal blooms and oxygen depletion.	Ecosystems
Tropospheric ozone formation (eco) (OFt)	kg NO _x eq	It assesses the impact of ground-level ozone on terrestrial ecosystems, focusing on damage to plant life and overall ecosystem health.	Ecosystems
Terrestrial ecotoxicity (TE)	kg 1,4-DCB	It evaluates the impact of toxic substances on terrestrial ecosystems, using characterization factors to assess potential harm to soil organisms and plant life.	Ecosystems
Terrestrial acidification (TA)	kg SO ₂ eq	It measures the impact of acidifying emissions, such as SO ₂ , on soil quality and plant life, leading to potential ecosystem degradation.	Ecosystems
Land use/transformation (LU)	m ² a crop eq	It assesses the impact of land occupation and conversion on biodiversity, focusing on species loss and ecosystem disruption.	Ecosystems
Marine ecotoxicity (MEc)	kg 1,4-DCB	It evaluates the impact of toxic substances on marine ecosystems, using characterization factors to assess potential harm to marine life.	Ecosystems
Marine eutrophication (ME)	kg N eq	It measures the impact of nutrient enrichment, particularly nitrogen, on marine ecosystems, leading to issues like algal blooms and oxygen depletion.	Ecosystems
Mineral resources (MRS)	kg Cu eq	It assesses the impact of mineral extraction on resource scarcity, expressed in terms of increased future extraction costs.	Resource availability
Fossil resources (FRS)	kg oil eq	It measures the impact of fossil fuel extraction on resource scarcity, focusing on the depletion of reserves and increased future extraction costs.	Resource availability

- LCI

The inventory data used is based on primary data sources, obtained from Vetik OÜ and NOFIMA. These organizations, both partners in the TACO ALGAE, provided direct data for the WH and the biorefinery extraction stages, respectively. In cases where primary data were unavailable, the inventory was supplemented using secondary data sources, including the Ecoinvent database v3.9.1 and published literature sources, particularly for aspects such as biogas production.

To account for the energy consumption associated with scaling up the pilot-level processes conducted by NOFIMA to the FU level, formulas and reference values from the

literature were employed [156]. Table 2.6 provides the technical data about the types of machinery used.

Table 2.6

Technical information on the technical equipment used for the biorefinery. From approbation publication No. 4 [135]

Equipment	Commercial name	Technical information	Others	Reference
Mixer	Blaastrom Ydra	22 kW power	Capacity 1 ton/h	Primary data [156]
Tricanter	Flottweg Z3E	10 kWh/ton		
Centrifuge	Alfa Laval Separator AFPX 810	10 kWh/ton		
Microfiltration	Alfa Laval 6,3 UF/MF	5 kWh/ton	It is assumed that it separates half of the water input	[156]
Ultrafiltration		10 kWh/ton		[155]

For the heating and drying processes, equations (2.1) and (2.2) were used. As reported by Piccinno et al. (2016) [156].

$$Q_{heat} = C_p * m_{mix} * (T_r - T_0) \quad (2.1)$$

$$Q_{dry} = \frac{C_{p,liq} * m_{liq} * (T_{boil} - T_0) + \Delta H_{vap} * m_{vap}}{\eta_{dry}} \quad (2.2)$$

Noticing these aspects, the electricity consumption for the CB design has been calculated (Table 2.7).

Table 2.7

Electricity calculation for the CB case. From approbation publication No. 4 [135]

<i>1st line (pigments extraction)</i>									
Mass IN	15.384 tons								
Mixing	338.45 kWh	P	22 kW						
Tricanter	153.84 kWh	P	10 kWh/ton						
Centrifuge	153.84 kWh	P	10 kWh/ton						
Mass IN	7.712 tons								
Microfiltration	38.56 kWh	P	5 kWh/ton						
Mass IN	3.866 tons								
Ultrafiltration	38.66 kWh	P	10 kWh/ton						
<i>2nd line (proteins extraction)</i>									
Mass IN	23.016 tons								
Heating	1.44E+09 J	C _p	4.18E+06 J/(t K)	m_{mix}	23.016 tons	T_r	313.5 K	T₀	298.1 K
Tricanter	230.16 kWh	P	10 kWh/ton						
Centrifuge	17.33 kWh	P	52 kW						
Mass IN	15.44 tons								
Microfiltration	154.44 kWh	P	10 kWh/ton						
									Note, this value was provided by NOFIMA, time of reaction 20 min
<i>3rd line (carrageenan extraction)</i>									
Mass IN	22.716 tons								
Heating	6.83E+09 J	C _p	4.18E+03 J/(kg K)	m_{mix}	22,716 kg	T_r	368.1 K	T₀	313.1 K
Tricanter	227.16 kWh	P	10 kWh/ton					n	0.765
Drying	3.94E+05 kJ	C _{p,liq}	4.18E+03 kJ/(t K)	m_{liq}	15.344 tons	T_{boil}	373.1 K	T₀	368.1 K

ΔH_{vap}	2.26E+0	m_{vap}	15.14	n_{dry}	0.9
	kJ/t		tons		

Table 2.8 shows the electricity calculation for the SPEp (only pigment).

Table 2.8

Electricity calculation for the SPEp. From approbation publication No. 4 [135]

<i>Single-product extraction of pigments</i>			
Mass IN	15.384 tons		
Mixing	338.45 kWh	P	22 kW
Tricanter	153.84 kWh	P	10 kWh/ton
Centrifuge	153.84 kWh	P	10 kWh/ton
Mass IN	7.712 tons		
Microfiltration	38.56 kWh	P	5 kWh/ton
Mass IN	3.866 tons		
Ultrafiltration	38.66 kWh	P	10 kWh/ton

Table 2.9 shows the electricity calculation for the SPEpr (only proteins).

Table 2.9

Electricity calculation for the SPEpr. From approbation publication No. 4 [135]

<i>Single-product extraction of proteins</i>								
Mass IN	23.076 tons							
Heating	1.45E+09 J	C_p	4.18E+06	m_{mix}	23.076 tons	T_r	313.5 K	T_0 298.15 K
			J/(t K)					
Tricanter	230.76 kWh	P	10 kWh/ton					
Centrifuge	17.33 kWh	P	52 kW					Note, this value was provided by NOFIMA, time of reaction 20 min
Mass IN	15.484 tons							
Microfiltration	154.84 kWh	P	10 kWh/ton					

Table 2.10 shows the electricity calculation for the SPEc (only carrageenan).

Table 2.10

Electricity calculation for the SPEc1-only carrageenan case. From approbation publication No. 4 [135]

<i>Single-product extraction of carrageenan</i>									
Mass IN	22.076 tons								
Heating	6.94E+09 J	C_p	4.18E+03	m_{mix}	23,076 kg	T_r	368.1 K	T_0	313.1 K
			J/(kg K)						
Tricanter	230.70 kWh	P	10 kWh/ton					n	0.765
Drying	4.01E+05 kJ	C_p	4.18E+03	m_{liq}	15.584	T_{boil}	373.1 K	T_0	368.1 K
		.liq	kJ/(t K)	tons					
				ΔH_{vap}	2.26E+03	m_{vap}	15.38	n_{dry}	0.9
				kJ/t		tons			

Table 2.11 shows the electricity calculation for the TLE.

Table 2.11

Electricity calculation for the TLE. From approbation publication No. 4 [135]

<i>Line 1 (pigment)</i>			
Mass IN	5.128 tons		
Mixing	112.82 kWh	P	22 kW

Tricanter	51.28 kWh	P	10 kWh/ton						
Centrifuge	51.28 kWh	P	10 kWh/ton						
Mass IN	2.584 tons								
Microfiltration	12.92 kWh	P	5 kWh/ton						
Mass IN	1.302 tons								
Ultrafiltration	1.302 kWh	P	10kWh/ton						
Line 2 (proteins)									
Mass IN	7.692 tons								
Heating	4.82E+08 J	C _p	4.18E+06 J/(t K)	m_{mix}	7.692 tons	T_r	313.5 K	T₀	298.1 K
Tricanter	76.92 kWh	P	10 kWh/ton						
Centrifuge	17.33 kWh	P	52 kW						
Mass IN	5.228 tons								Note, this value was provided by NOFIMA, time of reaction 20 min
Microfiltration	52.28 kWh	P	10 kWh/ton						
Line 3 (carrageenan)									
Mass IN	7.692 tons								
Heating	2.31E+09 J	C _p	4.18E+03 J/(kg K)	m_{mix}	7,692 kg	T_r	368.15 K	T₀	313.15 K
Tricanter	76.92 kWh	P	10 kWh/ton					n	0.765
Drying	1.37E+05 kJ	C _{p,liq}	4.18E+03 kJ/(t K)	m_{liq}	5.328 tons	T_{boil}	373.15 K	T₀	368.15 K
				ΔH_{vap}	2.26E+03 kJ/t	m_{vap}	5.128 tons	n_{dry}	0.9

With the determination of specific energy consumption values, the LCI was finalized. The upstream processes associated with WH are identical to those outlined in Table 2.2 and remain unchanged across all scenarios, including the CB, SPE, and TLE. Likewise, the pre-treatment phase and the biorefinery plant dimensions are consistent for all configurations.

In this case, the difference regards the *cut-off* model adoption for the Ecoinvent database. Given the circular configuration of the biorefinery, which includes the valorization of residual biomass through pathways like fertilizer production or biogas generation, the cut-off model prevents the overestimation of environmental benefits that might otherwise result from double-counted recycling credits. This approach ensures a more conservative and attributional portrayal of the system.

Consequently, Table 2.12 compiles the data for these two stages, facilitating a standardized comparison across the different system designs. The values presented in the dataset are normalized according to the FUs and the lifetime of the equipment, where applicable. The dataset corresponds to the total *F. lumbricalis* harvest for the year 2021, amounting to 181 tons of FW.

Table 2.12

LCI for the pre-treatment and biorefinery plant. This is independent of the design selected.

From approbation publication No. 4 [135]

	Category	Material/ component	Input on SimaPro software	Quantity	Unit	Source
Pre-treatment						
<i>Inputs</i>	Raw materials	Water for washing	Tap water {Europe without Switzerland} market for tap water Cut-off, U	7.69E+04	kg	Ecoinvent 3.9.1
<i>Outputs</i>	Waste	From washing	Wastewater, average {Europe without Switzerland} market	76.92	m ³	Ecoinvent 3.9.1

Category	Material/ component	Input on SimaPro software	Quantity	Unit	Source	
		for wastewater, average Cut-off, U				
Biorefinery plant						
<i>Inputs</i>	Raw materials	Biorefinery plant	Chemical factory, organics {RER} chemical factory construction, organics Cut-off, U	4.00E-07	p	Ecoinvent 3.9.1

The values in the LCI vary depending on the selected biorefinery design. Each configuration has distinct input and output values reflecting differences in resource consumption, energy use, and extraction efficiencies. Table 2.13 presents the LCI specifically for the CB, detailing the material and energy flows associated with this system.

The inventory does not account for the materials used in constructing the extraction machinery, as their contribution to the overall environmental impact is negligible. Additionally, since NOFIMA operates these machines for various experimental purposes, isolating their specific usage for the *F. lumbricalis* biorefinery was not feasible.

For SPE and TLE, the same processes as those applied in the CB system were used within the SimaPro software. However, variations in input values reflect differences in energy consumption and material flows across the scenarios. To determine the energy consumption values for each design, reference should be made to Table 2.8, Table 2.9, and Table 2.10 for SPE, while Table 2.11 provides the data for TLE.

The corresponding mass input and output flows are detailed in the mass balance tables in Chapter 2.1, where Fig. 2.7 illustrates the mass distribution for SPE, and Fig. 2.8 presents the mass balance for TLE. These references ensure an accurate allocation of resources and highlight process variations between the different biorefinery designs, facilitating a comparative evaluation of their environmental impacts.

Table 2.13
LCI for the CB. From approbation publication No. 4 [135]

Category	Material/ component	Input on SimaPro software	Quantity	Unit	Source	
1st line (pigments extraction)						
<i>Inputs</i>	Raw materials	Water for mixing	Tap water {Europe without Switzerland} market for tap water Cut-off, U	7,692	kg	Ecoinvent 3.9.1
	Energy	Mixing	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	338.45	kWh	Ecoinvent 3.9.1
		Tricanter	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	153.84	kWh	Ecoinvent 3.9.1
		Centrifuge	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	153.84	kWh	Ecoinvent 3.9.1
		Microfiltration	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	38.56	kWh	Ecoinvent 3.9.1

Category	Material/ component	Input on SimaPro software	Quantity	Unit	Source	
<i>Outputs</i>		Ultrafiltration	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	38.56	kWh	Ecoinvent 3.9.1
	Raw materials, waste	Wastewater	Wastewater, average {Europe without Switzerland} market for wastewater, average Cut-off, U	7.692	m3	Ecoinvent 3.9.1
2nd line (proteins extraction)						
<i>Inputs</i>	Raw materials	Enzyme	Enzymes {GLO} market for enzymes Cut-off, U	7.35	kg	Ecoinvent 3.9.1
<i>Inputs</i>		KOH	Potassium hydroxide {GLO} market for potassium hydroxide Cut-off, U	5.88	kg	Ecoinvent 3.9.1
		Water	Tap water {Europe without Switzerland} market for tap water Cut-off, U	15,344	kg	Ecoinvent 3.9.1
	Energy	Enzymatic extraction	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	802	kWh	Ecoinvent 3.9.1
		Decantation	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	230.16	kWh	Ecoinvent 3.9.1
		Centrifuge	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	17.33	kWh	Ecoinvent 3.9.1
		Microfiltration	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	154.44	kWh	Ecoinvent 3.9.1
<i>Outputs</i>	Raw materials, waste	Wastewater	Wastewater, average {Europe without Switzerland} market for wastewater, average Cut-off, U	15.356	m ³	Elementary flow
3rd line (carrageenan extraction)						
<i>Inputs</i>	Raw materials	KOH	Potassium hydroxide {GLO} market for potassium hydroxide Cut-off, U	5.28	kg	Ecoinvent 3.9.1
		Water	Tap water {Europe without Switzerland} market for tap water Cut-off, U	15,144	kg	Ecoinvent 3.9.1
	Energy	Heating	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	1,897	kWh	Ecoinvent 3.9.1
		Decantation	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	227	kWh	Ecoinvent 3.9.1
		Drying	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	109.57	kWh	Ecoinvent 3.9.1
<i>Outputs</i>	Emissions to air	Water to air	Water_Air emissions	15,144	kg	Elementary flow
Fertilizers production						
<i>Outputs</i>	Avoided products	Nitrogen fertilizer	Inorganic nitrogen fertiliser, as N {EE} market for inorganic nitrogen fertiliser, as N Cut-off, U	25.84	kg	Ecoinvent 3.9.1
		Potassium fertilizer	Inorganic potassium fertiliser, as K2O {EE} market for	3.06	kg	Ecoinvent 3.9.1

Category	Material/ component	Input on SimaPro software	Quantity	Unit	Source
	Phosphorous fertilizer	inorganic potassium fertiliser, as K2O Cut-off, U Inorganic phosphorus fertiliser, as P2O5 {EE} market for inorganic phosphorus fertiliser, as P2O5 Cut-off, U	26.52	kg	Ecoinvent 3.9.1

Note that the use of “electricity, medium voltage” in the modeling reflects the intention to simulate conditions representative of a potential industrial-scale operation.

For AS1, an LCI model for biogas production has been developed and is presented in Table 2.14. This inventory accounts for all relevant inputs and outputs associated with the anaerobic digestion process, including substrate preparation, biogas yield, and energy recovery through electricity cogeneration. The data used in this modeling are based on literature references [123] and adapted to align with the *F. lumbricalis* biorefinery framework, ensuring consistency with the overall system assessment.

Table 2.14

LCI for the AS1. From approbation publication No. 4 [135]

Category	Material/ component	Input on SimaPro software	Quantity	Unit	Source		
End-of-life biogas production							
<i>Inputs</i>	Raw materials	Air	Inputs from nature	2,504.44	kg	Elementary flow	
		Lime	Lime, packed {Europe without Switzerland} market for lime, packed Cut-off, U	8.42	kg	Ecoinvent 3.9.1	
<i>Outputs</i>	Emissions to soil	NH ₃ leakage	Ammonia	0.01156	kg	Elementary flow	
		H ₂ S leakage	Hydrogen sulfide	0.09724	kg	Elementary flow	
	Emissions to air	Methane leakage	Methane	18.5	kg	Elementary flow	
		NH ₃	Ammonia, EE	0.4056	kg	Elementary flow	
		SO ₂	Sulfur dioxide, EE	0.50898	kg	Elementary flow	
		NO ₂	Nitrogen dioxide, EE	6.16E-04	kg	Elementary flow	
	Waste and wastewater	Waste gypsum	CO	Carbon dioxide	3.75E-05	kg	Elementary flow
			Waste gypsum {Europe without Switzerland} market for waste gypsum Cut-off, U	12.44	kg	Ecoinvent 3.9.1	
		Waste lime	Limestone residue {CH} market for limestone residue Cut-off, U	1.6932	kg	Ecoinvent 3.9.1	
		Combustion ash	Digester sludge {CH} treatment of, municipal incineration with fly ash extraction APOS, U	14.28	kg	Ecoinvent 3.9.1	
Digestate		Refinery sludge {Europe without Switzerland} treatment of refinery sludge, landfarming Cut-off, U	241.40	kg	Ecoinvent 3.9.1		
Wastewater	Wastewater, average {Europe without Switzerland} market for wastewater, average Cut- off, U	7.82	kg	Ecoinvent 3.9.1			

Category	Material/ component	Input on SimaPro software	Quantity	Unit	Source
Avoided products	Ammonia	Urea ammonium nitrate mix {RER} market for urea ammonium nitrate mix Cut-off, U	0.007956	kg	Ecoinvent 3.9.1
	Inorganic phosphate	Inorganic phosphorus fertiliser, as P2O5 {EE} market for inorganic phosphorus fertiliser, as P2O5 Cut-off, U	0.007956	kg	Ecoinvent 3.9.1
Outputs	Electricity	Electricity, medium voltage {EE} market for electricity, medium voltage Cut-off, U	428.4	kWh	Ecoinvent 3.9.1

For AS2, a drying process available in the Ecoinvent 3.9.1 database ("*Drying of grass {ESTONIA}| processing | Cut-off, U*") was selected and modified to reflect Estonia's electricity mix. This adaptation ensures that the energy consumption accurately represents the regional energy profile, aligning the process with the environmental conditions of the study.

For AS4, the electricity production process chosen for solar panel energy generation is "*Electricity, low voltage {FI}| electricity production, photovoltaic, 3 kWp slanted-roof installation, multi-Si, panel, mounted | Cut-off, U.*" This energy source replaces conventional grid electricity in all biorefinery stages where standard electricity consumption is considered, thereby evaluating the potential reduction in environmental impact from integrating renewable energy into the system.

- Sensitivity analysis

To strengthen the reliability of the findings, a Monte Carlo analysis was conducted, targeting a $\pm 20\%$ variation in electricity consumption for the extraction processes, a critical parameter characterized by both high uncertainty and substantial environmental impact. The analysis involved 10,000 simulations, employing a normal distribution centered on a mean electricity consumption. The resulting values were then integrated into the LCA model to assess the overall environmental impacts across the three evaluated biorefinery designs (i.e., CB, SPE, and TLE).

2.3. Life cycle costing

LCC is a methodological approach used to assess the total costs associated with a product, process, or service throughout its entire life cycle. Unlike conventional cost analyses that focus solely on initial production or purchase expenses, LCC encompasses all financial aspects, including maintenance, operational costs, disposal, and end-of-life expenditures. Its primary objective is to provide a holistic view of the economic implications of a product or service, enabling more informed financial decision-making [166].

The methodology is structured into several key phases, which follow the approach of a conventional LCA. The first phase, scope definition, establishes the FU and system

boundaries, which are essential for determining the elements to be included in the analysis and ensuring the comparability of results. The second phase, data collection, involves identifying and quantifying costs incurred at each life cycle stage. This includes direct expenses such as raw materials, labor, and energy, as well as indirect costs like administrative overhead, transportation, and operational expenditures. The accuracy and reliability of collected data play a crucial role in ensuring the robustness of the assessment.

The third phase, cost analysis, aggregates the financial data to calculate the total costs over the product's life span. This step helps in evaluating different economic scenarios and identifying cost-efficient solutions. The final phase, interpretation of results, focuses on uncovering potential savings and optimization opportunities, supporting decision-making processes aimed at improving financial performance and resource allocation.

LCC is widely applied across various sectors, including project management, corporate sustainability strategies, and public procurement [166]. However, it presents notable challenges, particularly in estimating future costs due to uncertainties related to inflation, fluctuating interest rates, and raw material price volatility. Additionally, integrating economic assessments with environmental considerations requires combining LCC with methodologies such as LCA, which quantifies environmental impacts and links them to economic costs. This multidisciplinary approach is essential for fostering sustainable decision-making that accounts for both financial feasibility and environmental responsibility.

This chapter presents the methodology and data utilized to model the LCC assessment within the framework of the *F. lumbricalis* biorefinery system. It details the scope definition, cost categories considered, and the approach taken to quantify economic factors associated with each phase of the biorefinery process. The analysis includes direct costs, such as raw materials, energy consumption, and labor, as well as indirect costs, including transportation, maintenance, and equipment depreciation. Furthermore, assumptions related to the economic lifespan of the biorefinery infrastructure and potential financial uncertainties are addressed. By integrating these factors, the LCC evaluation provides a comprehensive understanding of the financial feasibility and long-term sustainability of the proposed biorefinery model.

Proposed LCC framework

There is currently no universally established framework for conducting LCC analyses specifically tailored to macroalgae biorefineries with standardized guidelines. To bridge this gap, this Thesis proposes a reproducible methodology, structured into a series of macro-steps that can be applied to similar research. Comparable approaches have been explored in the literature, such as the work of Thomassen et al. (2016) [167], which, despite focusing on microalgae biorefineries, offers a structured methodology for techno-economic assessments.

The LCC framework developed in this study comprises four key steps:

1. Technological scheme definition

It involves outlining the technological designs under evaluation, constructing a detailed technological flow diagram, and verifying the mass balances. This step, already described in the previous sections (i.e., the definition of CB, SPE, and TLE), ensures consistency in system boundaries and process inputs.

2. Inventorying

It focuses on compiling a comprehensive dataset necessary for the LCC analysis. This builds upon the inventory developed in the LCA study, refining it with additional cost-related data required for economic evaluation. In this study, the inventorying process was conducted using an Excel-based spreadsheet, allowing for systematic data organization and parameter tracking.

3. Economic assessment

It includes calculating core financial indicators for each biorefinery configuration, including return on investment (ROI), net present value (NPV), internal rate of return (IRR), and profit index (PI). These metrics provide a quantitative basis for assessing the economic feasibility of different process designs.

4. Sensitivity assessment

It entails creating alternative scenarios to test the robustness of the LCC results. These scenarios account for market dynamics, such as fluctuations in product pricing and variations in operational costs, to evaluate their impact on the overall economic viability of the system.

This structured approach enhances the transparency and reproducibility of the LCC methodology, ensuring that it can be adapted and applied to other macroalgae biorefinery studies. By integrating detailed cost assessments with technological and environmental considerations, this framework supports more comprehensive decision-making in the development of sustainable and economically viable macroalgae-based biorefineries.

LCC definition

As outlined in the introduction of this chapter, the proposed LCC follows the standardized LCA structure to ensure consistency and comparability when analyzing the final sustainability index in the LCSA. By aligning the LCC methodology with the LCA framework, the assessment facilitates a seamless integration of economic and environmental evaluations, enabling a more comprehensive understanding of the overall sustainability performance of the *F. lumbricalis* biorefinery system.

- Goal and scope

The primary objective of this LCC study is to assess the economic feasibility of five distinct *F. lumbricalis* biorefinery designs, CB, SPEp (pigment), SPEpr (protein), SPEc (carrageenan), and TLE, by determining the most viable solution and identifying key economic parameters that influence each configuration. The FU for this analysis is set at “1,000 and 2,000 DW of *F. lumbricalis* processed within the biorefinery.” These quantities align with the current harvesting license limit of 1,000 tonnes in Kassari Bay, Estonia, while also considering a potential expansion to 2,000 tonnes in the future. Defining the geographical boundaries of the system is crucial, as it directly impacts economic factors

such as labor costs, land expenses, and utility rates, all of which are fundamental to the cost assessment. For this study, the biorefinery facilities are assumed to be located in Estonia, ensuring that region-specific economic conditions are accurately reflected in the analysis.

- Life-cycle inventory for the economic analysis

The economic life-cycle inventories (e-LCI) for this analysis were derived from the LCI used in the LCA, where the original values, expressed in terms of mass, energy, and volume inputs, were converted into monetary values (EUR) and normalized based on the defined FU. The e-LCI is structured into three key macro-categories for economic evaluation:

- Operating costs. Quantified using multiple data sources, including Orbis [168] for publicly available commercial data on fuel, electricity, and consumables, as well as specific cost figures provided by Vetik OÜ. The analysis considered Estonia's manufacturing sector under NACE code 20.5 ("other chemical products"), where cost impacts were calculated through filtered datasets to ensure relevance. Labor costs were estimated using Teatmik [169], referencing financial data from seaweed-related companies in Estonia.
- Investments. Assessed through detailed process evaluations conducted in collaboration with Alfalaval [170], a Swedish company specializing in separation technologies, heat transfer, and fluid handling. This consultation provided insights into the optimal machinery sizing required for processing 1,000 tonnes of *F. lumbricalis* while ensuring scalability for an increased input capacity of up to 2,000 tonnes. The selection of equipment, its specifications, and properties are detailed in the next subchapter.
- Product sales. The pricing structure was established through a market analysis, referencing analogous products. For pigments, the valuation was based on phycocyanin rather than phycoerythrin, as the latter necessitates additional processing steps and higher purity levels that are currently unattainable within the studied biorefinery system. This choice to prioritize the more valuable product also reflects recommendations derived from the microalgae cultivation case study [142], reinforcing the rationale behind the valuation approach.

Protein pricing was determined by aligning with the market value of soy-protein feed commonly used in animal nutrition, while carrageenan was evaluated against commercial gelling agents typically employed in the cosmetics industry. The final biorefinery residues were assigned a minimal market value and sold to biogas producers as a means to mitigate waste treatment and disposal costs. Notably, the option of utilizing the residual material as a fertilizer was deemed unfeasible from laboratory tests further developed during the TACO algae project due to the extensive multi-stage extraction process, which significantly depleted the mineral content of *F. lumbricalis*. Consequently, biogas production remained the only viable end-of-life solution for these residues.

Table 2.15 presents the e-LCI for the 1,000-ton case.

Table 2.15

e-LCI for the 1,000-ton case. * Unit costs are calculated as a lump-sum amount

Cost/ income items	Cost/ income inventory	Unit costs/ incomes	CB	SPEp	SPEpr	SPEc	TLE
<i>Operating costs</i>							
Seaweed harvesting	Diesel for harvesting	1.47 €/l	2,148 4 63 €	2,148 4 63 €	2,148 46 3 €	2,148 46 3 €	2,148 4 63 €
	Diesel for transport to the harbor	1.47 €/l	39,572 €	39,572 €	39,572 €	39,572 €	39,572 €
	Lubricant oil	16.99 €/kg	28,883 €	28,883 €	28,883 €	28,883 €	28,883 €
	Epoxy paint	27.51 €/kg	17,606 €	17,606 €	17,606 €	17,606 €	17,606 €
Seaweed harvesting	Antifouling agent	79.12 €/kg	16,615 €	16,615 €	16,615 €	16,615 €	16,615 €
	Working on boat	20.00 €/h	140,00 0 €	140,00 0 €	140,000 €	140,000 €	140,000 €
	Electricity for unloader	0.06 €/kW h	317 €	317 €	317 €	317 €	317 €
Seaweed transport	Diesel for transport	1.47 €/l	601,56 8 €	601,56 8 €	601,568 €	601,568 €	601,568 €
Pigment extraction	Water	2.18 €/m ³	167,68 5€	167,68 5€	167,685€	167,685 €	167,685 €
	Water	2.18 €/m ³	16,768 €	16,768 €	0.00 €	0.00 €	5,589 €
	Processing labor	11.61 €/h	111,45 6€	111,45 6€	0.00 €	0.00 €	111,456 €
	Electricity	0.06 €/kW h	433,95 0 €	43,395 0 €	0.00 €	0.00 €	14,479 €
Proteins extraction	Regular maintenance of equipment	11.61 €/h	111,45 6 €	111,45 6 €	0.00 €	0.00 €	111,456 €
	Enzyme	110.00 €/h	808,50 0 €	0.00 €	825,000 €	0.00 €	275,000 €
	KOH	1.16 €/kg	6,832 €	0.00 €	6,972 €	0.00 €	2,324 €
	Water	2.18 €/m ³	33,449 €	0.00 €	33,539 €	0.00 €	11,179 €
	Processing labor	11.61 €/h	111,45 6 €	0.00 €	111,456 €	0.00 €	111,456 €
	Electricity	0.06 €/kW h	72,235 €	0.00 €	70 794 €	0.00 €	24 871 €
	Regular maintenance of equipment	11.61 €/h	111,45 6 €	0.00 €	111,456 €	0.00 €	111,456 €
Carrageenan extraction	KOH	1.16 €/kg	6,135 €	0.00 €	0.00 €	6,972 €	2,324 €
	Water	2.18 €/m ³	33,013 €	0.00 €	0.00 €	33,539 €	11,179 €
	Processing labor	11.61 €/h	111,45 6 €	0.00 €	0.00 €	111,456 €	111,456 €
	Electricity	0.06 €/kW h	134,01 4 €	0.00 €	0.00 €	136,131 €	45,428 €
Management and administrative costs*	Office suppliers	€/unit	10,000 €	10,000 €	10,000 €	10,000 €	10,000 €
	Legal and accounting consultancy services	€/unit	45,000 €	35,000 €	35,000 €	35,000 €	45,000 €

Cost/ income items	Cost/ income inventory	Unit costs/ incomes	CB	SPEp	SPEpr	SPEc	TLE
Selling and packaging*	Security costs	€/unit	32,000 €	23,000 €	23,000 €	23,000 €	32,000 €
	Research and development costs	€/unit	170,00 0 €	110,00 0 €	110,000 €	110,000 €	170,000 €
	Commercial and marketing costs	€/unit	100,00 0 €	75,000 €	75,000 €	75,000 €	100,000 €
	Packaging materials and equipment	€/unit	125,00 0 €	70,000 €	70,000 €	70,000 €	125,000 €
<i>Investments</i>							
Seaweed harvesting*	Trawling boats	€/unit	100,00 0 €	100,00 0 €	100,000 €	100,00 0 €	100,000 €
	Nylon harvesting net	€/unit	16.15 €	16.15 €	16.15 €	16.15 €	16.15 €
	Fishing license	€/unit	35,000 €	35,000 €	35,000 €	35,000 €	35,000 €
Pigment extraction*	Electrical unloader	€/unit	1,900 €	1,900 €	1,900 €	1,900 €	1,900 €
	Decanter (Foodec 65)	€/unit	700,00 0 €	700,00 0 €	0.00 €	0.00 €	2,100,00 0 €
	High-Speed Separator (Clara 200 High Flow)	€/unit	260,00 0 €	260,00 0 €	0.00 €	0.00 €	260,000 €
Protein extraction*	Micro and Ultrafiltration batch	€/unit	250,00 0 €	250,00 0 €	0.00 €	0.00 €	250,000 €
	Decanter (Foodec 65)	€/unit	0.00 €	0.00 €	700,000 €	0.00 €	0.00 €
	High-Speed Separator (PurePulp 75)	€/unit	440,00 0 €	0.00 €	440,000 €	0.00 €	440,000 €
Carrageenan extraction*	Microfiltration batch	€/unit	250,00 0 €	0.00 €	250,000 €	0.00 €	250,000 €
	Decanter	€/unit	0.00 €	0.00 €	0.00 €	700,00 0 €	0.00 €
Construction and infrastructure*	Dryer	€/unit	20,000 €	0.00 €	0.00 €	20,000 €	20,000 €
	Cost of land	4.50 €/m ²	1,642 €	1,434 €	1,389 €	1,190 €	2,170 €
<i>Incomes (intermediate case scenario)</i>							
Products	Pigments	124,736 €/ton	2,494,7 15 €	2,494,7 15 €	0.00 €	0.00 €	831,987 €
	Protein	1,500 €/ton	150,00 0 €	0.00 €	150,000 €	0.00 €	49,950 €
	Carrageenan	15,000 €/ton	3,000,0 00 €	0.00 €	0.00 €	3,000,0 0 €	1,000,05 0 €
	Residues for biogas	10.00 €/ton	73,760 €	76,720 €	75,920 €	74,980 €	75,960 €

For the scenario involving 2,000 tonnes of *F. lumbricalis*, the values presented in Table 2.15 are proportionally doubled to reflect the increased processing scale.

- Economic analysis parameters

The economic analysis commenced with defining several fundamental parameters that significantly influence the final outcome. In particular, the proportion of investment

financed through equity versus external loans was determined, alongside the required return on equity and the applicable interest rate for external financing. These values were selected based on the average financial data retrieved from Orbis [168]. The selected parameters are summarized in Table 2.16.

Table 2.16

Capital costs definition

Macro category	Impact
Share of investment covered by equity (EV)	70 %
Remaining share of investment covered by external funds (DV)	30 %
Required rate of return from equity (Re)	10 %
Interest rate for external loan (Rd)	4.25 %

Once the key parameters were established, financial metrics were calculated over a 10-year period to assess the cost-effectiveness of each scenario. These metrics included investment, inbound cash flows, outbound cash flows, depreciation, taxable income, taxes, net cash flows, discounted cash flows, ROI, NPV, IRR, and PI.

Investment represents the initial financial commitment required to launch the biorefinery, encompassing capital costs for equipment, infrastructure, and technology. This factor significantly impacts cash flows and long-term profitability, making it a crucial criterion for scenario comparison [171]. In this Thesis, investments were distributed over the 10-year analysis period, considering the useful life of different assets: licenses (1 year), nylon nets (5 years), machinery and equipment (10 years), and trawling boats (20–30 years). These lifespans also dictated the depreciation rates applied in the economic model. Depreciation serves as an accounting method that systematically allocates the cost of long-term assets over their operational lifespan, ensuring that their diminishing value due to wear, tear, or obsolescence is reflected in financial calculations [172].

Inbound cash flows refer to the revenue generated by the biorefinery, mainly derived from product sales and, in some cases, financial incentives [173]. For this analysis, revenue was exclusively based on product sales to highlight scenarios with the highest economic potential. Outbound cash flows include all operational costs required to sustain the process, excluding depreciation. These costs encompass expenditures on raw materials, labor, and maintenance [174] and were computed as the total operating costs previously defined in the inventory.

Taxable income represents the net income subject to taxation, calculated by deducting eligible expenses, including depreciation and interest payments, from gross revenue [175]. In this Thesis, taxable income was determined as the difference between cash inflows and outflows, further adjusted for depreciation. The depreciation periods, aligned with asset lifetimes, were assumed to reflect the average depreciation periods used for taxation purposes.

Taxes, representing compulsory government-imposed levies on business profits, were calculated using a corporate tax rate of 20%, consistent with the standard taxation rate for businesses in Estonia [176].

Net cash flows denote the balance between cash inflows and outflows, further adjusted for taxation and investments. This metric provides insight into the financial sustainability of each scenario [177]. A positive net cash flow indicates profitability, while persistent negative cash flow suggests financial viability concerns, necessitating external funding or operational modifications. Discounted cash flows account for the time value of money, expressing future cash flows in present-day terms using a discount rate. In this study, the Weighted average cost of capital (WACC) was determined at 8.02%, calculated using the financial parameters presented in Table 2.16 and applying Equation 2.3.

$$WACC = (EV \times Re) + (DV \times Rd \times (1 - Tc)) \quad (2.3)$$

Finally, the four key economic indicators were assessed to evaluate the financial viability of each scenario.

1. ROI. It serves as a fundamental measure of profitability, representing the ratio of net profit to the total investment cost. It is calculated using Equation 2.4, where n denotes a single time period within the analysis and N represents the total number of time periods. Higher ROI values signify more economically advantageous scenarios, indicating greater profitability relative to the initial capital investment [178].

$$ROI = \frac{\sum_{n=0}^N \text{Net cash flow}_n}{\sum_{n=0}^N \text{Investment}_n} \quad (2.4)$$

2. IRR. It is the discount rate at which the NPV of all future cash flows of a project equals zero. It represents the rate of return that equates the present value of cash inflows to the initial investment, offering a measure of investment efficiency and profitability. A higher IRR indicates a more favorable investment opportunity, as it reflects a greater potential return relative to the cost of capital [178].
3. NPV. It provides the total present value of all future cash flows associated with a project, discounted at a specified rate of return, minus the initial investment cost. In life cycle analysis, NPV serves as a key metric for evaluating the financial feasibility and attractiveness of investment scenarios [178]. It is calculated using Equation 5, where n denotes the individual time period of analysis and N represents the total number of time periods. A positive NPV indicates that the projected earnings exceed the anticipated costs, making the investment financially viable, while a negative NPV suggests that the investment may not be economically sustainable.

$$NPV = \sum_{n=0}^N \frac{\text{Net cash flow}_n}{(1 + IRR)^n} \quad (2.5)$$

4. PI. It is used to assess the efficiency of an investment by comparing the present value of future cash flows to the initial investment. It is calculated as shown in Equation 2.6. A PI greater than 1 indicates that the investment is expected to generate value, as the present value of future cash flows exceeds the initial cost. Conversely, a PI less than 1 suggests that the investment may not be economically viable [178]. This metric is

particularly useful for ranking investment opportunities and determining whether a project should proceed based on its potential return relative to its cost.

$$PI = \frac{NPV}{\text{Initial investment}} \quad (2.6)$$

These economic parameters have proven to be a robust basis for conducting techno-economic assessments in algal systems. When applied to the microalgae cultivation case study [142], they enabled a clear identification of the most economically viable scenario, supported by a well-defined investment trend and an estimated return on investment within a specified timeframe. Moreover, the methodology effectively highlighted the economic advantages among different design options and helped identify critical process bottlenecks, such as capital investment requirements, equipment costs, and operational expenses like energy consumption, that significantly influence overall economic feasibility.

- Alternative scenarios

Given that the primary uncertainties in the analysis stem from product market prices, the sensitivity analysis focuses on these variables. Among the biorefinery products, pigment holds the highest unit price and plays a crucial role in determining the overall economic performance of the system. To account for potential market fluctuations, a range of pigment prices was defined based on findings from online market research. In addition to the intermediate scenario, where the pigment price was set at 124,736 €/ton (see Table 2.15), two benchmark scenarios were considered to assess the economic resilience of the biorefinery system under different market conditions.

- Worst case scenario (WCS). It assumes the lowest pigment unit price of 100,000 €/ton, combined with the minimum *F. lumbricalis* harvestable under the current license limits (1,000 tons).
- Best case scenario (BCS). It accounts for the highest pigment unit price of 500,000 €/ton, alongside the maximum harvestable quantity of *F. lumbricalis* under the extended license limits (2,000 tons).

Equipment information and plant dimension calculations for the LCC

This sub-chapter provides details on the equipment specifications shared by Alfalaval and the calculations performed for sizing the machinery required for the *F. lumbricalis* biorefinery.

The following figures illustrate the technical specifications and operational aspects of the selected machinery. The decanter chosen for the process, the Foodec 65, manufactured by Alfalaval, is depicted in Fig. 2.14. This equipment is fundamental for the separation of biomass from liquids, a critical step in extracting high-value products such as pigments, proteins, and carrageenan. The high efficiency and processing capacity of the Foodec 65 contribute to the overall productivity of the biorefinery while enhancing its economic viability by reducing waste and optimizing resource utilization.

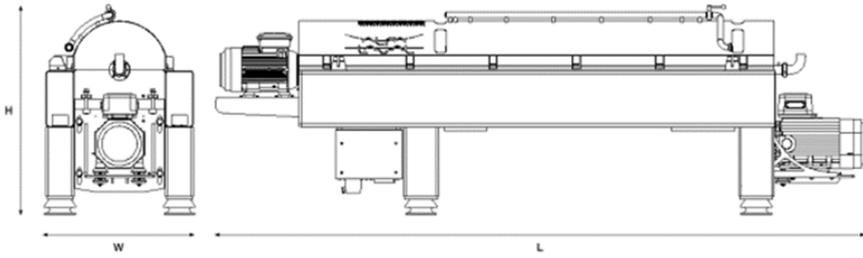


Fig. 2.14. Foodec 65 decanter. Picture provided by Alfalaval [170].

The second set of machinery selected for the *F. lumbricalis* biorefinery includes the PurePulp 750 and Clara 200 high-flow high-speed separators. These advanced separators are specifically designed for high-capacity operations, ensuring rapid and efficient separation of valuable components from the biomass. Their integration into the biorefinery process enhances the purity and quality of the extracted products, which is essential for their application in industries such as food, pharmaceuticals, and cosmetics. By incorporating these high-performance separators, the biorefinery can optimize product recovery and improve overall process efficiency. Fig. 2.15 provides detailed illustrations of the PurePulp 750.

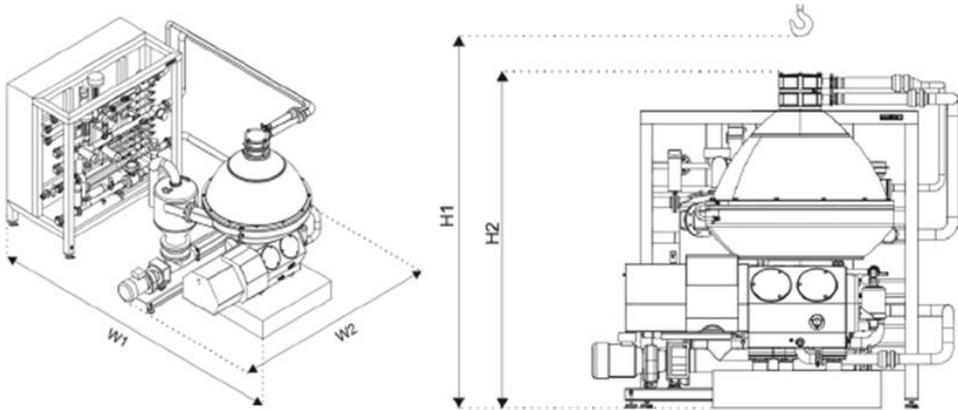


Fig. 2.15. Pure Pulp 750. Picture provided by Alfalaval [170].

Fig. 2.16 depicts the technical prospect for the Clara 200 High Flow high-speed separators.

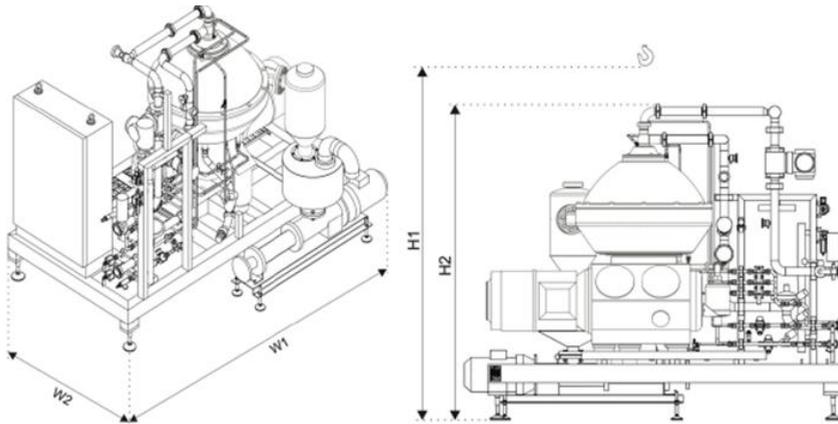


Fig. 2.16. Clara 200 High Flow high-speed separators. Picture provided by AlfaLaval [170].

For the filtration stage, filtering columns manufactured by AlfaLaval have been selected. These advanced filtration systems play a crucial role in eliminating impurities, thereby ensuring that the final products meet industrial and market standards. Their integration into the biorefinery process enhances product quality, making them suitable for applications in high-value industries such as pharmaceuticals, cosmetics, and food production. Investing in these cutting-edge filtration technologies is essential for improving the competitiveness and long-term sustainability of the biorefinery. Fig. 2.17 provides an illustration of the selected filtration equipment.

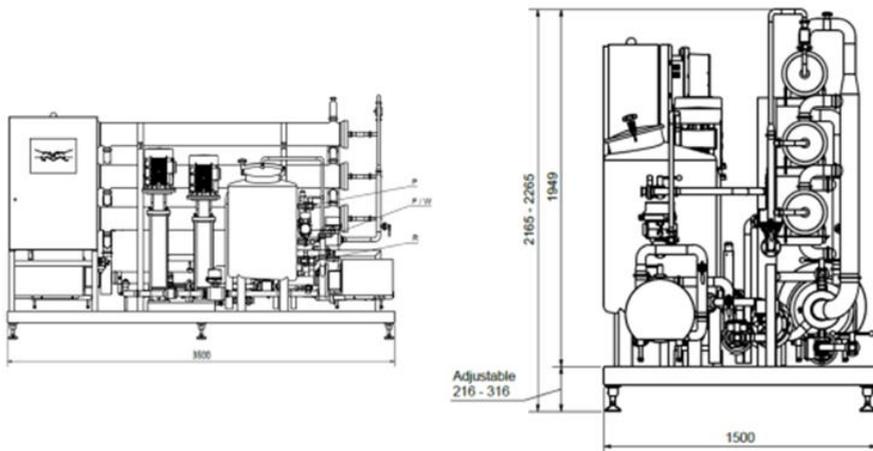


Fig. 2.17. Micro and ultrafiltration machinery. Picture provided by AlfaLaval [170].

The technical specifications of the selected machinery were utilized to estimate the total hypothetical area (m^2) required for the *F. lumbricalis* biorefinery. The calculation was based on determining the number of machines needed and summing their spatial requirements. To ensure adequate spacing for operational efficiency, safety, and maintenance, an additional

2 meters in both length and width were allocated per unit to accommodate operator passage corridors and access for servicing.

However, the drying equipment was excluded from this calculation due to the unavailability of detailed sizing information from AlfaLaval. Although an estimated price was incorporated into the financial assessment based on online sources, the absence of precise dimensional data led to its omission from the spatial analysis to avoid inaccuracies.

Table 2.17 presents a comprehensive breakdown of the estimated productive area requirements.

Table 2.17

Sizing of the productive area

Process	Machinery	Size (mm)	Area (m ²)	Total area with safety distances (m ²)
Pigment extraction	Decanter (Foodec 65)	6,502x1,450x1,791	9.42	29.33
	High-Speed Separator (Clara 200 High Flow)	2,470x1,905x2,500	4.70	17.46
	Micro and Ultrafiltration batch	3,500x1,500x2,300	5.25	19.25
Protein extraction	Decanter (Foodec 65)	6,502x1,450x1,791	9.42	29.33
	High-Speed Separator (PurePulp 75)	3,600x2,450x3,000	8.82	24.92
	Microfiltration batch	3,500x1,500x2,300	5.25	19.25
Carrageenan extraction	Decanter	6,502x1,450x1,791	9.43	29.33

The precise number of equipment units required for the biorefinery, determined based on the capacity of each machine, is provided in Table 2.18. However, due to the proprietary nature of AlfaLaval's machinery specifications, the specific capacity values are not disclosed. Instead, only the number of units necessary to process the defined functional unit (1,000 tons or 2,000 tons of *F. lumbricalis*) is reported. This approach ensures transparency in the biorefinery system design while maintaining compliance with confidentiality agreements regarding sensitive technical data.

Table 2.18

Number of equipment

Process	Machinery	CB	SPEp	SPEpr	SPEc	TLE
Pigment extraction	Decanter (Foodec 65)	2	2	0	0	6
	High-Speed Separator (Clara 200 High Flow)	2	2	0	0	2
	Micro and Ultrafiltration batch	1	1	0	0	1
Protein extraction	Decanter (Foodec 65)	0	0	2	0	0
	High-Speed Separator (PurePulp 75)	1	0	1	0	1
	Microfiltration batch	1	0	1	0	1
Carrageenan extraction	Decanter	0	0	0	2	0

The total space requirements for the biorefinery plant were estimated based on the number of machines needed for each production design. In addition to the primary processing area, the layout includes allocations for essential supporting facilities such as

main corridors, secondary corridors, office spaces, storage warehouses, packaging zones, and other necessary operational areas. These considerations ensure an optimized workflow, efficient material handling, and proper accessibility for maintenance and personnel movement. The final dimensions, incorporating all these factors, are detailed in Table 2.19, providing a comprehensive overview of the spatial requirements for the facility.

Table 2.19

System sizing						
Design	Process	Total size (m ²)	Main corridors (2m)	Secondary corridors (1m)	Space for other needs (m ²)	Total area (m ²)
CB	Pigment extraction	112.82	2	4	200	364.99
	Protein extraction	44.17				
	Carrageenan extraction	0.00				
SPEp	Pigment extraction	112.82	2	2	200	318.82
SPEpr	Protein extraction	102.83	2	2	200	308.83
SPEc	Carrageenan extraction	58.66	2	2	200	264.66
TLE	Pigment extraction	230.15	2	4	200	482.32
	Protein extraction	44.17				
	Carrageenan extraction	0.00				

Monetization of environmental externalities for the LCC

As this Thesis includes an LCA of the same system, an additional layer of novelty has been introduced by incorporating the monetization of environmental externalities into the economic evaluation. The adopted approach follows the methodology developed by Ponsioen et al. (2020) [179], which provides monetization factors for LCA results derived from both mid-point and end-point impact categories using the ReCiPe 2016 method, the same LCA framework is applied in this LCA of this thesis. Whenever mid-point values are available, they are prioritized over their corresponding end-point values to enhance accuracy and consistency in the economic assessment. The monetization factors applied in this study are summarized in Table 2.20.

Table 2.20

Monetization of environmental externalities			
Impact category	Categories	Unit of the environmental indicators	Value (€/unit)
Global warming	Midpoint	kg CO ₂ eq	0.15
Stratospheric ozone depletion	Endpoint	kg CFC11 eq	38.00
Ionizing radiation	Endpoint	kBq Co-60 eq	0.00061
Ozone formation, Human health	Endpoint	kg NO _x eq	0.066
Fine particulate matter formation	Midpoint	kg PM _{2.5} q	14.00
Ozone formation, Terrestrial ecosystems	Endpoint	kg NO _x eq	0.0093
Terrestrial acidification	Midpoint	kg SO ₂ eq	2.73
Freshwater eutrophication	Midpoint	kg P eq	2.00
Marine eutrophication	Midpoint	kg N eq	3.10
Terrestrial ecotoxicity	Endpoint	kg 1,4-DCB	0.00013

Impact category	Categories	Unit of the environmental indicators	Value (€/unit)
Freshwater ecotoxicity	Endpoint	kg 1,4-DCB	0.008
Marine ecotoxicity	Endpoint	kg 1,4-DCB	0.0012
Human carcinogenic toxicity	Endpoint	kg 1,4-DCB	0.24
Human non-carcinogenic toxicity	Endpoint	kg 1,4-DCB	0.016
Land use	Midpoint	m ² a crop eq	0.10
Mineral resource scarcity	Endpoint	kg Cu eq	0.20
Fossil resource scarcity	Endpoint	kg oil eq	0.39
Water consumption	Midpoint	m ³	0.045

2.4. Social life cycle assessment

Since the 1960s, awareness has grown regarding the limitations of continuous economic and industrial expansion within the constraints of a finite planet. This realization has led to the concept of sustainable development, emphasizing the need to meet present human needs without compromising the ability of future generations to do the same. Sustainability, as outlined in various international frameworks such as the United Nations Sustainable Development Goals, extends beyond environmental concerns to include social dimensions. This broader perspective has driven the evolution of LCA into S-LCA, which aims to evaluate the social impacts of products and systems throughout their life cycle [166].

S-LCA follows a methodological structure similar to environmental LCA, consisting of goal and scope definition, inventory analysis, impact assessment, and interpretation. However, while LCA focuses on environmental aspects such as resource use, emissions, and ecological damage, S-LCA evaluates how products and processes affect human well-being across different stakeholder groups, including workers, local communities, consumers, and decision-makers. The overarching objective is to assess the social implications of a product's life cycle and how they contribute to or detract from overall human well-being.

Despite its conceptual alignment with LCA, S-LCA is still in its early stages and lacks the standardization achieved by environmental LCA. While LCA has well-established guidelines, S-LCA remains methodologically fragmented. The most significant step toward its standardization has been the development of the Guidelines for S-LCA by the UNEP-SETAC Life Cycle Initiative [180], which aimed to establish a foundational structure for assessing social impacts. More recently, the introduction of ISO 14075, titled "Environmental Management - Principles and Framework for Social Life Cycle Assessment" [181], represents a significant step toward formalizing the methodology. Nevertheless, key challenges persist, particularly in the consistent definition and quantification of social impacts.

In S-LCA, the primary Area of Protection is human well-being, meaning that assessments seek to identify and characterize social changes caused by a product or system and evaluate their contributions to societal sustainability. Stakeholders considered in S-LCA typically include workers across the supply chain, affected local or regional communities, and consumers who use the final product. Additionally, other stakeholders,

such as company shareholders and decision-makers, may also be included if their well-being is affected by life cycle activities.

As interest in social sustainability continues to grow, S-LCA provides a crucial tool for integrating social considerations into sustainability assessments. However, further methodological advancements and harmonization efforts are required to enhance its reliability and applicability in decision-making processes.

In light of these challenges, this thesis proposes a hybrid methodology that integrates the latest advancements in S-LCA to ensure a comprehensive and up-to-date assessment approach applicable to the *F. lumbricalis* biorefinery system.

The methodological framework defined for the S-LCA

The S-LCA study conducted in this work is structured across two analytical scales, aligning with established methodologies in the literature [182]. This multi-scale approach enables a more comprehensive evaluation of potential social impacts, ensuring a broader set of indicators that would not be attainable through a single-scale assessment.

1. Impact Pathway Approach (IPA) - Scale 1. It operates at the macro-national level and facilitates a social hotspot analysis. This level of analysis aims to identify major potential social risks associated with the macroalgae sector, particularly concerning WH and OFC. Characterization models are employed to represent impact pathways through a causal-effect chain, linking quantitative inventory indicators, such as instances of child labor, to broader damage categories following a top-down approach. To implement this, the Social Hot Spot Database (SHDB) [183] within the SimaPro software is utilized, allowing for the identification of social “hotspots” associated with the modeled product system.
2. Reference Scale Approach (RSA) - Scale 2. It focuses on the small-enterprise level, offering a detailed social assessment of individual companies. This micro-level analysis provides insights that cannot be captured at the macro scale, allowing for a deeper understanding of the social dynamics within specific enterprises. This approach evaluates the social performance of organizational activities within the product system using Performance Reference Points (PRP). PRPs serve as benchmarks based on internationally recognized thresholds, objectives, or best practices (e.g., a semi-quantitative 0–5 scale). To support this assessment, tailored questionnaires will be developed to establish a set of indicators relevant to the *F. lumbricalis* biorefinery system.

This dual-scale methodology ensures a nuanced and context-sensitive evaluation of social impacts across different levels of the supply chain.

Description of the IPA framework application

The IPA closely mirrors the standard LCA methodology, incorporating key phases such as goal and scope definition, system boundary delineation, inventory analysis, and LCIA. This structured approach ensures that social impacts are systematically identified, quantified, and assessed within a defined product system. By establishing clear system

boundaries, the IPA facilitates a comprehensive evaluation of potential social risks and benefits, integrating inventory data with impact characterization models.

- Goal and scope

The primary goal is to explore the social dimensions associated with a macroalgae value chain, starting with the biomass collection methods and conducting an S-LCA of different macroalgae harvesting and cultivation techniques.

In this way, the specific objective of the research is to identify key social issues linked to the WH (i.e., reference scenario described in Chapter 2.1) of the red macroalgae *F. lumbricalis* at a national scale through a social hotspot analysis. WH was chosen for reference analysis as it is the predominant method for collecting this species [134] and is underrepresented in existing literature [184].

Additionally, the S-LCA includes an exploratory scenario involving a hypothetical OFC system for *F. lumbricalis*. This scenario was developed based on the literature information [136] to assess the potential social impacts of such a system. By incorporating this hypothetical scenario, the study broadens its scope, facilitating a more comprehensive social hotspot analysis. This approach enables the identification of both positive and negative social aspects of each system, providing insights beyond what a single-scenario analysis could achieve.

To ensure comparability across scenarios, the FU is defined as “1 USD generated during an 8-hour standard working day for each system.” This FU reflects the unique characteristics of the systems under study and establishes a consistent basis for assessment. Additionally, it is in accordance with the structure of the SHDB, which requires monetary inputs for its analysis.

- System description in the context of SHDB

The WH system described follows the same structure detailed in Chapter 2.1. A key additional consideration specific to the S-LCA definition is that, within the FU’s timeframe (i.e., one 8-hour working day), the total amount of fresh *F. lumbricalis* harvested is 10 tons.

For the OFC setup, the life cycle inventory modeling is based on literature sources [136], particularly on artificial substrate cultivation systems for *F. lumbricalis*. All data have been normalized to ensure the same biomass yield (10 tons) as in the WH scenario. The process begins with the collection of the initial inoculum of *F. lumbricalis* from Kassari Bay, which is the same harvesting area as in the WH scenario. The biomass is then transferred into specialized nylon mesh bags (ø 5.5 cm, height 20 cm) with an internal plastic frame and a mesh size of 1 mm, carefully selected to minimize light reduction within the bags. These seeded bags are submerged at a depth of 4 meters and left to grow for three months under optimal environmental conditions in Kassari Bay. During this period, the reported growth rate increases by 155 %, and routine maintenance of the cultivation lines is assumed to occur once per month. After the three-month cultivation period, the total biomass reaches 10 tons and is harvested following the same unloading procedures as in the WH scenario. Similar to WH, the OFC scenario accounts for an annual one-month maintenance period for the harvesting vessel. A schematic visualization is reported in Fig. 2.18.

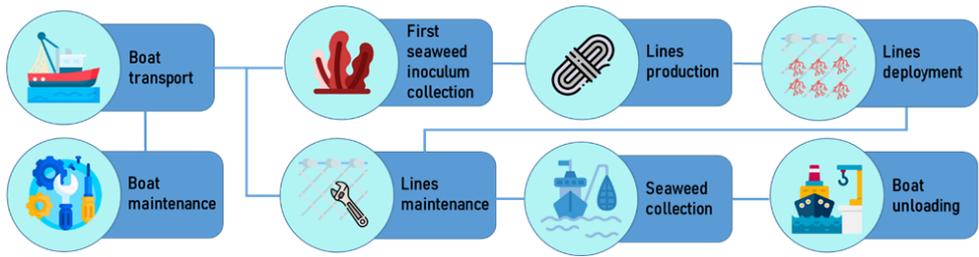


Fig. 2.18. System diagram for the OFC scenario.

Once the WH and OFC systems were defined, it became essential to describe the integration of the SHDB into this analysis. The SHDB was employed to identify and analyze potential social risks associated with the product supply chains, focusing on key areas where significant social issues may arise in both WH and OFC systems. As a comprehensive resource, the SHDB integrates data from global organizations such as the International Labour Organization and the World Bank to assess social risks related to multiple factors, including child labor, forced labor, inadequate working conditions, wages, and health and safety [183]. In total, 30 different social issues were evaluated in this study.

Widely applied in S-LCA, the SHDB uses a risk-based approach to assess social impacts, enabling users to identify high-risk regions within supply chains. In the results discussion, an example will be provided to illustrate how social impacts extend throughout the entire value chain. The SHDB covers a broad spectrum of social concerns across more than 150 industrial sectors and nearly 200 countries [183]. The database operates with transparency, relying on widely recognized data sources to generate reliable insights. Its application has been broadly utilized across various sectors. For instance, Backers and Traverso (2022) [185] employed the SHDB to map potential social risks in carbon-reinforced concrete supply chains across different countries, while Zamani et al. (2016) [186] applied it to identify social hotspots in the Swedish clothing industry.

For this study, the selected SHDB processes specifically pertain to Estonia, aligning with the defined system boundaries. The database was used to determine the sector that best represents each operation performed within the WH and OFC scenarios. This selection process was guided by the sector descriptions provided within the SHDB. However, since a direct one-to-one match for certain processes was not always available, some activities were represented using *proxy* processes. In these cases, the SHDB operation that most closely resembled the actual activity was selected as a substitute.

Table 2.21 outlines the SHDB processes chosen for the WH scenario. Some activities required the use of *proxy* processes to approximate real-world operations. For example, boat unloading is represented by the "manufactures nec (omf)/EST U" process, a broad manufacturing category that serves as a reasonable but non-specific *proxy* for boat unloading. Similarly, boat maintenance is modeled using the "trade (trd)/EST U" process, which covers general repair services but does not explicitly specify boat maintenance. However, key processes such as macroalgae harvesting and transportation to the harbor

were directly matched to more specific SHDB processes, ensuring an accurate reflection of their characteristics within the social impact assessment.

Table 2.21

SHDB processes description for the WH scenario

Process stage	SHDB process	Description from SHDB
<i>Seaweed harvesting</i>	Fishing (fsh)/EST U	Fishing in Estonia
<i>Transport to harbor</i>	Water transport (wtp)/EST U	Water transport in Estonia
<i>Boat unloading</i>	Manufactures nec (omf)/EST U	Other manufacturing Estonia
<i>Boat maintenance</i>	Trade (trd)/EST U	Retail, wholesale, and repair services in Estonia

Table 2.22 details the SHDB processes selected for the OFC scenario. In this case, the stages "first seaweed inoculum collection" and "seaweed collection" have been merged, as both involve the harvesting of *F. lumbricalis*. The proxy processes utilized in this scenario largely align with those applied in the WH scenario. Specifically, boat unloading and boat maintenance are represented using "manufactures nec (omf)/EST U" and "trade (trd)/EST U," respectively, ensuring consistency in modeling these operations.

Additionally, line deployment and line maintenance are mapped using the "Fishing (fsh)/EST U" proxy, as these activities closely resemble traditional fishing operations in terms of labor and resource demands. In contrast, certain processes, such as boat transport, first seaweed inoculum collection, and line production, are directly represented within the SHDB, eliminating the necessity for proxy substitutions. This selection approach ensures a balanced and comprehensive assessment of social risks, maintaining methodological consistency while accurately reflecting the nuances of the OFC system.

Table 2.22

SHDB processes description for the OFC scenario

Process stage	SHDB process	Description from SHDB
<i>Boat transport</i>	Water transport (wtp)/EST U	Water transport in Estonia
<i>First seaweed inoculum collection</i>	Fishing (fsh)/EST U	Fishing in Estonia
<i>Seaweed collection</i>		
<i>Lines production</i>	Chemical, rubber, plastic products (crp)/EST U	Chemical, rubber, and plastic products in Estonia
<i>Lines deployments</i>	Fishing (fsh)/EST U	Fishing in Estonia
<i>Lines maintenance</i>	Fishing (fsh)/EST U	Fishing in Estonia
<i>Boat unloading</i>	Manufactures nec (omf)/EST U	Other manufacturing Estonia
<i>Boat maintenance</i>	Trade (trd)/EST U	Retail, wholesale, and repair services in Estonia

As a final remark, it is crucial to highlight that the SHDB requires process inputs to be expressed in monetary terms (i.e., USD 2011). Given the lack of precise data on the total monetary value of the systems under study, a reverse modeling approach was adopted. Specifically, the model utilizes 1 USD generated during an 8-hour workday as the input, aligning with the time frame defined by the FU for both the WH and OC systems. To ensure consistency and accuracy in the assessment, the allocation factors presented in Tables 2.23

and 2.24 were subsequently applied to distribute the economic inputs proportionally across the different system components.

- LCI for the S-LCA

Table 2.23 presents the LCI for the WH scenario, detailing the operations involved, the corresponding allocation factors, and the assumptions applied in their calculations. This comprehensive inventory provides a structured breakdown of each process step, ensuring transparency in data attribution and methodological consistency. The allocation factors were derived based on operational characteristics, resource consumption, and system constraints, facilitating an accurate representation of the environmental and economic contributions of each activity within the WH framework.

Table 2.23

LCI for the WH scenario

Stage	Description	Assumption/calculation	Daily time to perform the operation	Allocation to FU
<i>Seaweed harvesting</i>	Harvesting by throwling. Boat capacity up to 12 tons	Assumed to be the remaining time of the 8 hours, subtracting the time required by the other operations	4.50 hours	56 %
<i>Transport to harbor</i>	40 minutes of sailing distance	Considered 2 times, for go and back	1.33 hours	17 %
<i>Boat unloading</i>	Electric unloader	-	1.50 hours	19 %
<i>Boat maintenance</i>	1 time per year (1 month)	Normalization of the 1 month period to the 8 hours of FU	0.67 hours	8 %

Table 2.24 presents the data for the OFC scenario, initially mapped over the 3-month cultivation period necessary to achieve a yield of 10 tons of *F. lumbricalis*. These values were then normalized to align with the defined FU, ensuring consistency in comparison with the WH scenario. The normalization process accounts for variations in resource consumption, labor inputs, and operational factors, facilitating a standardized assessment of the OFC system's social and economic implications.

Table 2.24

LCI for the OFC scenario

Stage	Description	Assumption/calculation	Time to perform the operation (3 months)	Normalization on the FU	Allocation to FU
<i>Boat transport</i>	40 minutes of sailing distance	It includes boat transport for the first seaweed inoculum collection, for the lines' deployment, lines maintenance, and for the final seaweed collection. Considered 2 times, for go and back.	1.33 hours (for 1 single trip, go and back) 6.00 hours (for all the trips)	0.24 hours	2.96 %
<i>First seaweed</i>	603 kg are needed to	It has been calculated knowing the rate and the	0.50 hours	0.020 hours	0.25 %

Stage	Description	Assumption/calculation	Time to perform the operation (3 months)	Normalization on the FU	Allocation to FU
<i>inoculum collection</i>	obtain 10 000 kg of fresh <i>F. lumbricalis</i>	time of growth, and the final target amount			
<i>Lines production</i>	Nylon mesh bags	The amount of nylon required was calculated by normalizing data from the literature [36], resulting in a total of 33 kg for this case study. The production rate of nylon has been assumed 250 kg per 8-hour workday [187], and this value was normalized to the 33 kg	1.06 hours	0.042 hours	0.52 %
<i>Lines deployments</i>	-	Assumed half of the time needed for the seaweed collection	2.25 hours	0.089 hours	1.11 %
<i>Lines maintenance</i>	1 time per month	3 times in total. Assumed is the same time needed for the line deployment	6.75 hours	0.27 hours	3.33 %
<i>Seaweed collection</i>	-	Since the amount is the same, it is assumed to be the same as the wild harvesting	4.50 hours	0.18 hours	2.22 %
<i>Boat unloading</i>	Electric unloader	Since the amount is the same, it is assumed to be the same as the wild harvesting	1.50 hours	0.059 hours	0.74 %
<i>Boat maintenance</i>	1 time per year (1 month)	Normalization of the 1-month period to the 3-month period needed for the seaweed growth	180 hours	7.11 hours	88.86 %

In both scenarios, the LCI is streamlined by basing calculations on the total work hours performed. This approach allows for a direct comparison between the two systems, emphasizing the potential social risks faced by an individual worker within each operational framework. By normalizing data to work hours rather than production output alone, this method provides a more accurate assessment of labor conditions, workforce exposure to risks, and overall social implications across the WH and OFC systems.

- LCIA for the S-LCA

The analysis utilizes as LCIA the “*Social Hotspot 2022 Category Method w Norm*”, which is available within the SHDB on SimaPro software v. 9.5. A schematic representation of this methodology is provided in Fig. 2.19.

The Social Hotspot 2022 Subcategory Method w Norm / Global per Capita Annual includes 30 social risk subcategories and a condensed set of five broader social damage risk categories [188]. The characterization process for each subcategory involves multiplying the LCI inventory results by a factor that represents the relative probability of adverse working conditions or community risks for a given indicator. The outcome of this process

is measured in medium risk-hour equivalents (mrheq.), providing a quantifiable metric to assess and compare social risks across different life cycle stages.

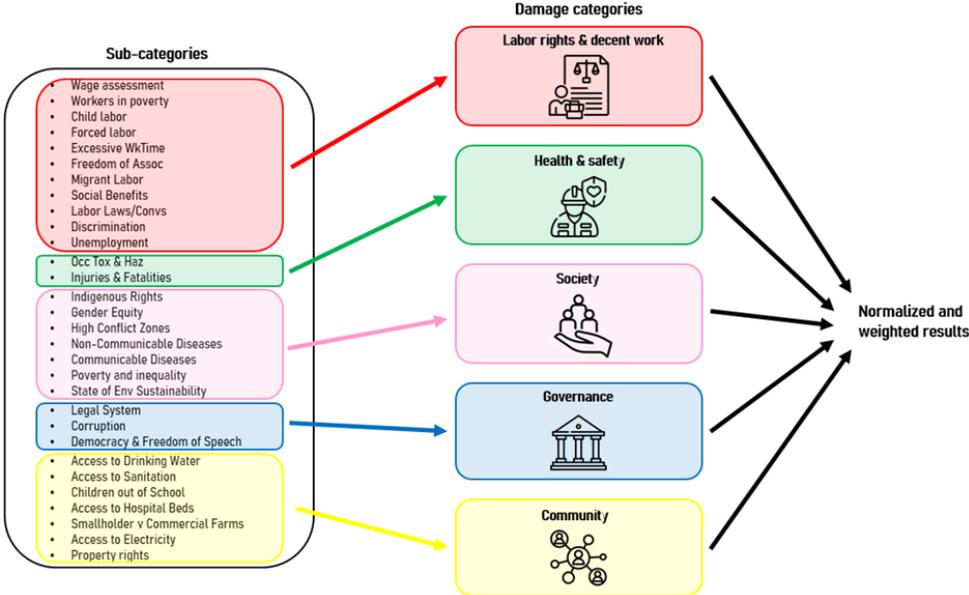


Fig. 2.19. Schematic visualization of the Social Hotspots 2022 Subcategory Method with normalization (Norm.).

These probability levels are defined in relation to the likelihood of an adverse condition occurring at a medium risk level. A low-risk level corresponds to approximately one-tenth of the likelihood compared to medium risk, assigning it a characterization factor of 1 medium risk-hour equivalent. A very high-risk level represents a tenfold increase in likelihood relative to medium risk, resulting in a characterization factor of 10 medium-risk-hour equivalents per very high-risk hour. Meanwhile, a high-risk level falls between these extremes, corresponding to about half the likelihood of very high risk or five times that of medium risk, leading to a characterization factor of 5 medium risk-hour equivalents per high risk-hour [188].

Following the risk classification, the sub-category characterization results are aggregated using a damage assessment approach at the category level. This methodology, referred to as the "Category method", generates weighted sums of sub-category results for each category. The weighting process ensures that category-level results are not skewed by the number of sub-categories within each category. Consequently, categories with fewer sub-categories will yield scores comparable to those with more sub-categories, provided the risk levels remain the same.

One version of the Category method incorporates normalization data at the category level and allows for optional user-defined weighting of category results. Finally, a weighting procedure is applied, with results expressed in annual global per capita units (S-Pt). This unit represents the total per capita annual medium risk-hour equivalents for each

sub-category, based on the total global economic output over one year. This approach facilitates meaningful comparisons of social risks across different product systems and industries.

- Uncertainty analysis

The social study analyzed two scenarios, WH and OFC, yet significant uncertainties exist within these systems. As this is the first application of S-LCA to macroalgae harvesting and production activities, the modeled systems remain hypothetical, introducing a high degree of uncertainty in defining parameters. This is particularly relevant to the allocation factors, which estimate the time required for different activities detailed in Fig. 2.3 and 2.18.

To account for these uncertainties, a Monte Carlo simulation was conducted using SimaPro software. The simulation applied a normal distribution to adjust the initial input allocations within a predefined range of values, running 5,000 simulations in total. The variation range was set according to the uncertainty level associated with each parameter and its relative importance in the allocation process. Further details on the allocation factors and their corresponding uncertainties are provided in Table 2.25.

Table 2.25

Definition of parameter variation for the Monte Carlo simulation

Process stage	Range of values
WH scenario	
<i>Seaweed harvesting</i>	±30 %
<i>Transport to harbor</i>	±10%
<i>Boat unloading</i>	±20 %
<i>Boat maintenance</i>	±20 %
OFC scenario	
<i>Boat transport</i>	±10 %
<i>First seaweed inoculum collection & seaweed collection</i>	±30 %
<i>Lines production</i>	±10 %
<i>Lines deployments</i>	±30 %
<i>Lines maintenance</i>	±30 %
<i>Boat unloading</i>	±20 %
<i>Boat maintenance</i>	±20 %

The range of values used in the Monte Carlo simulation was determined by evaluating the processes with the highest levels of uncertainty in the time required for specific operations. For the WH scenario, seaweed harvesting was identified as the most uncertain stage. In the OFC scenario, key uncertainties were associated with the first seaweed inoculum collection, seaweed collection, line deployment, and line maintenance. These operations, conducted in open sea conditions, are highly dependent on weather variations, which can significantly affect the time needed to complete them. Consequently, a larger uncertainty range (±30 %) was assigned to these stages to reflect their variability.

An intermediate uncertainty range (±20 %) was applied to boat maintenance and unloading. While these operations are primarily linked to general boat activities, variations can arise depending on the frequency and intensity of boat usage. This intermediate range provides a conservative yet reasonable estimate of potential fluctuations.

For more stable activities, such as transport to the harbor and line production, a lower uncertainty range ($\pm 10\%$) was assumed. These processes are generally fixed and less susceptible to external factors, making them more predictable.

The Monte Carlo simulation results play a crucial role in refining the analysis by establishing a broader range of possible outcomes. This approach enhances the reliability of the findings by accounting for potential variations, reducing overall uncertainty. It also provides a clearer understanding of the system's performance under different conditions, helping to mitigate the uncertainties linked to initial system data modeling.

Description of the RSA framework application

At the small-enterprise level, no existing databases comprehensively capture the potential social risks within the macroalgae sector. To address this gap, a direct assessment was conducted using custom-designed questionnaires tailored specifically for this research. These questionnaires were distributed to a panel of experts specializing in macroalgae production, maritime activities, and sustainability. The overall framework followed the guidelines provided in the UNEP Social LCA Handbook [180] and the ISO 14075 [181] to ensure methodological rigor.

Two questionnaires were developed. The first questionnaire, which is fully detailed in Annex 1, provided a broad assessment by listing a wide range of stakeholder groups potentially affected by social impacts and their associated social indicators, fully developed from the UNEP Social LCA handbook. This questionnaire served as an initial screening tool to identify the most relevant stakeholders and social indicators applicable to the system under analysis. It was developed and subsequently shared with other partners in the TACO algae project for validation and screening.

Based on the responses from the first questionnaire, a total of nine subcategories and four stakeholder groups were identified as the most significant indicators of social impact for this case study (Table 2.26).

Table 2.26

Selected indicators to define the potential social impacts for each category of stakeholder

	Categories of stakeholders			
	<i>Workers</i>	<i>Local community</i>	<i>Consumers</i>	<i>Other value chain actors</i>
Indicators of social impact	<ul style="list-style-type: none"> ○ Health and safety ○ Working hours ○ Social benefits/social security 	<ul style="list-style-type: none"> ○ Safe and healthy living conditions ○ Local economic development 	<ul style="list-style-type: none"> ○ Health and Safety ○ Transparency 	<ul style="list-style-type: none"> ○ Fair competition ○ Promoting social responsibility ○ Wealth distribution

The second questionnaire, developed based on the findings of the first, is included in Annex 2. It specifically targets four key stakeholder groups identified during the initial survey (Table 2.26), comprising 14 questions related to the selected 10 social indicators. To

structure the assessment, the questions were divided into two main stages that define the macroalgae sector: collection or cultivation and processing.

This second questionnaire was distributed to a panel of eight experts, including project partners HYNDLA and VETIK OÜ. Although over 20 experts were invited to participate, only eight responded or agreed to take part in the survey.

Following the collection of questionnaire responses, potential social risks were assessed qualitatively and classified into four categories: high, medium, low, or zero risk. To facilitate a structured and quantitative interpretation of these qualitative results, a reference scale (RS) was established. This scale converts qualitative assessments into numerical values, ranging from -2 (high risk) to +2 (no risk). Additionally, a color-coded system was integrated to enhance readability and simplify interpretation. Tables 2.27-2.36 present the specific RSs defined for the selected indicators.

Health and safety for a worker refers to the protection of workers' physical, mental, and social well-being in the workplace. According to the International Labour Organization and the World Health Organization, it aims to prevent work-related health issues, mitigate risks arising from occupational hazards, and ensure that work environments are adapted to workers' physiological and psychological capacities. A safe workplace, as defined by the Occupational Safety and Health Administration, is one that is free from serious recognized hazards and complies with safety standards. This indicator assesses both the frequency of workplace incidents, including injuries, illnesses, and fatalities, and the effectiveness of prevention and management measures in place. Workplace health is not only about the absence of disease or injury, but also includes factors such as safety policies, hygiene standards, and mental well-being [189].

Table 2.27

RS for the *health and safety* indicator for workers stakeholder

Scale level	Stakeholder category: WORKERS Impact subcategory: HEALTH AND SAFETY
+2	There are no risks for workers of the processes under analysis to be exposed to accidents/damages for health and safety as well as to carry out strenuous activities.
+1	There is a low risk for workers of the processes under analysis to be exposed to accidents/damages to health and safety as well as to carry out strenuous activities.
0	There is not a shared position of the experts, or there is not enough data available.
-1	There is a medium risk for workers of the processes under analysis to be exposed to accidents/damages to health and safety as well as to carry out strenuous activities.
-2	There is a very high risk for workers of the processes under analysis to be exposed to accidents/damages to health and safety as well as to carry out strenuous activities.

Working hours evaluates compliance with legal and industry standards regarding working hours, ensuring that workers do not regularly exceed 48 hours per week and receive at least one rest day per seven-day period. Overtime is voluntary, limited to 12 hours per week, not systematically required, and compensated at a premium rate. The organization of

working hours considers workers' needs, with stricter regulations for night shifts. The assessment focuses on verifying adherence to International Labour Organization standards, ensuring that actual working hours align with legal requirements. It also examines whether workers receive appropriate compensation for overtime, either as financial remuneration or additional time off. This indicator applies across various work arrangements, from part-time to full-time employment, and different workplaces, including home-based, field, and manufacturing jobs [189].

Table 2.28

RS for the *working hours* indicator for workers stakeholder

Scale level	Stakeholder category: WORKERS Impact subcategory: WORKING HOURS
+2	There is no risk . 0 % of workers exceed 40-48 hours per week.
0	There is not a shared position of the experts, or there is not enough data available.
-1	There is a medium risk . From 0 % to <50 % of workers exceed 40-48 hours per week.
-2	There is a very high risk . More than 50 % of workers exceed 40-48 hours per week.

Social benefits/Social security considers the non-monetary compensation provided to workers, which is typically available to full-time employees but may not extend to part-time, contract, or home-based workers. Social security, a subset of social protection, includes government-backed schemes such as social insurance and assistance, covering benefits like retirement, disability, dependents, and survivor benefits. Additional social benefits may include medical, dental, and paramedical insurance, wage protection, paid parental and sick leave, education and training programs, meal vouchers, and access to wellness facilities like gyms or childcare services. The availability of these benefits varies by country, depending on national policies. Some nations provide universal healthcare, while others rely on employer-provided insurance. The assessment aims to determine whether an organization offers social benefits and social security coverage to its workers and to what extent these provisions contribute to their well-being and job security [189].

Table 2.29

RS for the *social benefits/Social security* indicator for workers stakeholder

Scale level	Stakeholder category: WORKERS Impact subcategory: SOCIAL BENEFITS/SOCIAL SECURITY
+2	There are no risks . The workers have more than 6 social benefits.
+1	There is a low risk . The workers have between 2 and 6 social benefits.
0	There is not a shared position of the experts, or there is not enough data available.
-1	There is a medium risk . The workers have only 1 or 2 benefits.
-2	There is a very high risk . The workers do not have any social benefits.

Safe and healthy living conditions for the local community pose the risk evaluation through equipment accidents, structural failures, or land use changes that may lead to disasters such as landslides. Additionally, changes in land use can contribute to the spread of diseases, for example, poor water drainage fostering malaria outbreaks. The influx of workers may also facilitate the spread of communicable diseases. Hazardous material use and pollution emissions further exacerbate health risks, necessitating robust environmental risk management to prevent and mitigate adverse effects. Beyond risk management, organizations can positively influence local health by providing shared access to healthcare facilities and ensuring transparency about potential health and safety risks. In cases where operations cause harm, companies are expected to engage in remediation or compensation efforts. This indicator assesses an organization's commitment to maintaining safe operational conditions and minimizing negative public health impacts [189].

Table 2.30

RS for the *safe and healthy living conditions* indicator for the local community stakeholder

Scale level	Stakeholder category: LOCAL COMMUNITY Impact subcategory: SAFE AND HEALTHY LIVING CONDITIONS
+2	The presence of the process under analysis does not increase the perception of the risk of affecting the local living conditions.
+1	The presence of the process under analysis slightly increases the perception of the risk of affecting local living conditions.
0	There is not a shared position of the experts, or there is not enough data available.
-1	The presence of the process under analysis increases the perception of the risk of affecting local living conditions.
-2	The presence of the process under analysis highly increases the perception of the risk of affecting local living conditions.

Local economic development evaluates the extent to which organizations contribute to local employment and community development through hiring practices and supplier relationships. Prioritizing local hiring provides income opportunities and skill development for community members, fostering economic growth. Establishing partnerships with locally-based suppliers further amplifies these benefits by encouraging business development within the region.

Beyond direct employment, organizations can enhance local development by offering technical and transferable skills training and equipping workers with long-term career opportunities. A significant impact can also be seen when local employees are appointed to senior management positions, as this fosters trust, open communication, and stronger community ties. This indicator assesses how effectively an organization supports local employment and economic sustainability [189].

Table 2.31

RS for the *local economic development* indicator for the local community stakeholder

Scale level	Stakeholder category: LOCAL COMMUNITY Impact subcategory: LOCAL ECONOMIC DEVELOPMENT
+2	The presence of the processes under analysis could highly contribute to the development of the local economy by workforce hired locally, as well as prioritizing buying goods and services from local suppliers, and by the development of “satellite activities”.
+1	The presence of the processes under analysis could positively contribute to the development of the local economy.
0	There is not a shared position of the experts, or there is not enough data available.
-1	The presence of the processes under analysis has a low contribution to economic development.
-2	The presence of the processes under analysis has a very low contribution to economic development

The *health and safety* for the consumer evaluates an organization's commitment to ensuring the health and safety of consumers by minimizing risks associated with its products and services. It assesses whether systematic measures are in place to prevent harm, ensure product reliability, and provide early warnings in case of hazardous products, recalls, or bans [190]. Organizations are expected to adhere to safety standards and consumer protection regulations, ensuring that their products and services meet performance expectations without posing risks to users. The assessment aims to determine the extent of these efforts throughout the product or service life cycle [189].

Table 2.32

RS for the *health and safety* indicator for the consumers stakeholder

Scale level	Stakeholder category: CONSUMERS Impact subcategory: HEALTH AND SAFETY
+2	The presence of the processes under analysis could highly contribute to the development of zero hunger, good health & well-being, and responsible consumption & production practices
+1	The presence of the processes under analysis could positively contribute to the development of zero hunger, good health & well-being, and responsible consumption & production practices
0	There is not a shared position of the experts, or there is not enough data available.
-1	The presence of the processes under analysis has a low contribution to the development of zero hunger, good health & well-being, and responsible consumption & production practices
-2	The presence of the processes under analysis has a very low contribution to the development of zero hunger, good health & well-being, and responsible consumption & production practices

Transparency accounts for an organization's commitment to transparency in its communication with consumers, ensuring that information about products and social

responsibility is clear, accurate, and not misleading. It assesses whether the organization provides accessible and verifiable details through certifications, labels, and indices that reflect social and environmental performance. By fostering transparency, organizations enable consumers to make informed choices based on reliable and ethical disclosures. The assessment aims to determine the extent to which organizations openly share relevant product and corporate responsibility information [189].

Table 2.33

RS for the *transparency* indicator for the consumers stakeholder

Scale level	Stakeholder category: CONSUMERS Impact subcategory: TRANSPARENCY
+2	The selected “actor” highly contributes to promoting transparency using practices like certification standards, labels, and special indices. Moreover, it provides sustainability reports based on standardized methodology such as life cycle impact assessment.
+1	The selected “actor” positively contributes to promoting transparency using practices like certification standards, labels, and special indices.
0	There is not a shared position of the experts, or there is not enough data available.
-1	The selected “actor” has a low contribution to promoting transparency using practices like certification standards, labels, and special indices. Moreover, it provides sustainability reports based on standardized methodology such as life cycle impact assessment.
-2	The selected “actor” has a very low contribution to promoting transparency using practices like certification standards, labels, and special indices. Moreover, it provides sustainability reports based on standardized methodology such as life cycle impact assessment.

Fair competition for the value chain actors analyzes whether an organization engages in fair competition and complies with laws preventing anti-competitive behavior, antitrust violations, and monopoly practices. It assesses actions such as price fixing, bid rigging, predatory pricing, and collusion that restrict market competition. Additionally, it examines whether the organization engages in practices that create unfair barriers to entry, abuse market dominance, or form cartels. The assessment aims to determine if the organization’s competitive conduct aligns with legal and ethical standards, ensuring a fair and open marketplace [189].

Table 2.34

RS for the *fair competition* indicator for the value chain actors stakeholder

Scale level	Stakeholder category: VALUE CHAIN ACTORS Impact subcategory: FAIR COMPETITION
+2	The selected “actor” highly contributes to promoting fair competition. Conducting competitive activities in a fair way and in compliance with preventing anti-competitive behavior, anti-trust, or monopoly practices.
+1	The selected “actor” positively contributes to promoting fair competition. Conducting competitive activities in a fair way and in compliance with preventing anti-competitive behavior, anti-trust, or monopoly practices.

Scale level	Stakeholder category: VALUE CHAIN ACTORS Impact subcategory: FAIR COMPETITION
0	There is not a shared position of the experts, or there is not enough data available.
-1	The selected “actor” has a low contribution to promoting fair competition. Conducting competitive activities in a fair way and in compliance with preventing anti-competitive behavior, anti-trust, or monopoly practices.
-2	The selected “actor” has a very low contribution to promoting fair competition. Conducting competitive activities in a fair way and in compliance with preventing anti-competitive behavior, anti-trust, or monopoly practices.

Promoting social responsibility accounts for an organization's commitment to social responsibility (SR) in its operations and supply chain. It assesses whether the organization actively considers stakeholders' interests, including customers, employees, shareholders, and communities, and integrates SR into its core business practices. The assessment also examines whether the organization manages suppliers in a socially responsible manner, through monitoring, auditing, and corrective actions when necessary. Key factors include the presence of supplier codes of conduct, contractual agreements addressing social and environmental responsibilities, and efforts to encourage suppliers to adopt ethical standards. Additionally, promoting SR certifications, product labels, and participation in related foundations or initiatives serves as a measure of an organization's commitment to ethical business practices [189].

Table 2.35

RS for the *promoting social responsibility* indicator for the value chain actors stakeholder

Scale level	Stakeholder category: VALUE CHAIN ACTORS Impact subcategory: PROMOTING SOCIAL RESPONSIBILITY
+2	The selected “actor” highly contributes to promoting social responsibility among its suppliers and through its own actions. These measures include monitoring (to consider human rights records when selecting suppliers), auditing, and training efforts.
+1	The selected “actor” positively contributes to promoting social responsibility among its suppliers and through its own actions. These measures include monitoring (to consider human rights records when selecting suppliers), auditing, and training efforts.
0	There is not a shared position of the experts, or there is not enough data available.
-1	The selected “actor” has a low contribution to promoting social responsibility among its suppliers and through its own actions. These measures include monitoring (to consider human rights records when selecting suppliers), auditing, and training efforts.
-2	The selected “actor” has a very low contribution to promoting social responsibility among its suppliers and through its own actions. These measures include monitoring (to consider human rights records when selecting suppliers), auditing, and training efforts.

The *wealth distribution* assesses the fairness of value distribution across all participants in the value chain. Equitable wealth distribution ensures that each actor receives a fair share of the total value created, with pricing structures that cover production costs and provide acceptable profit margins. However, market pricing does not always guarantee fair returns at all levels, particularly in the production stage, where intermediate manufacturing

processes often contribute less to total value compared to upstream (e.g., R&D, design) and downstream (e.g., marketing, sales) activities. The presence of contractual agreements at the sector or cluster level can help protect weaker actors in the value chain by ensuring fair trading conditions. The assessment aims to determine whether the value generated by a product or service is distributed fairly among all stakeholders, promoting economic equity and sustainable business practices [189].

Table 2.36

RS for the *wealth distribution* indicator for the value chain actors stakeholder

Scale level	Stakeholder category: VALUE CHAIN ACTORS Impact subcategory: WEALTH DISTRIBUTION
+2	The selected “actor” highly contributes to the wealth distribution among the supply chain using contract instruments, the creation of the interbranch organization, and the definition of a fair price for the products.
+1	The selected “actor” positively contributes to the wealth distribution among the supply chain using contract instruments, the creation of the interbranch organization, and the definition of a fair price for the products.
0	There is not a shared position of the experts, or there is not enough data available.
-1	The selected “actor” has a low contribution to the wealth distribution among the supply chain using contract instruments, the creation of the interbranch organization, and the definition of a fair price for the products.
-2	The selected “actor” has a very low contribution to the wealth distribution among the supply chain using contract instruments, the creation of the interbranch organization, and the definition of a fair price for the products.

2.5. Life cycle sustainability assessment

The LCSA is a holistic framework designed to evaluate the sustainability performance of products, processes, or systems by integrating three key dimensions: LCA or E-LCA, LCC, and S-LCA aspects. This approach enables decision-makers to assess trade-offs between sustainability pillars, facilitating more informed and balanced choices throughout the entire life cycle of a product. To effectively apply LCSA, a Multi-Criteria Decision Analysis (MCDA) is employed, helping to navigate the complexity of decision-making by systematically comparing alternatives based on defined criteria. MCA draws from disciplines such as engineering, economics, and social sciences, allowing for structured evaluations that combine both quantitative and qualitative aspects. Various MCA methods exist, including parameters and results granularity [191], function definitions [192], thresholds [193] and pairwise comparisons [194], [195], with TOPSIS (Technique for Order of Preference by Similarity to Ideal Solution) [196] being particularly relevant in sustainability studies due to its ability to identify the most balanced option based on proximity to an ideal solution.

LCSA follows a life cycle perspective, meaning it considers all phases of a product's existence, from raw material extraction and production to use, disposal, and end-of-life management. This comprehensive view helps to prevent burden-shifting, ensuring that

improvements in one area do not inadvertently lead to negative impacts elsewhere. Furthermore, LCSA adheres to the principle of completeness, requiring that all relevant sustainability aspects be considered to provide a well-rounded assessment. The LCSA framework, first conceptualized by Walter Klöpffer [197], underscores the importance of interpreting environmental, economic, and social indicators together rather than aggregating them into a single score. This approach allows for a balanced analysis where trade-offs and synergies among the three pillars can be clearly identified. See Fig. 20.

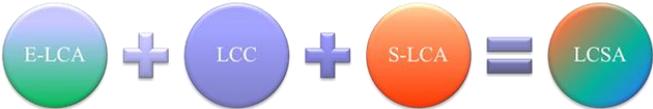


Fig. 2.20. The Life Cycle Sustainability Assessment framework [198].

The adoption of LCSA provides several advantages, making it a valuable tool for businesses, policymakers, and researchers. It enables structured data management, organizes complex sustainability information systematically, and clarifies trade-offs between environmental, economic, and social impacts. It helps organizations assess and improve the sustainability of their products and supply chains while raising awareness of sustainability impacts among stakeholders. LCSA aids in detecting weaknesses in production systems, optimizing resource allocation, and guiding technology and product selection toward the most sustainable alternatives. It supports consumer decision-making by identifying environmentally and socially responsible products and stimulates innovation by encouraging the development of sustainable production methods and technologies. Additionally, LCSA plays a crucial role in certification and labeling initiatives, contributing to eco-labeling for sustainable product marketing and helping shape policies and strategies for more responsible production and consumption patterns.

Given the increasing global emphasis on sustainability, LCSA is becoming an essential tool for guiding product innovation, policy development, and responsible consumption. By integrating environmental, economic, and social indicators, LCSA ensures a comprehensive sustainability assessment, supporting organizations in making evidence-based, future-oriented decisions.

The TOPSIS methodology

The TOPSIS method is recognized for its efficiency in multi-criteria decision-making, requiring minimal user input while delivering clear and easily interpretable results. Developed by Hwang and Yoon [196], TOPSIS ranks alternatives based on their proximity

to an ideal solution, making it a valuable tool for evaluating multiple options against selected criteria [199].

By determining the closest distance to the ideal solution and the furthest distance from the anti-ideal solution, TOPSIS systematically identifies the best-performing alternative among all proposed options. Like other MCA methods, it incorporates a subjective component through the assignment of weights to each criterion, influencing the final ranking [191].

The significance of these distances is illustrated in Fig. 2.21, where two alternatives, A and B, are evaluated under equal weight distribution for the criteria. In this scenario, Alternative A is positioned closer to the ideal solution and further from the anti-ideal solution compared to Alternative B, leading TOPSIS to determine Alternative A as the superior choice.

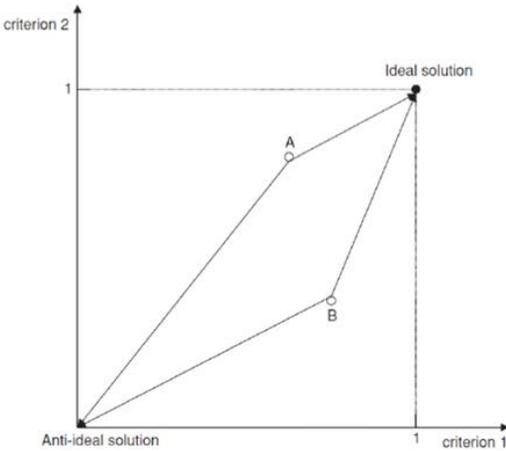


Fig. 2.21. TOPSIS method [199].

TOPSIS involves six distinct computational steps:

1. Collecting the performance data of alternatives across different criteria in quantitative terms.
2. Normalizing the quantitative performance values for each alternative within the decision matrix.
3. Weighting the normalized performance values for each alternative, resulting in a weighted normalized decision matrix.
4. Comparing the weighted scores of each alternative to both an ideal and anti-ideal solution.
5. Calculating the distances of each alternative from the ideal and anti-ideal solutions.
6. Determining the relative closeness of each alternative to the ideal and anti-ideal solutions, ultimately identifying the best alternative.

These steps are explained in further detail below.

From a mathematical perspective, the core of the TOPSIS analysis is a data matrix, where the evaluation criteria are represented as $x_1, x_2, \dots, x_j, \dots, x_m$. The performance of n

$$A^- = (v_1^-, \dots, v_j^-, \dots, v_m^-) \quad (2.10)$$

Where $v_j^+ = \max_j(v_{ij})$ if the criterion is to be maximized and $v_j^- = \min_j(v_{ij})$ if the criterion is to be minimized.

5. Step 5

Calculate the Euclidean distance far from the ideal solution:

$$d_i^+ = \sqrt{\sum_{j=1}^m (v_j^+ - v_{ij})^2} \quad (2.11)$$

And from the anti-ideal solution:

$$d_i^- = \sqrt{\sum_{j=1}^m (v_j^- - v_{ij})^2} \quad (2.12)$$

6. Step 6

Calculate the relative closeness coefficient of each action:

$$C_i = \frac{d_i^-}{d_i^+ + d_i^-} \quad (2.13)$$

The closeness coefficient is always between 0 and 1. Preferred alternatives are scored with 1; therefore, for alternatives closer to the ideal than the anti-ideal, C_i will be closer to 1. For alternatives closer to the ideal than the anti-ideal, C will be closer to 0.

The data integration for the LCSA

The developed methodology is structured to conduct an MCDA to assess the LCSA of the three distinct biorefinery configurations developed in Chapter 2.1 (i.e., CB, SPE, and TLE). To ensure a comprehensive evaluation, the assessment criteria were categorized under the three pillars of sustainability, environmental, economic, and social, resulting in a total of 12 sub-criteria.

Four environmental indicators were chosen, derived from results in Chapter 3.1 (LCA results), representing the indicators with the most significant environmental impact on the systems. These indicators are:

- Global warming potential expressed in kg CO₂ eq.
- Fine particulate matter formation expressed in kg PM_{2.5}
- Human carcinogenic and non-carcinogenic toxicity expressed in kg 1.4-DCB
- Water consumption expressed in m³

From the economic perspective, four indicators from the results in Chapter 3.2 (LCC results), which overall describe the economic assessment of the system, have been taken:

- Return of Investment expressed in %
- Net Present Value expressed in EUR
- Internal Rate of Return expressed in %

- Profitability Index

For the SPE complete scenario, the single SPEp, SPEpr, and SPEc have been added in order to create a full, complete, comparable system.

Finally, for the social aspect, four social indicators have been selected. They have been chosen because they are those that are more able to underline the differences among the different biorefinery designs. They have been calculated in Chapter 3.3 (S-LCA results), and they are:

- Health & safety of the worker (HS)
- Working hours of a worker (WHS)
- Local economy development (LED)
- Wealth distribution among the supply chain (WD)

For the S-LCA results, certain adjustments were necessary to ensure comparability across the three biorefinery designs.

In the TLE configuration, the HS indicator was assumed to be equivalent to that of the CB design. In contrast, for the SPE configuration, the HS indicator was determined by summing the risks associated with the production of each individual product line, pigments, proteins, and carrageenan, following the approach outlined in Equations 2.14 and 2.15. Given that this setup is anticipated to present a higher level of HS risks compared to the CB design, the corresponding values were assigned negative weights to account for the increased risk exposure.

$$HS_{SPE} = HS_{SPE,pigments} - HS_{SPE,proteins} - HS_{SPE,carrageenan} \quad (2.14)$$

$$HS_{SPE,pigments} = HS_{SPE,proteins} = HS_{SPE,carrageenan} = HS_{CB} + \frac{HS_{CB}}{3} \quad (2.15)$$

In TLE, the WHS was assumed to be identical to that in CB. However, in SPE, WHS was determined by summing the risks associated with the production of each individual product, as outlined in Equations 2.16 and 2.17. Given that this configuration is expected to present a higher WHS risk compared to CB, the resulting value was assigned a negative weight, aligning with the approach applied to the HS indicator.

$$WHS_{SPE} = WHS_{SPE,pigments} - WHS_{SPE,proteins} - WHS_{SPE,carrageenan} \quad (2.16)$$

$$WHS_{SPE,pigments} = WHS_{SPE,proteins} = WHS_{SPE,carrageenan} = WHS_{CB} \quad (2.17)$$

LED and WD were calculated based on the total output of valuable products in each scenario, under the assumption that higher yields contribute to greater wealth distribution and reduced social risk. Consequently, in SPE and TLE, LED and WD were proportionally adjusted by comparing the percentage difference in product yields relative to CB. In SPE, for every 1 ton of DW biomass of *F. lumbricalis* processed per extraction line, the resulting yield was 20 kg of pigments, 100 kg of proteins, and 200 kg of carrageenan, aligning with the production levels of CB. As a result, LED and WD remained unchanged. However, in TLE, processing 1 ton DW of *F. lumbricalis* led to significantly lower yields, producing only 6.67 kg of pigments, 33.33 kg of proteins, and 66.67 kg of carrageenan [135]. To

accurately reflect these disparities in production efficiency, LED and WD for TLE were recalculated using Equations 2.18 and 2.19, ensuring a proportional adjustment in line with the observed differences.

$$LED_{TLE} = \frac{LED_{CB} * (pigments_{TLE} + proteins_{TLE} + carrageenan_{TLE})}{(pigments_{CB} + proteins_{CB} + carrageenan_{CB})} \quad (2.18)$$

$$WD_{TLE} = WD_{CB} + \frac{WD_{CB}}{3} \quad (2.19)$$

For WD_{TLE} , the value was incorporated into WD_{CB} to more accurately reflect the heightened risk associated with wealth distribution, stemming from the lower product yield in this system configuration. This adjustment ensures that the negative implications of reduced production are properly accounted for in the overall assessment.

To establish the weight (W) of each sustainability criterion, environmental, economic, and social, a distributive normalization approach was applied, as detailed in Eq. 2.20. This structured method provides a balanced framework for integrating the three pillars of sustainability within the overall evaluation, ensuring that each aspect is proportionally represented based on its relative impact.

$$W = \beta * \frac{1}{n} \quad (2.20)$$

Where n represents the number of criteria considered in this study (i.e., three), and β is the variation ratio, which was set to 1 in this analysis. Initially, an equal distribution of weights was applied across the three sustainability criteria, assigning each a value of 0.33 (i.e., 1/3).

To enhance the robustness of the results and assess the impact of varying the relative importance of each sustainability pillar, a sensitivity analysis was performed. This analysis involved systematically modifying the weight of one criterion while proportionally redistributing the remaining weight among the other two. The adjustments were made in increments of 10 %, 50 %, 150 %, 200 %, and 300 % to evaluate how different weighting scenarios influence the final sustainability ranking of the biorefinery designs.

3. RESULTS AND DISCUSSION

3.1. Environmental results

This chapter presents and analyzes the results of the environmental sustainability assessment. It begins with the LCA findings related to the upstream stage, focusing on biomass gathering operations. Subsequently, the discussion extends to the overall environmental performance of the complete *F. lumbricalis* biorefinery system, evaluating its impact across different processing configurations.

Results of the LCA on the different techniques for the upstream stage

The results presented in this section pertain to the environmental impact assessment of the three possible macroalgae “production” methods: WH, ONC, and OFC. These methods have been previously detailed in Chapter 2.1 (Fig. 2.3) and Chapter 2.2 (Fig. 2.11 and 2.12). The LCI data corresponding to each scenario are provided in Tables 2.2, 2.3, and 2.4, forming the basis for the comparative environmental evaluation.

- Results for the WH scenario

Table 3.1 presents the environmental impact results for the WH scenario across the selected midpoint indicators. The step with the highest environmental impact is highlighted in grey, indicating the most critical phase in terms of sustainability challenges.

Table 3.1

Midpoint results for the WH scenario. From approbation publication No. 2 [134]

Impact category	Unit	Seaweed harvesting	Transport to the harbor	Unloading the boat	Final transport
CC	kg CO ₂ eq	6.79E+01	4.53E+00	5.99E-01	1.14E+01
PM	disease inc.	6.33E-06	1.20E-06	5.19E-08	8.60E-07
AP	mol H+ eq	8.36E-01	1.60E-01	5.53E-03	4.54E-02
EF	kg P eq	1.54E-02	4.75E-05	4.26E-05	8.45E-04
EM	kg N eq	2.01E-01	3.87E-02	8.16E-04	1.32E-02
ECF	CTUe	1.63E+03	3.30E+01	7.00E+00	1.39E+02
LU	Pt	1.56E+02	7.87E+00	3.81E+00	1.02E+02
WU	m ³ depriv.	2.57E+01	1.09E-02	4.43E-02	6.05E-01
RF	MJ	8.32E+02	6.22E+01	8.21E+00	1.70E+02
RM&M	kg Sb eq	2.81E-03	8.15E-07	8.08E-07	4.72E-05

Fig. 3.1 illustrates that seaweed harvesting is the primary contributor to environmental impact, particularly affecting the RM&M and CC indicators. The values expressed in EcoPoints (Pt) provide a single-score representation of the results normalized to the environmental impact of an EU citizen over one year [200].

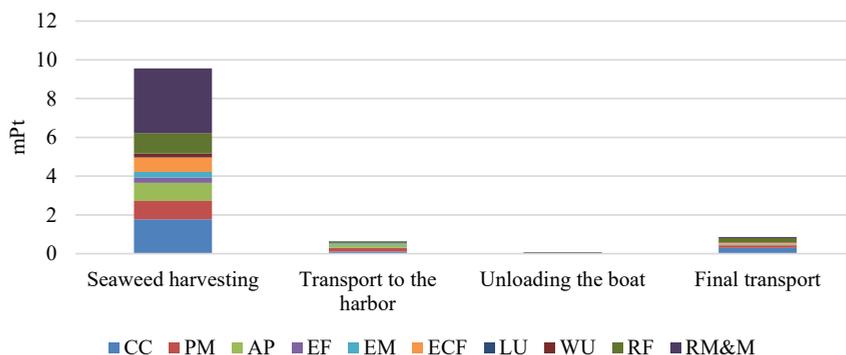


Fig. 3.1. Endpoint results for the WH. From approbation publication No. 2 [134].

Fig. 3.2 provides a detailed analysis of the seaweed harvesting procedure, identifying the antifouling agent as the most environmentally impactful material, contributing 3.18 mPt. Diesel fuel follows with an impact of 2.28 mPt, while the steel used in boat construction and maintenance accounts for 1.45 mPt. These results highlight the primary sources of environmental burden in the harvesting phase, emphasizing the need for targeted mitigation strategies, such as alternative antifouling solutions, improved fuel efficiency, or alternative vessel materials.

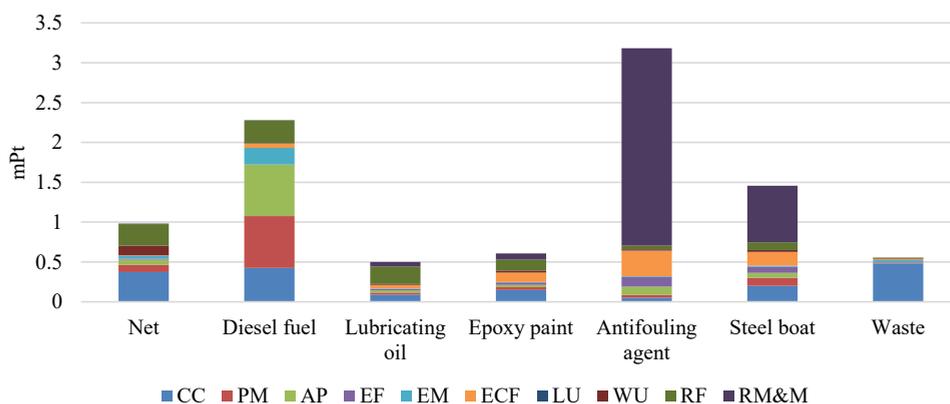


Fig. 3.2. Focus results for the seaweed harvesting procedures in the WH system. From approbation publication No. 2 [134].

- Results for the ONC scenario

Table 3.2 presents the environmental impact results for the ONC scenario across the selected midpoint indicators. Notably, the initial collection of raw algae for cultivation is excluded, as it is assumed to be performed manually, resulting in a negligible environmental impact. The table highlights the step with the highest environmental burden in grey, emphasizing the most critical process in terms of sustainability.

Table 3.2

Midpoint results for the ONC scenario. From approbation publication No. 2 [134]

Impact category	Unit	Transportation to facility	Cleaning	Cultivation
CC	kg CO ₂ eq	4.96E+00	2.70E+00	2.13E+02
PM	disease inc.	3.08E-07	2.46E-07	1.22E-05
AP	mol H ⁺ eq	2.00E-02	1.58E-02	8.11E-01
EF	kg P eq	6.93E-04	1.08E-03	6.82E-02
EM	kg N eq	4.38E-03	3.47E-03	2.00E-01
ECF	CTUe	9.47E+01	1.63E+02	7.83E+04
LU	Pt	2.99E+01	8.56E+01	4.76E+04
WU	m ³ depriv.	5.09E-01	3.06E+00	1.11E+02
RF	MJ	6.78E+01	5.09E+01	1.96E+03
RM&M	kg Sb eq	6.52E-05	5.25E-05	2.15E-03

Fig. 3.3 confirms that cultivation procedures represent the most significant environmental impact within the ONC scenario, contributing 56.206 mPt. This finding highlights the intensive resource and energy demands associated with maintaining controlled onshore macroalgae growth conditions. Understanding this impact is crucial for optimizing cultivation techniques to improve overall sustainability and reduce the environmental footprint of macroalgae-based biorefineries.

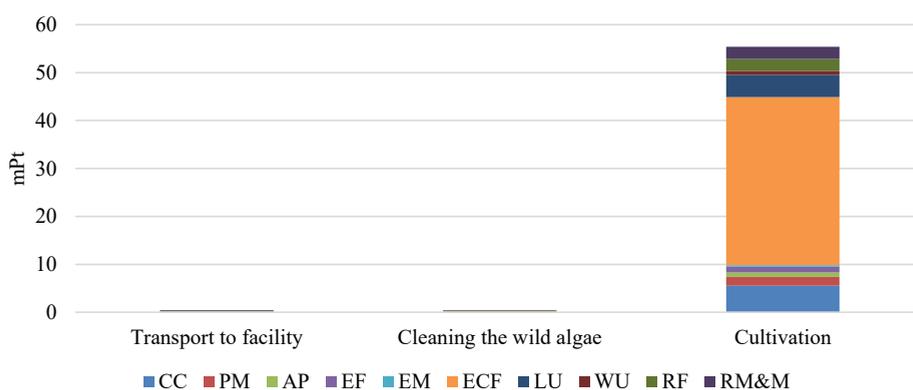


Fig. 3.3. Endpoint results for the ONC. From approbation publication No. 2 [134].

Fig. 3.4 illustrates that the primary environmental burden in the ONC scenario stems from the electricity required to generate artificial sunlight, contributing 43.49 mPt to the ECF indicator. This impact is closely linked to the infrastructure and operational processes involved in geothermal electricity production, particularly the drilling waste generated during extraction. These findings emphasize the need for energy-efficient lighting alternatives or a shift toward natural sunlight integration to mitigate the environmental footprint of onshore macroalgae cultivation.

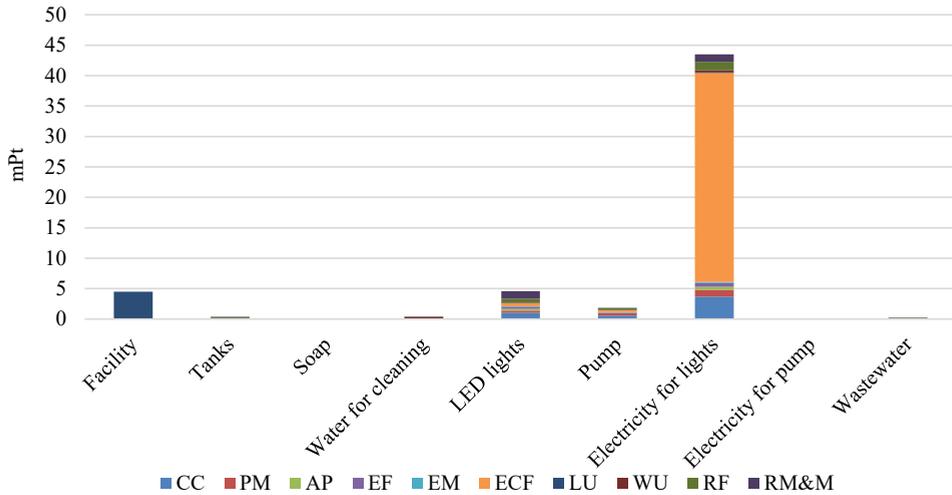


Fig.3.4. Focus results on the cultivation stage for the ONC system. From approbation publication No. 2 [134].

- Results for the OFC scenario

Table 3.3 presents the midpoint results for the OFC scenario, with the step exhibiting the highest environmental impact highlighted in grey. This allows for clearer identification of the most critical phase within the offshore cultivation process, facilitating targeted strategies for impact reduction.

Table 3.3

Midpoint results for the OFC scenario. From approbation publication No. 2 [134]

Impact category	Unit	Collection of raw algae	Spore release and seeding	Nursery phase	Deployment of lines	Harvesting	Final transport
CC	kg CO ₂ eq	7.28E-01	4.00E+01	1.89E+00	1.74E+02	3.21E+01	1.14E+01
PM	disease inc.	4.67E-08	1.80E-06	1.30E-07	7.09E-06	8.49E-07	8.60E-07
AP	mol H ⁺ eq	8.47E-03	2.14E-01	1.38E-02	1.69E+00	1.10E+00	4.54E-02
EF	kg P eq	6.39E-05	9.07E-03	2.23E-04	1.63E-02	3.11E-04	8.45E-04
EM	kg N eq	1.07E-03	3.79E-02	2.90E-03	4.51E-01	2.76E-01	1.32E-02
ECF	CTUe	3.76E+01	6.77E+02	2.73E+01	1.16E+03	1.87E+02	1.39E+02
LU	Pt	8.92E+00	1.40E+02	8.84E+00	3.62E+02	4.53E+01	1.02E+02
WU	m ³ depriv.	3.88E-02	2.77E+01	4.29E-01	1.06E+02	7.89E-02	6.05E-01
RF	MJ	7.07E+01	1.04E+03	3.19E+01	3.13E+03	3.56E+02	1.70E+02
RM&M	kg Sb eq	1.51E-06	3.38E-04	8.75E-06	5.12E-04	5.06E-06	4.72E-05

The step of the OFC that gives the most noticeable environmental impact is the deployment of the lines (Fig. 3.5). Focusing on that, Fig. 3.6 demonstrates that the thin nylon ropes are the most impactful material (4.69 mPt), followed by the buoys (3.37 mPt) and the boat use (3.16 mPt). This is also in line with the literature because nylon production is one of the most harmful in terms of emissions and embodied energy [201].

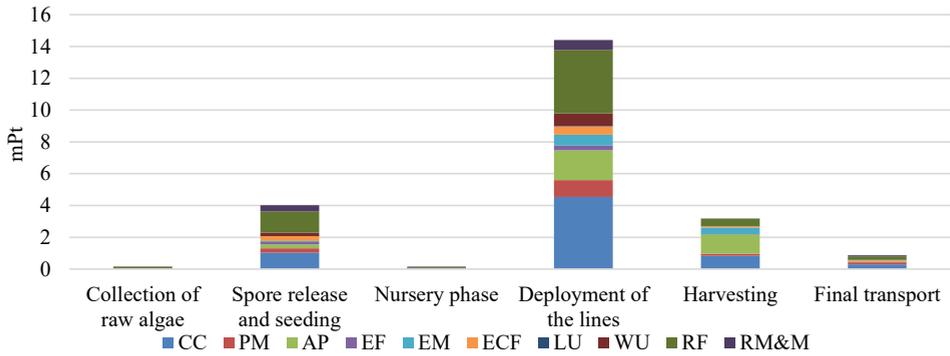


Fig. 3.5. Endpoint results for the OFC. From approbation publication No. 2 [134].

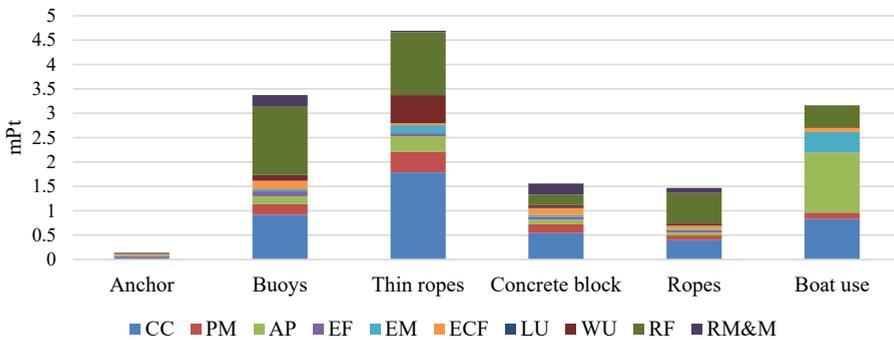


Fig. 3.6. Focus results on the seaweed harvesting stage for the OFC system. From approbation publication No. 2 [134].

- Comparison of the three scenarios

Table 3.4 presents the total results for each midpoint indicator, highlighting the highest values among the different macroalgae harvesting and cultivation techniques. The ONC scenario exhibits the highest environmental impact in CC, EF, ECF, LU, and WU. The OFC scenario records the maximum impact in PM, AP, EM, and RF. Meanwhile, the WH scenario has the highest impact on RM&M.

Table 3.4

Total midpoint results of the WH, ONC, and OFC scenarios. From approbation publication No. 2 [134]

Impact category	Unit	WH total	ONC total	OFC total
CC	kg CO ₂ eq	8.45E+01	2.20E+02	2.60E+02
PM	disease inc.	8.45E-06	1.28E-05	1.08E-05
AP	mol H+ eq	1.05E+00	8.47E-01	3.07E+00
EF	kg P eq	1.63E-02	7.00E-02	2.68E-02
EM	kg N eq	2.54E-01	2.08E-01	7.82E-01
ECF	CTUe	1.81E+03	7.86E+04	2.23E+03

Impact category	Unit	WH total	ONC total	OFC total
LU	Pt	2.69E+02	4.77E+04	6.67E+02
WU	m ³ depriv.	2.64E+01	1.15E+02	1.34E+02
RF	MJ	1.07E+03	2.08E+03	4.80E+03
RM&M	kg Sb eq	2.86E-03	2.27E-03	9.13E-04

The trend of Table 3.4 is also reflected in Fig. 3.7, which shows the normalized comparison of the three designs. ONC is the technique that has the highest absolute environmental impact (56.206 mPt), followed by OFC (22.76 mPt) and WH (11.10 mPt), which results in the best environmental solution. For the ONC, the most sensitive indicators were ECF (35.22 mPt) and CC (5.73 mPt). For the OFC, the indicators with the most remarkable values were CC (6.77 mPt) and RF (6.14 mPt). Finally, the indicators with the most significant input to the total impact were RM&M (3.39 mPt) and CC (2.20 mPt) for WH.

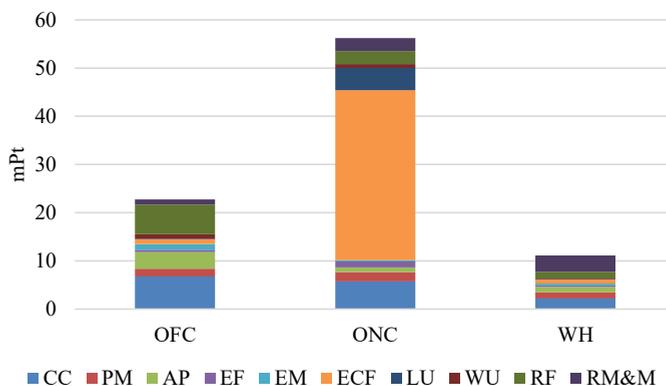


Fig. 3.7. Total Endpoint comparison between WH, ONC, and OFC scenarios. From approbation publication No. 2 [134].

- Comparison including the ASs

Fig.3.8 presents a comparative analysis between the baseline systems and all the alternative scenarios considered in the study. This comparison aims to provide a more comprehensive perspective on the results, highlighting the variations in environmental impacts across different system configurations. By incorporating alternative scenarios, the analysis enables a deeper understanding of potential improvements, trade-offs, and optimization opportunities within the macroalgae biorefinery processes.

A significant contribution to reducing the environmental impact of the WH technique is achieved by selecting a less harmful antifouling agent. The WH_AS3 scenario demonstrates a 28.02 % decrease in environmental impact compared to the baseline WH system. In contrast, improving transport efficiency through reduced diesel consumption does not yield substantial improvements, with only a 2.07 % reduction.

Regarding OFC, the selected parameters in the OFC_AS1 and OFC_AS4 scenarios do not significantly influence the final results. However, integrating solar energy through solar

panels slightly increases the environmental impact due to the materials required for panel construction, which outweighs the electricity savings. The results do not account for the carbon sequestration capacity or the nutrient removal potential of seaweed, aligning with the work of Seghetta et al. (2016) [36], which provided the LCI. Nonetheless, recent research suggests incorporating these factors [202], [203], [204]. Future studies focusing on the carbon sequestration and nutrient removal properties of *F. lumbricalis* may reveal additional environmental benefits, potentially reducing the impact of the OFC scenarios.

For ONC, replacing artificial lighting with natural sunlight in ONC_AS2 is highly influential, leading to a 78.25 % reduction in impact compared to the baseline, bringing it nearly in line with WH levels. Using solar panels for electricity generation in ONC_AS1 also results in a reduction (-26.9 %), though this is limited by the environmental footprint of the materials required for panel installation. A crucial consideration for the ONC scenario is the assumption that 100 % of electricity is sourced from a geothermal plant. If the Icelandic national energy mix were used instead (it includes 71 % hydropower), the baseline ONC scenario’s impact would decrease to 36.38 mPt, a reduction of 35.28 %. This suggests that geothermal energy is not the most efficient solution for mitigating environmental impacts in this system.

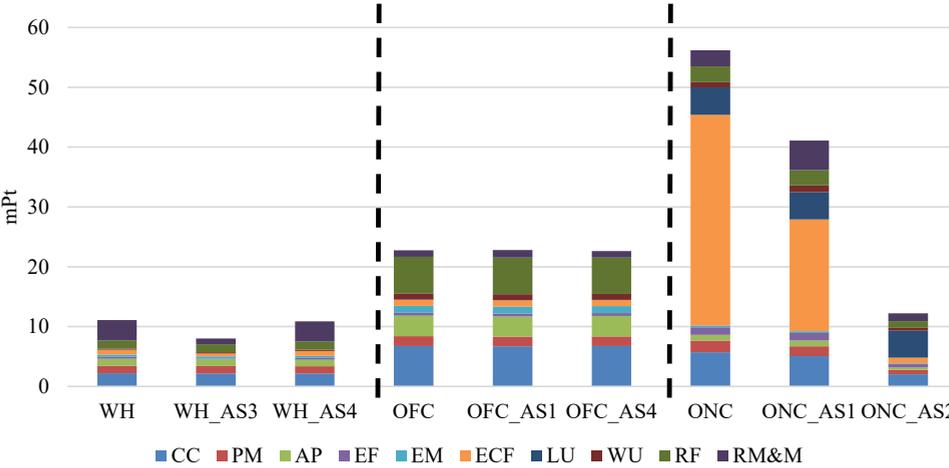


Fig.3.8. Comparison of the baseline and alternative scenarios. Note that, AS1: use of 50 % solar panel plant energy. AS2: natural sunlight instead of artificial light. AS3: Environmental friendly antifouling agent. AS4: Reduce diesel consumption of boats by 10%. From approbation publication No. 2 [134].

The scenario with the lowest environmental impact is WH_AS3, which replaces the antifouling agent with a less hazardous alternative, resulting in an impact of 7.99 mPt. Conversely, the highest impact occurs in the baseline ONC scenario (56.21 mPt). The most significant reduction in impact (-78.25 %) is observed in ONC_AS2, highlighting the crucial role of electricity consumption in this design. However, this scenario may be

impractical for a system located in Iceland due to the limited sunlight availability throughout the year, though it could be highly effective in lower-latitude regions.

For WH, the study assumes a relatively favorable macroalgae collection area located just 40 minutes from the harbor. However, this may not be the case in all locations, and given that diesel fuel is the second-largest impact contributor for this technique, results could vary significantly for longer transport distances.

These findings should not be interpreted as absolute environmental impact values for macroalgae cultivation and harvesting techniques but rather as general insights into the critical parameters that must be considered during the planning phase. Besides environmental aspects, other factors, such as total biomass production and process control capabilities, are essential in determining the most suitable system.

Results of the LCA on the full biorefinery system

The results in this section focus on the environmental impact assessment of the complete CB design for *F. lumbricalis* red macroalgae (Fig. 2.4) based on the LCI detailed in Table 2.13. The analysis begins with a hotspot assessment to identify critical impact areas within the system, followed by reinforcement through a Monte Carlo simulation and comparisons with ASs to enhance the robustness of the results. Subsequently, the environmental performance of the CB system is compared with two alternative designs: SPE (Fig. 2.6) and TLE (Fig. 2.9). A final Monte Carlo simulation is then conducted to refine the comparative analysis and assess the reliability of the findings.

- CB hot-spot results

The results for the CB, expressed in midpoint impact categories using FU1, are presented in Table 3.5. A positive value indicates that the system contributes to the given impact category, signifying an environmental burden. Conversely, a negative value represents an avoided impact, meaning the system provides an environmental benefit by mitigating certain adverse effects. The results highlight the process stages with the most significant environmental impacts, visually represented using a color gradient ranging from dark red (indicating high impact) to light green (denoting lower impact).

The extraction stages within the biorefinery emerge as the most impactful, with the third stage, dedicated to carrageenan recovery, exhibiting the highest environmental burden. Conversely, the utilization of extraction residues for biostimulant fertilizer production demonstrates environmental benefits, as reflected by the negative score.

To further validate these findings, Fig. 3.9 presents normalized and weighted endpoint results, emphasizing the most critical impact stages.

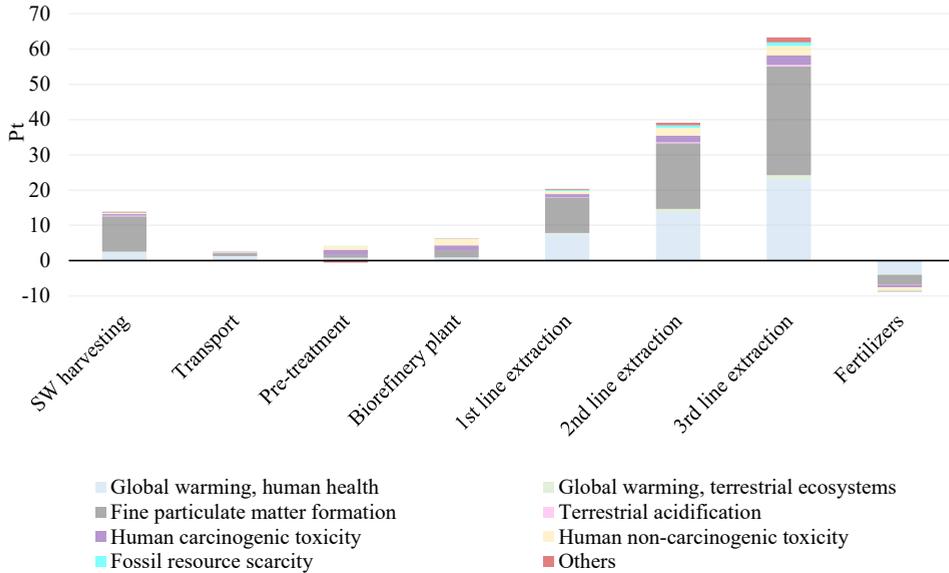


Fig. 3.9. Endpoints normalized and weighted results for the CB baseline scenario considering FU1. Note that others include all the other indicators contributing <1 % to the total impact. From approbation publication No. 4 [135].

Table 3.5

Midpoint results for the CB, baseline scenario, considering FU1. From approbation publication No. 4 [135]

	Unit	SW harv.	Trans.	Pre-treat.	Bio. plant	1 st line	2 nd line	3 rd line	Fert.	Total
GWP	kg CO ₂ eq	1.56E+02	7.87E+01	5.38E+01	5.51E+01	4.82E+02	9.02E+02	1.49E+03	2.46E+02	2.97E+03
SOD	kg CF C11 eq	1.39E-04	3.82E-05	1.33E-04	2.28E-05	3.35E-04	9.04E-04	1.00E-03	5.33E-04	2.04E-03
IR	kBqCo-60 eq	2.76E+00	1.26E+00	1.25E+01	8.29E+00	8.77E+01	1.58E+02	2.70E+02	1.40E+01	5.26E+02
OFhh	kg NO _x eq	2.93E+00	2.38E-01	1.28E-01	1.77E-01	1.61E+00	3.00E+00	4.98E+00	4.21E-01	1.26E+01
PM	kg PM _{2.5} eq	9.56E-01	6.92E-02	7.41E-02	1.88E-01	9.50E-01	1.78E+00	2.94E+00	2.60E-01	6.70E+00
OFt	k NO _x eq	2.95E+00	2.54E-01	1.33E-01	1.83E-01	1.72E+00	3.19E+00	5.32E+00	4.52E-01	1.33E+01
TA	kg SO ₂ eq	3.01E+00	1.48E-01	1.76E-01	4.74E-01	2.37E+00	4.67E+00	7.34E+00	1.02E+00	1.72E+01
EF	kg P eq	2.41E-02	5.92E-03	2.17E-01	6.21E-02	1.17E-01	2.59E-01	3.02E-01	5.16E-02	9.34E-01
ME	kg N eq	4.86E-03	2.11E-03	3.58E-01	2.69E-03	4.97E-02	2.10E-01	4.37E-02	1.97E-02	6.51E-01

	Unit	SW harv.	Trans.	Pre- treat.	Bio. plant	1 st line	2 nd line	3 rd line	Fert.	Total
TE	kg 1,4-DCB	1.89E+02	1.24E+03	2.26E+02	2.74E+03	1.53E+03	3.13E+03	4.73E+03	- 1.55E+03	1.22E+04
ECF	kg 1,4-DCB	3.02E+00	1.92E+00	3.78E+00	3.24E+01	9.18E+00	2.22E+01	2.83E+01	- 1.48E+01	8.59E+01
MEc	kg 1,4-DCB	4.06E+00	3.23E+00	5.13E+00	4.18E+01	1.28E+01	2.84E+01	3.94E+01	- 1.91E+01	1.16E+02
HCT	kg 1,4-DCB	8.37E+00	3.91E+00	2.47E+01	2.50E+01	1.66E+01	3.43E+01	4.85E+01	- 1.43E+01	1.47E+02
HNC T	kg 1,4-DCB	8.53E+01	6.13E+01	2.81E+02	5.01E+02	2.47E+02	5.98E+02	6.99E+02	- 2.42E+02	2.23E+03
LU	m _{2a} crop eq	2.49E+00	3.13E+00	1.36E+00	5.83E+00	3.86E+01	1.19E+02	1.19E+02	- 8.83E+00	2.81E+02
MRS	kg Cu eq	4.89E-01	2.04E-01	5.06E-01	2.62E+00	5.78E-01	1.44E+00	1.75E+00	- 3.56E+00	4.03E+00
FRS	kg oil eq	4.99E+01	2.48E+01	1.19E+01	1.30E+01	1.41E+02	2.59E+02	4.38E+02	- 7.21E+01	8.66E+02
WU	m ³	4.91E-01	1.56E-01	- 1.47E+01	6.37E-01	1.87E+00	1.20E+01	2.53E+01	- 3.55E+00	2.23E+01

Note: SW harv. (seaweed harvesting), trans. (transportation), Bio. Plant (biorefinery plant), 1st line (pigment extraction), 2nd line (protein extraction), 3rd line (carrageenan extraction), fert.. (fertilizers)

The total environmental burden of the CB is quantified to 140.08 Pt. The most sensitive impact categories for the system include PM (70.22 Pt) and GWP (46.02 Pt), primarily driven by energy consumption in the extraction phases and diesel fuel usage during harvesting operations.

The third-line extraction stage (63.28 Pt) is identified as the most significant contributor to the total impact, followed by the second-line extraction stage (39.09 Pt), both of which involve energy-intensive processes for protein and carrageenan recovery. Within the WH phase, the primary contributors to environmental impact are diesel fuel consumption (11.8 Pt) and the steel used in boat construction (0.65 Pt). These results differ from those presented in the previous sub-chapter due to variations in the LCIA methods employed and updates in the Ecoinvent database over the years, leading to shifts in impact calculations. The adoption of more efficient engines or alternative fuel technologies [205] could significantly reduce these impacts.

Furthermore, the environmental benefit of fertilizer production from biorefinery residues (-8.82 Pt) is highlighted, demonstrating the potential for offsetting non-bio-based market alternatives. These findings reinforce the importance of optimizing energy use and integrating more sustainable harvesting and extraction methods to improve the overall environmental performance of the *F. lumbricalis* cascade biorefinery.

To strengthen the reliability of the results, a Monte Carlo analysis was conducted, addressing the uncertainty associated with electricity consumption in the extraction processes, which represents both a critical and highly uncertain parameter. A $\pm 20\%$

variation was applied, with a total of 10,000 simulations performed under a normal distribution, centered on a mean value of 4,160.75 kWh for total energy consumption. The generated values were integrated into the LCA model to evaluate the resulting environmental impacts across different scenarios. The findings of this uncertainty analysis are illustrated in Fig. 3.10.

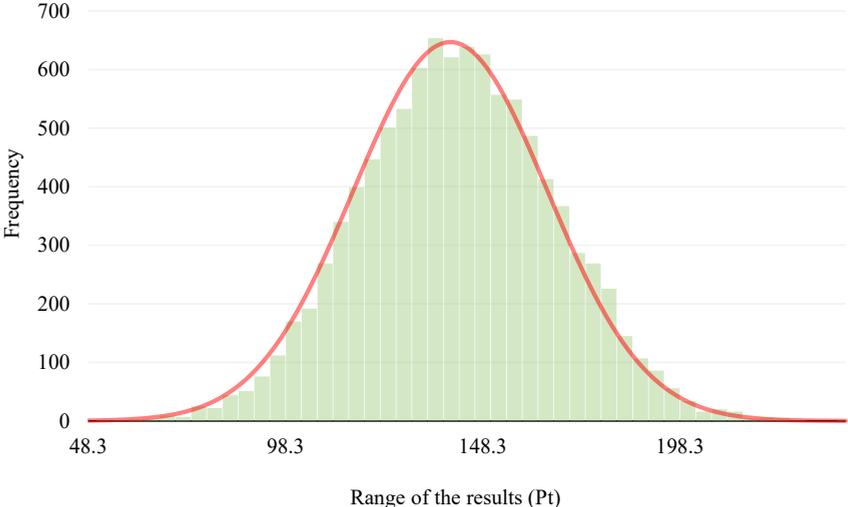


Fig. 3.10. Monte Carlo analysis range of results of the CB. From approbation publication No. 4 [135].

This analysis offers a clearer perspective on result variability. The simulations, which accounted for fluctuations in electricity consumption, produced a spectrum of possible outcomes, with the most favorable scenario reaching 48.31 Pt and the most unfavorable extending to 238.45 Pt. Despite this broad range, the majority of results remained close to the initial 140.08 Pt obtained in the baseline assessment. Within the Monte Carlo simulation, most values clustered between 132.31–136.31 Pt and 140.31–144.31 Pt, suggesting that these intervals can be considered reliable reference points for future studies.

- Comparison with ASs

Fig. 3.11 illustrates the results of the different alternative scenarios examined in the analysis.

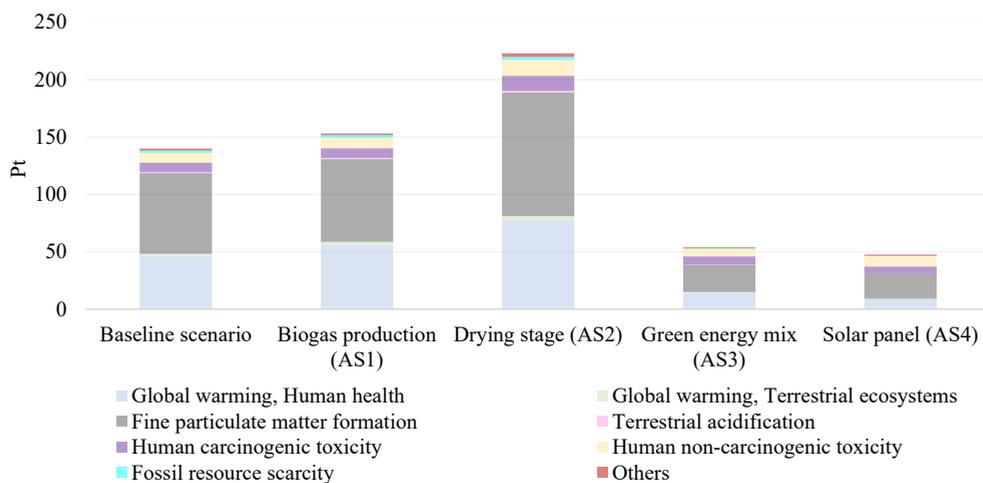


Fig.3.11. Comparison with the ASs considering FU1. Note that others include all the other indicators contributing <1 % to the total impact. From approbation publication No. 4 [135].

Among the evaluated scenarios, AS2 exhibits the highest environmental impact, reaching 222.89 Pt, due to the integration of a drying stage for biomass conservation. The substantial increase in energy consumption required to reduce the water content in *F. lumbricalis* is the primary driver of this impact, with the drying stage alone contributing 37 % of the total. The inclusion of drying in the biorefinery process is a critical consideration that necessitates careful assessment during process design. This evaluation should determine whether drying is essential and, if so, identify the most efficient technological solutions to minimize energy consumption.

Scenarios incorporating alternative electricity sources demonstrate significant reductions in environmental impact. AS3, which utilizes a REC energy mix, results in an impact of 53.97 Pt, while AS4, integrating solar panel production, achieves the lowest footprint at 47.48 Pt. These scenarios outperform the baseline case, which relies on the Estonian energy mix, with AS3 and AS4 reducing environmental impact by 61 % and 66 %, respectively.

A comparison between AS1, involving biogas production from extraction residues, and the baseline scenario highlights that fertilizer production remains the more sustainable end-of-life strategy. Fertilizer application results in an impact of -8.82 Pt, significantly lower than the 4.30 Pt associated with biogas generation. This difference arises from the absence of additional technological development required for fertilizer application. The negative impact values stem from system boundary expansion, assuming that *F. lumbricalis*-derived fertilizer substitutes conventional market fertilizers. A similar assumption applies to the biogas scenario, where electricity generated from biogas cogeneration is assumed to replace electricity from the national grid. However, biogas cogeneration necessitates additional technological advancements and infrastructure, leading to higher associated impacts.

- Comparison with alternative designs (SPE and TLE)

This section compares the environmental performance of the CB with the two alternative designs (SPE and TLE) using FU2, which focuses on the process output to ensure a more precise characterization of the analysis. By normalizing the results based on product yield, FU2 provides a clearer assessment of the efficiency and environmental trade-offs associated with each biorefinery design. The comparative results are illustrated in Fig. 3.12, highlighting the differences in environmental impact across the three scenarios.

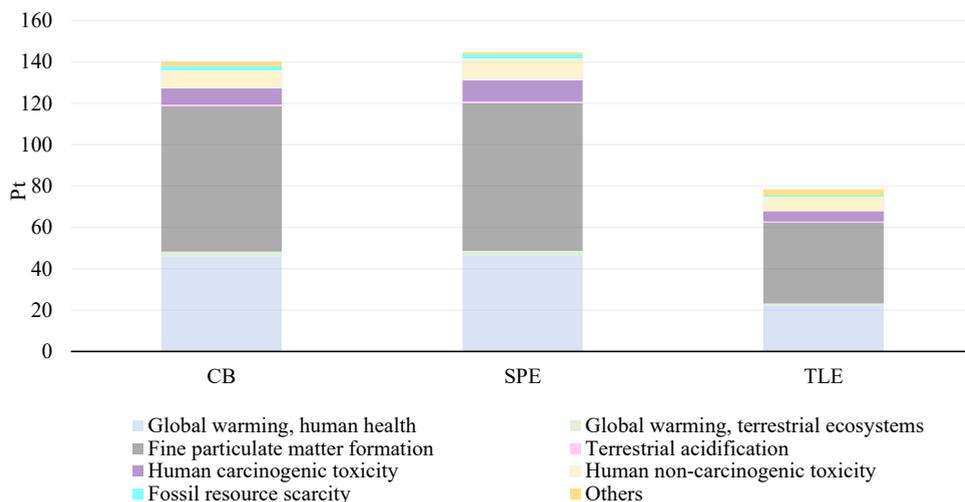


Fig. 3.12. Comparison between the CB system, the SPE, and the TLE referring to the FU2. Note that others include all the other indicators contributing <1 % to the total impact. From approbation publication No. 4 [135].

The SPE scenario represents the least efficient biorefinery design, with an environmental impact of 144.60 Pt. While the CB demonstrates a modest improvement, reducing the impact by approximately 4 % compared to SPE, its primary advantage lies in optimizing resource utilization rather than achieving absolute reductions in environmental burden. The cascade approach enables the extraction of equivalent product yields using only 1 ton of biomass, whereas SPE requires 3 tons, highlighting the superior efficiency of the CB design. In contrast, TLE exhibits the lowest impact at 78.39 Pt, suggesting that a three-line extraction biorefinery could be a viable alternative to the cascade model in terms of sustainability. However, TLE does not fully achieve the same product yield as the CB. To ensure comparability with FU2, additional conventional market products have been integrated into the analysis to balance system outputs. This strategy not only aligns with the increasing demand for bio-based compounds in pharmaceutical [206], cosmetics [207], and food applications [208] but also facilitates market expansion by catering to emerging consumer preferences for sustainable products [49], [209].

- Sensitivity analysis

A Monte Carlo analysis was conducted, applying the same parameters established in Chapter 2.2, with 10,000 simulations incorporating a $\pm 20\%$ variation in energy consumption during the extraction phase. This analysis provided deeper insights into how these fluctuations impact the comparative evaluation of all scenarios within a single framework (Fig. 3.13). The results fluctuated between 19.65 Pt and 239.45 Pt, with SPE showing a range from 36.37 Pt to 239.48 Pt and TLE varying from 19.65 Pt to 129.88 Pt. These findings highlight the significant overlap between the CB and SPE, reinforcing the similarities observed in Fig. 3.12. Furthermore, an intriguing overlap emerged among all three scenarios, spanning 47.64 Pt to 143.64 Pt, covering approximately 17% of the simulation dataset. This suggests that in 83% of cases, TLE exhibited a lower environmental impact compared to both SPE and the CB, further supporting the potential advantages of a three-line extraction biorefinery in terms of sustainability.

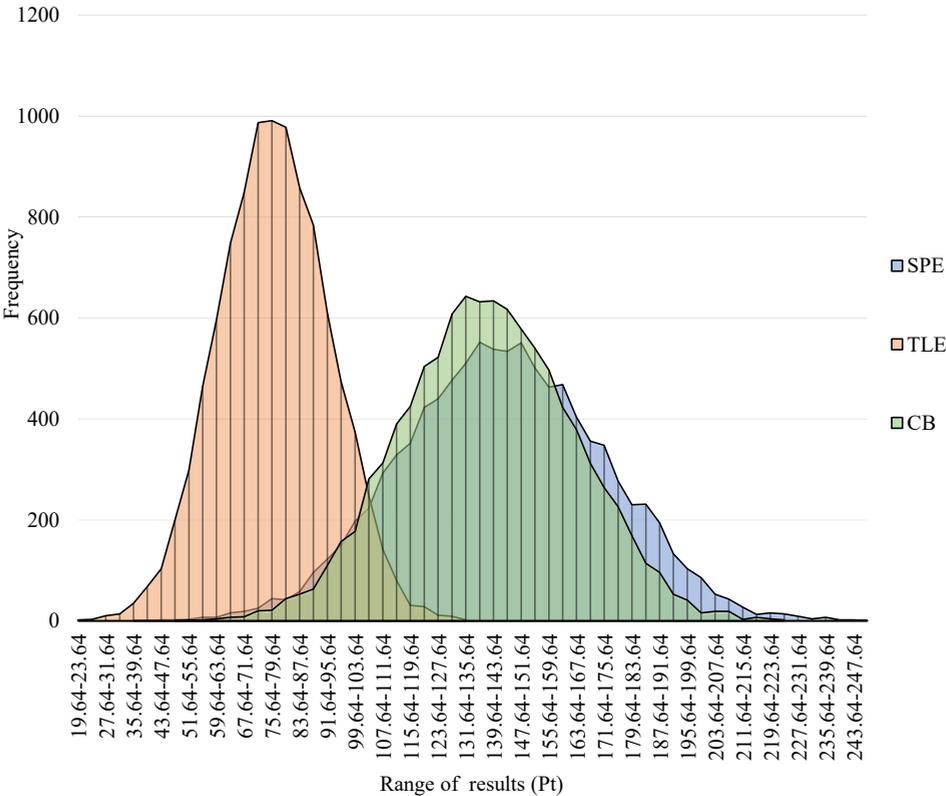


Fig. 3.13. Monte Carlo analysis for the comparison between CB, SPE, and TLE. From approbation publication No. 4 [135].

- Take-home messages

This LCA study quantifies the environmental impact of a macroalgae-based biorefinery and explores potential improvements for enhancing its overall sustainability. The findings highlight the benefits of adopting a CB approach over an SPE system, emphasizing the

efficiency gained through the systematic reuse of extractive residues. This biorefinery model holds significant potential in the Baltic Sea Region, not only fostering new economic opportunities for local communities but also promoting the sustainable exploitation of an abundant natural resource. Despite these advantages, opportunities remain to further optimize the system and reduce its environmental impact.

One of the most critical factors influencing the environmental footprint is energy consumption, particularly the electricity required for biomass heating during protein and carrageenan extraction. This aligns with Nilsson et al. (2022) [163], who identified chemical extraction as the primary contributor to environmental impacts in biorefineries. Specifically, protein and carrageenan recovery exhibit higher energy demand than pigment extraction, reinforcing the strategic advantage of selecting aqueous extraction methods with lower energy requirements.

To mitigate the environmental impact associated with energy use, two key strategies can be considered. First, from a technological standpoint, alternative equipment and innovative extraction techniques could be introduced to reduce energy consumption, particularly for protein and carrageenan extraction, which are the most energy-intensive processes. EAE employs specific enzymes to break down cell walls, facilitating compound release at lower temperatures and reducing mechanical and thermal energy requirements [210]. Similarly, MAE and UAE enhance cell disruption, improving extraction efficiency while minimizing energy use and processing time [211], [212]. scCO₂ utilizes CO₂ as a solvent at lower temperatures, offering a non-toxic and energy-efficient alternative for bioactive compound extraction [213]. However, these technologies remain in the experimental stage and require further development before achieving industrial-scale implementation [26], [214]. Arias et al. (2025) [138] recently compared EAE, UAE, and traditional water extraction, highlighting that EAE achieved the greatest energy savings, resulting in over 90 % impact reduction compared to water extraction and over 15 % compared to UAE. These findings offer a promising foundation for future developments. Nonetheless, further comparative analyses of the same macroalgae species are needed, as environmental performance is highly species-specific. Additionally, scaling up such assessments to an industrial level would provide a more realistic understanding of environmental sustainability.

The second strategy involves transitioning to a cleaner energy mix or incorporating renewable energy sources such as photovoltaics. As evidenced in this study, shifting to renewable energy can reduce the environmental footprint by over 50 % without altering the initial energy demand or technological setup. Golbert et al. (2021) [127], proposed an Organic Rankine Cycle system that integrates solar thermal energy with macroalgae biorefineries, enhancing energy efficiency by capturing and converting low-grade thermal energy into electricity, further reducing reliance on fossil-based power sources.

Another critical factor influencing the environmental performance of the biorefinery is the drying stage, which plays a vital role in preserving biomass integrity throughout the cascade process. This stage is closely linked to energy consumption and significantly impacts overall sustainability. The findings align with van Oirschot et al. (2017) [215], who identified drying as the most environmentally impactful step in macroalgae processing,

contributing over 70 % of the total impact. Evaluating the necessity of this stage during the design phase is essential, as it may be possible to eliminate drying through improved biomass management and better alignment between harvesting and processing schedules.

Beyond optimizing the macroalgae-based biorefinery, these results lay the groundwork for future comparative studies between macroalgae and traditional land-based biorefineries, such as those utilizing corn, sugarcane, or forestry residues. While direct comparisons are complex due to differences in feedstock characteristics, system boundaries, and methodologies [216], macroalgae offer clear advantages, particularly in avoiding land use conflicts and reducing freshwater and fertilizer requirements. However, land-based systems benefit from well-established agricultural infrastructure and supply chains, contributing to greater extraction efficiency and lower energy demands [217].

The LCA analysis acknowledges inherent uncertainties, particularly given the early-stage development of macroalgae biorefineries and the limited availability of industrial data. These challenges necessitate certain simplifications, including assumptions related to machine sizing, extraction efficiencies, purity levels, and the applicability of extracted compounds. While data accuracy for WH was robust, detailed extraction phase information was limited, requiring reliance on empirical formulas and proxy data for scaling up pilot-scale operations. Future research should incorporate advanced chemical process simulation tools such as SuperPro Designer or Aspen Plus to improve the accuracy of cascade biorefinery models and better reflect industrial conditions. Sensitivity analyses of key parameters would also help refine process decisions, ensuring more robust LCA results.

Additionally, integrating techno-economic assessments through simulation software would provide a clearer understanding of the feasibility of different technological configurations, allowing for better market-based decision-making.

The potential ecological consequences of *F. lumbricalis* harvesting in Estonia's marine environment were not explicitly considered in the results. Monitoring the long-term effects of macroalgae depletion on surrounding ecosystems, such as nutrient cycling and biodiversity, remains a challenge, as quantifying these changes within an LCA framework is inherently complex [218].

Further research is also needed to examine seasonal variations in the chemical composition of *F. lumbricalis*, as these fluctuations directly influence extraction yields and product quality. Understanding how harvesting timing impacts sustainability will be essential for optimizing the overall performance of macroalgae biorefineries [219].

3.2. Economic results

In this sub-chapter, the results of the LCC analysis are presented, following the framework outlined in Chapter 2.3. The analysis begins with a general economic characterization of the systems, including an overview of operational costs, capital investments, and potential revenue streams. This initial assessment aims to identify economic hotspots and key financial drivers within the system, offering a preliminary understanding of its economic viability.

Subsequently, a comprehensive economic comparison of the different biorefinery designs, CB, SPEp, SPEpr, SPEc, and TLE, is conducted. This evaluation integrates key financial indicators such as NPV, IRR, TER, and IR to provide a quantitative assessment of the economic feasibility and profitability of each design.

Economic analysis results

- Operational costs

Fig.3.14 presents a comparative analysis of the operating costs associated with the five biorefinery designs.



Fig. 3.14. Operative costs for the CB, SPEp, SPEpr, SPEc, and TLE. These values are reported for 1 year.

The differences in operational expenditures among the various production scenarios are relatively small. As anticipated, the CB incurs the highest operating cost, amounting to 5,354,337 €/year. This is attributed to its complexity and the simultaneous production of multiple high-value end products, necessitating a more extensive processing setup. Among the single-product extraction designs, SPEp (3,766,787 €/year) and SPEc (3,771,810 €/year) exhibit the lowest operational costs, demonstrating nearly identical expenditure levels. In contrast, SPEpr (4,642,929 €/year) and TLE (4,592,366 €/year) fall within an intermediate cost range, reflecting their slightly higher operational demands compared to the more cost-efficient single-extraction designs, yet remaining lower than the multi-product cascade system.

Fig. 3.15 gives a detailed breakdown of the operating costs associated with the CB design, which holds particular significance as it represents the highest overall cost among the evaluated biorefinery configurations and serves as the focal design approach of this Thesis. The cost distribution highlights the primary economic burdens within the cascade biorefinery system, providing insights into the key cost drivers and potential areas for optimization. Understanding these cost components is essential for assessing the financial feasibility of the CB model and identifying strategies for improving economic efficiency while maintaining the sustainability advantages of the multi-product extraction approach.

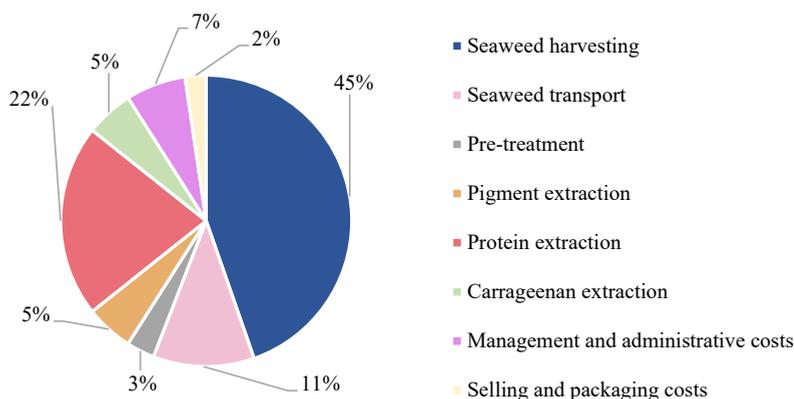


Fig. 3.15. Operative costs for the CB design. These values are reported for 1 year.

The most significant cost category within the CB design is related to *F. lumbricalis* harvesting procedures, amounting to 2,391,458 €/year, with 90 % of this expense attributed to fuel costs for extraction and transport, making harvesting nearly half of the total operating costs. The second most expensive stage is protein extraction, costing 1,143,930 €, primarily driven by the high cost of enzymes (e.g., alcalase) required for the extraction process. The third highest expense is seaweed transport, totaling 601,568 €, which is also heavily influenced by fuel consumption. These findings indicate that fuel dependency and enzyme costs are the dominant economic burdens within the cascade biorefinery system, emphasizing the need for targeted strategies to optimize fuel efficiency and explore alternative or more cost-effective enzymatic extraction methods.

- Investments

Fig. 3.16 illustrates the investment costs associated with the different biorefinery designs. The TLE design requires the highest investment cost (3,459,086€) due to the necessity of operating three extraction lines simultaneously, which demands a greater number of machines and infrastructure. CB follows with an investment cost of 2,058,558 €, reflecting the complexity of the cascade biorefinery system. In contrast, the SPE designs require the lowest initial investments, as anticipated, since they focus on single-product extraction, reducing equipment and processing requirements.

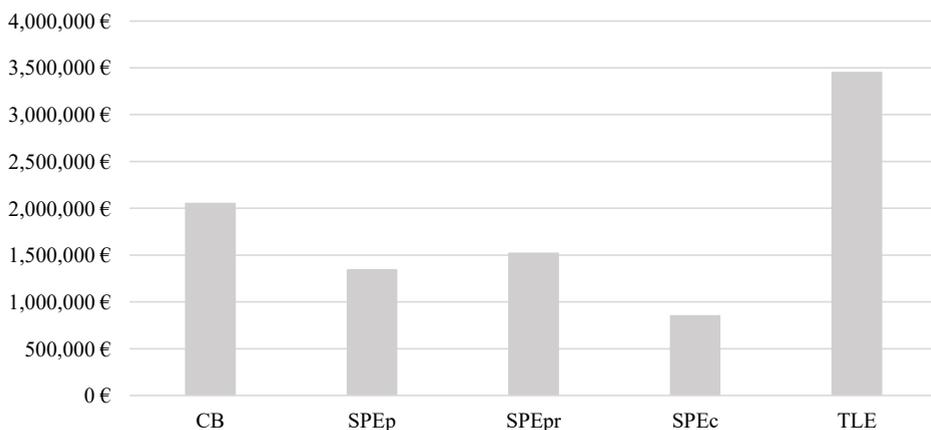


Fig. 3.16. Investment costs for the CB, SPEp, SPEpr, SPEc, and TLE. These values are reported for 1 year.

Fig. 3.17 provides a detailed breakdown of the investment costs for CB, offering insights that are broadly applicable to the other biorefinery designs. The Foodec 65 decanters represent the largest investment, accounting for 34 % of the total, as they are crucial for separating feedstock from liquids, a fundamental step in producing high-quality pigments, proteins, and carrageenan. The micro- and ultrafiltration stages follow, constituting 24 % of the total investment, essential for ensuring product purity. The PurePulp 750 and Clara 200 High Flow high-speed separators contribute 22 % and 13 %, respectively, reflecting their role in refining the extracted bio-compounds. Additional investments, although smaller in scale, include the trawling boat, nylon nets for harvesting, annual fishing licenses, an electric unloader (5-6 kW hydraulic pump), and land acquisition. These elements, while representing a smaller share of the total investment, are necessary for securing a functional and efficient seaweed biorefinery system.

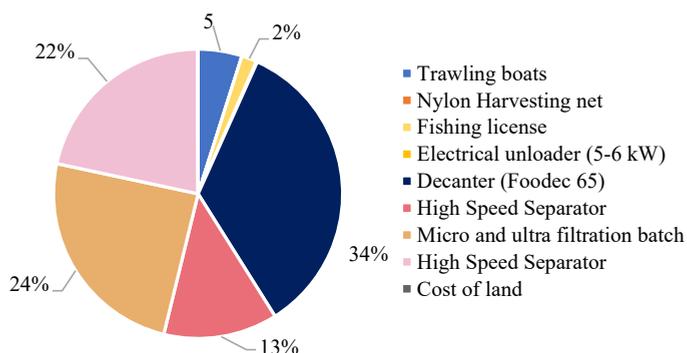


Fig. 3.17. Breakdown of the investment cost for the CB biorefinery design.

- Sales scenarios

An analysis of the sales impact, incorporating both the WCS and BCS, has been conducted to assess the financial resilience of the biorefinery designs under different market conditions. The results are presented in Fig. 3.18, illustrating the revenue variations across different economic scenarios. The WCS assumes the lowest pigment unit price and the minimum harvestable *F. lumbricalis* under current licensing conditions, whereas the BCS reflects the highest pigment market value and an extended harvesting limit.

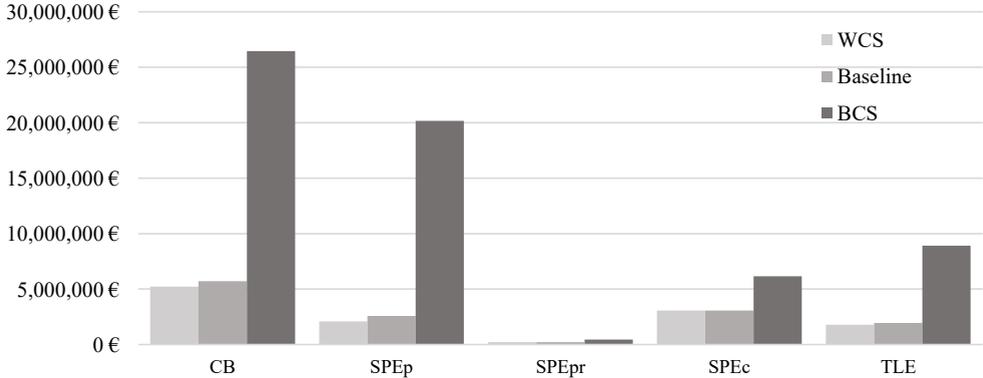


Fig. 3.18. Comparative analysis of the sales scenarios, including WCS and BCS.

These findings, centered on product sales, offer an initial perspective on the economic viability of various biorefinery configurations. As expected, CB and SPEp, which prioritize pigment extraction, achieve the highest revenue generation. In contrast, SPEpr, which focuses on protein production, exhibits significantly lower earnings, increasing the likelihood of economic infeasibility. While these data do not constitute a comprehensive economic assessment, they serve as a fundamental basis for the analysis. Establishing this foundation ensures that subsequent economic evaluations can be analyzed with greater precision, enhancing the study’s overall validity and reliability.

- Environmental monetarization results

The environmental externalities were assessed using the methodology detailed in Chapter 2.3. Fig. 3.19 illustrates a comparative analysis of the environmental costs across different biorefinery configurations. The results align with the initial LCA findings, with CB incurring the highest environmental cost at 707,731 €, followed by TLE (441,437 €), SPEc (421,116 €), SPEpr (304,595 €), and SPEp (202,964 €). However, when factoring in the total product output of each system, the interpretation of these results changes. To achieve the same product yield as CB, the combined outputs of SPEp, SPEpr, and SPEc would be required, culminating in a total environmental cost of 928,676 €, thereby rendering this combination more environmentally costly than CB.

A closer analysis of the factors driving environmental costs indicates that GWP constitutes the largest share (63 %), followed by PM (13 %) and FRS (12 %). These findings deviate from the initial LCA results, where PM had the most significant impact, and FRS played a minor role. This discrepancy highlights the influence of the monetization factors

defined in Table 2.20, emphasizing how their application can substantially alter the interpretation of environmental costs

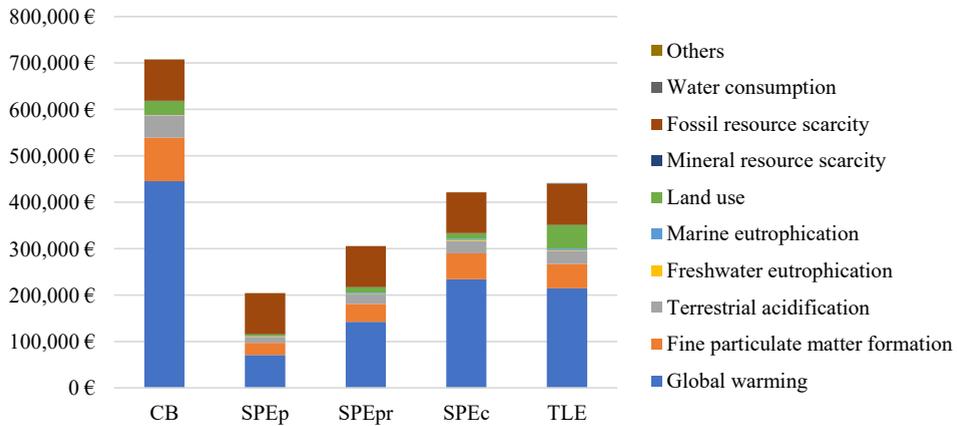


Fig. 3.19. Comparative analysis of the environmental externalities. Note that others include the sum of all the ReCiPe2016 indicators with an impact lower than 1 €.

Comparative economic assessments

Table 3.6 presents a comprehensive economic assessment of various biorefinery scenarios over a 10-year period.

Table 3.6

Comparative economic analysis of the different scenarios

	CB	SPEp	SPEpr	SPEc	TLE
<i>Worst case scenario (WCS)</i>					
ROI	-154 %	-802 %	-1,934 %	-389 %	-1,262 %
NPV	-3,168,612€	-13,623,485 €	-31,904,995 €	-6,964,791 €	-21,060,533 €
IRR	Not Calculable				
PI	-1.54	-6.62	-15.50	-3.38	-10.23
<i>Baseline scenario</i>					
ROI	41 %	-596 %	-1,934 %	-389 %	-1,194 %
NPV	-17,318 €	-10,306,897 €	-31,904,995 €	-6,964,791 €	-19,954,451 €
IRR	-0.17 %	Not Calculable	Not Calculable	Not Calculable	Not Calculable
PI	-0.01	-5.01	-15.50	-3.38	-9.69
<i>Best case scenario (BCS)</i>					
ROI	5,147 %	4,111 %	-3,768 %	-679 %	-209 %
NPV	82,437,798 €	65,710,001 €	-61,516,773 €	-11,636,363 €	-4,055,130 €
IRR	559 %	447 %	Not Calculable	Not Calculable	Not Calculable
PI	40.05	31.92	-29.88	-5.65	-1.97

In the WCS, all ROI values are negative, indicating sustained financial losses across all designs. SPEpr and TLE exhibit the most severe deficits, with ROIs of -1,934 % and -1,262 %, respectively, rendering them highly unattractive for investment. NPV analysis further confirms the unprofitability of all designs, with SPEpr standing out as the worst

performer, reporting an NPV of -31,904,995 €. None of the scenarios yields a calculable Internal IRR, reinforcing their inability to generate positive returns. The PI values are also negative, with SPEpr displaying the lowest figure (-15.50), making it the least viable option.

In the baseline scenario, CB is the only design with a positive ROI of 41 %, suggesting some potential for profitability, whereas all other configurations remain unprofitable, with SPEpr still at the lowest ROI (-1,934 %). Despite CB's relative advantage, its NPV remains negative, indicating difficulty in recouping initial investment costs. The other designs continue to show negative NPVs, with SPEpr again suffering the largest loss (-31,904,995 €). IRR analysis reflects these findings, as CB's IRR hovers near zero, making it difficult to ensure a stable return. Additionally, CB's PI is nearly neutral, implying it is close to breaking even, whereas the other scenarios maintain negative PIs, confirming their ongoing unprofitability. While CB demonstrates the best financial performance among the evaluated options, its stability remains questionable for industrial-scale implementation.

In the BCS, both CB and SPEp exhibit highly positive ROIs of 5,147 % and 4,111 %, respectively, indicating substantial profitability under optimal market conditions. However, SPEpr, SPEc, and TLE continue to show significant financial losses. CB achieves an NPV of 82,437,798 €, followed by SPEp at 65,710,001 €, both indicating strong financial viability. In contrast, SPEpr maintains the highest financial loss (-61,516,773 €). The IRR results mirror these trends, with CB and SPEp demonstrating strong positive IRRs of 559 % and 447 %, while the other designs remain financially nonviable. Similarly, CB and SPEp report positive PIs of 40.05 and 31.92, confirming their economic feasibility, while SPEpr, SPEc, and TLE remain in negative territory.

Overall, CB emerges as the most robust and viable design, showing resilience in the Baseline Scenario and demonstrating strong profitability in the BCS while incurring comparatively lower losses in the WCS. These findings underscore that the economic sustainability of an *F. lumbricalis* biorefinery is heavily reliant on revenue generation from high-value pigment extraction, which significantly drives profitability. Conversely, SPEpr, SPEc, and TLE are consistently unprofitable across all scenarios, suffering substantial losses. This analysis highlights that biorefinery models focusing on single-product extractions (except for pigments) or straying from a cascade biorefinery approach are economically unsustainable.

Economic discussion and take-home messages

The analysis highlights that raw material extraction and transport are the most significant contributors to operating costs, emphasizing logistics and *F. lumbricalis* procurement as the primary expense factors. This is primarily due to the cost of fuel for the diesel-powered trawling boat, a concern exacerbated by the fluctuating energy prices observed in Europe in recent years [220], largely influenced by geopolitical instability and the urgent shift toward sustainable alternatives to fossil fuels, which further increases transportation costs.

Among the analyzed biorefinery designs, CB incurs the highest operating costs due to its comprehensive approach, encompassing all necessary steps to produce a full range of

marketable products, including pigments, proteins, and carrageenan. While this strategy maximizes biomass valorization, it also presents significant financial challenges.

Investment costs are also substantial, with the Foodec 65 decanter representing the most expensive item. This machinery, along with advanced filtration technologies, is essential for ensuring efficient feedstock separation and high-quality end-product recovery, both crucial requirements for the operational success of a sustainable biorefinery. This finding was further reinforced by the preliminary economic comparison conducted on alternative pigment extraction methods. Regardless of whether EAE, UAE, or water-based extraction was selected, the analysis consistently highlighted capital expenditure as the most critical economic factor. In particular, the total investment required approached € 1.5 million, underlining the importance of carefully managing infrastructure-related costs during early-stage biorefinery planning [138].

The economic viability of the project depends heavily on market conditions and biorefinery design choices. Simulations reveal significant variability in financial returns, largely dictated by market prices and production volumes. In optimistic scenarios, where the pigment price reaches 500,000 €/ton, CB demonstrates strong profitability and resilience, offering substantial returns. However, under unfavorable conditions with pigment prices at 100,000 €/ton, profit margins shrink drastically, often resulting in financial losses across all designs. This underscores the project's vulnerability to market fluctuations and highlights the need for robust risk management and adaptive market strategies, given that pigment sales constitute the primary revenue stream for the *F. lumbricalis* biorefinery.

From a financial perspective, securing government subsidies and funding that promote circular and sustainable economic models could support the purchase of machinery and technological advancements. Estonia's access to resources such as the Just Transition Fund (3.5 billion euros for 2021–2027) [221], the EU-funded Recovery and Resilience Plan (936.3 million euros) [222], the LIFE programme, and the Green Fund (90 million euros) [223] provides potential avenues for financial assistance. Additionally, leasing options could reduce upfront investment costs and allow for more frequent equipment upgrades, though careful market analyses would be necessary to identify viable suppliers. Operational optimization could also enhance profitability by implementing feedback systems to monitor equipment performance, predictive maintenance based on real-time data, and detailed process analyses to improve efficiency and ensure stable workflows.

Despite these opportunities, this research also faces limitations. The study primarily focuses on products intended for the animal feed market, excluding higher-value applications in human nutrition or pharmaceuticals due to technological and purity constraints. Developing technologies to achieve higher-purity products could significantly enhance profitability. The analysis also assumes stable and secure markets for all biorefinery outputs, a critical limitation, as insufficient market demand could make the project financially unsustainable. Additional market research is necessary to validate product demand. Furthermore, it is essential to recognize the preliminary nature of the results, which serve as an initial guideline. Future industrial implementations will require

refinements as technology matures and more precise market data becomes available. The investment in machinery represents a substantial expense, and this Thesis did not include a cost-benefit analysis comparing quotations from multiple suppliers due to its exploratory nature. Conducting such analyses in future evaluations could lead to significant cost savings. Other risks include potential regulatory changes affecting seaweed harvesting limits and fluctuations in input costs, such as enzymes and energy.

While the *F. lumbricalis* biorefinery demonstrates strong potential for profitability under favorable market conditions, particularly with the CB design, its success is heavily dependent on external factors such as market prices and regulatory environments. Addressing these challenges through financial support, operational optimization, and comprehensive market analysis will be crucial in mitigating risks and maximizing benefits. The findings of this study provide a foundational reference for future efforts to establish industrial-scale *F. lumbricalis* biorefineries. The future prospects of these biorefineries are closely tied to the market potential of their extracted products. The pigment market, for example, is expanding rapidly due to growing demand for natural and sustainable colorants. Algal pigments like phycocyanin (from blue-green algae) and fucoxanthin (from brown macroalgae) are increasingly used in food, cosmetics, and pharmaceuticals [224]. The global pigment market, including algal pigments, was valued at \$ 24.13 billion in 2023 and is projected to reach \$ 35.60 billion by 2032, growing at a compound annual growth rate (CAGR) of 6 % [225]. Similarly, the algae protein market is rising, driven by the demand for sustainable protein alternatives. Expected to grow at a CAGR of 7 % from 2023 to 2032 [226], this sector benefits from increasing consumer awareness of plant-based proteins' health benefits and the rise of veganism. Algal proteins are valued for their high nutritional content and reduced environmental footprint compared to animal-based proteins [227], making them particularly appealing to environmentally and health-conscious consumers. Carrageenan, another key product, is also experiencing market growth, with an estimated value of \$ 850 million in 2022, projected to reach \$ 1.55 billion by 2032 [228], fueled by demand in the food, pharmaceutical, and cosmetics industries. Its multifunctional properties and natural origin make it especially attractive in regions with stringent additive regulations [229].

On the technological front, advancements in macroalgae harvesting, cultivation, and processing will be critical to improving productivity and sustainability. The development of on-shore photobioreactors optimized for light and nutrient usage has the potential to significantly enhance algal yields [230]. Additionally, genetic engineering could facilitate the creation of strains with increased productivity and greater resilience to environmental stresses [231]. Innovations in advanced membrane technologies promise to reduce operational costs and improve biomass purity [232]. The integration of low-energy processing techniques and efficient extraction methods, such as green solvents and supercritical extraction, can enhance overall biorefinery sustainability [233]. Furthermore, the incorporation of solar panels and renewable energy sources could reduce dependence on fossil fuels, lower greenhouse gas emissions, and stabilize energy costs while improving public perception of sustainability efforts.

Despite these promising developments, several challenges remain. High production costs, particularly substantial initial investments in infrastructure and advanced technologies, represent a significant financial barrier, as demonstrated by this study's findings. Operational costs, especially energy requirements, further compound economic challenges. Another major obstacle is the scalability of macroalgae processing technologies [234]. While systems may perform well at laboratory or pilot scales, transitioning to industrial-scale operations presents challenges such as altered growth dynamics, increased logistical complexity, and reduced energy efficiency. Moreover, managing a macroalgae biorefinery requires specialized expertise in fields such as biomass biology, chemical engineering, and environmental management, necessitating skilled and highly paid personnel.

These challenges underscore the complexity of macroalgae biorefineries while also highlighting opportunities for technological innovation, sustainable development, and job creation. With further refinement, *F. lumbricalis* biorefineries could play a pivotal role in the circular bioeconomy, provided that financial, technological, and environmental challenges are strategically addressed through policy support, investment, and innovation.

3.3. Social results

In this Chapter, the social aspects associated with the *F. lumbricalis* biorefinery system will be evaluated. The analysis will first present the results obtained using the IPA, which assesses social risks and potential impacts through a sector-level analysis. This will be followed by the results derived from the RSA, which focuses on a more localized, enterprise-level evaluation. By integrating both methodologies, the outcomes aim to provide a comprehensive understanding of the social implications across different scales, highlighting key risk factors, opportunities for social improvements, and potential trade-offs in the sustainability assessment of the *F. lumbricalis* biorefinery.

Results of the IPA approach

Using the IPA approach, the primary objective is to develop a social hotspot map for the WH and OFC value chains in the production of *F. lumbricalis*. These reference systems are detailed in Chapters 2.1 and 2.4, specifically in Fig. 2.3 and 2.18, while the corresponding LCI are provided in Tables 2.23 and 2.24. This analysis aims to identify potential social risks within the supply chains, evaluating key indicators such as labor conditions, community well-being, and stakeholder engagement

- WH results

Table 3.7 presents the results for the sub-categories in the WH scenario, using a color-coded scale ranging from red (indicating the highest potential social risk) to green (representing the lowest potential social risk). This visual representation facilitates a clear identification of critical social risks associated with the WH production system.

Table 3.7

Sub-category results for the WH scenario

Impact category	Seaweed harvesting	Maintenance of boat	Transport to the harbor	Boat unloading	Total	Unit
Wage assessment	0.01508	0.00454	0.00486	0.00695	0.0314	mrheq
Workers in poverty	0.01135	0.00301	0.00216	0.00433	0.0208	mrheq
Child labor	0.01274	0.00355	0.00197	0.00519	0.0234	mrheq
Forced labor	0.02167	0.00576	0.00506	0.00946	0.0419	mrheq
Excessive WkTime	0.01588	0.00431	0.00378	0.00704	0.0310	mrheq
Freedom of assoc	0.01269	0.00310	0.00248	0.00641	0.0247	mrheq
Migrant labor	0.00982	0.00241	0.00201	0.00421	0.0185	mrheq
Social benefits	0.00711	0.00190	0.00116	0.00352	0.0137	mrheq
Labor laws/conv	0.00387	0.00119	0.00125	0.00152	0.0078	mrheq
Discrimination	0.01868	0.00529	0.00403	0.00811	0.0361	mrheq
Unemployment	0.00666	0.00165	0.00170	0.00274	0.0127	mrheq
Occ tox & haz	0.01799	0.00476	0.00298	0.00860	0.0343	mrheq
Injuries & fatalities	0.02456	0.00658	0.00692	0.01012	0.0482	mrheq
Indigenous rights	0.00537	0.00143	0.00073	0.00230	0.0098	mrheq
Gender equity	0.00759	0.00214	0.00114	0.00311	0.0140	mrheq
High conflict zones	0.00958	0.00235	0.00222	0.00472	0.0189	mrheq
Non-Communicable diseases	0.00256	0.00058	0.00045	0.00114	0.0047	mrheq
Communicable diseases	0.00747	0.00210	0.00103	0.00296	0.0136	mrheq
Poverty and inequality	0.01278	0.00342	0.00240	0.00583	0.0244	mrheq
State of env sustainability	0.01572	0.00438	0.00237	0.00691	0.0294	mrheq
Legal system	0.01243	0.00312	0.00397	0.00569	0.0252	mrheq
Corruption	0.00680	0.00159	0.00190	0.00274	0.0130	mrheq
Democracy & freedom of speech	0.01739	0.00457	0.00633	0.00873	0.0370	mrheq
Access to drinking water	0.00799	0.00212	0.00118	0.00315	0.0144	mrheq
Access to sanitation	0.01306	0.00378	0.00194	0.00522	0.0240	mrheq
Children out of school	0.00922	0.00238	0.00192	0.00463	0.0182	mrheq
Access to hospital beds	0.01176	0.00321	0.00235	0.00449	0.0218	mrheq
Smallholder v commercial farms	0.00255	0.00062	0.00028	0.00231	0.0058	mrheq
Access to electricity	0.00547	0.00159	0.00048	0.00218	0.0097	mrheq
Property rights	0.01390	0.00393	0.00220	0.00546	0.0255	mrheq

The analysis clearly indicates that the harvesting operation represents the stage with the highest potential social risk, followed closely by the boat unloading procedures. Among the evaluated indicators, *Injuries & Fatalities* exhibits the highest potential risk (0.0482 mrheq), highlighting concerns related to occupational safety within the sector. Additionally, *Forced Labor* (0.0419 mrheq) and *Discrimination* (0.0361 mrheq) emerge as critical social risks, warranting further attention.

To provide a clearer perspective, Fig. 3.20 presents the normalized and weighted results, categorized into different damage categories, enabling a more comprehensive interpretation of the social implications associated with the WH scenario.

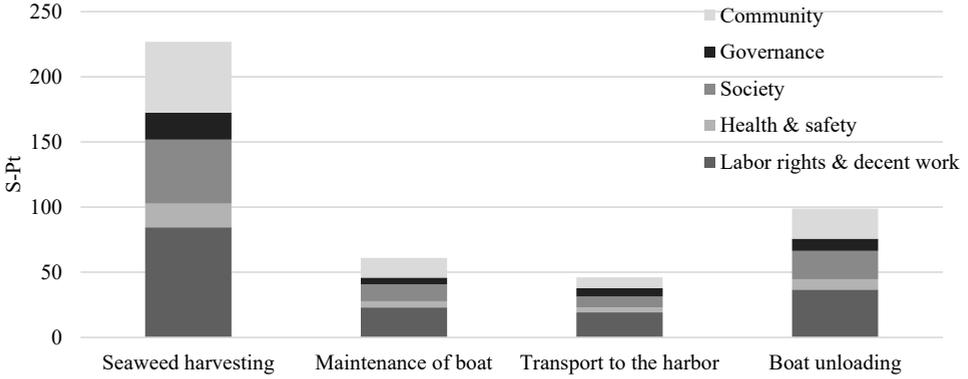


Fig. 3.20. Normalized and weighted results for the WH scenario.

Seaweed harvesting stands out as the stage with the highest potential social risk, with a total impact of 226.85 S-Pt. Among the individual indicators, *access to electricity* (22.36 S-Pt), *labor law/conventions* (20.51 S-Pt), and *occupational toxicity & hazards* (18.50 S-Pt) are the most significant contributors to the overall risk assessment. The most affected damage categories are labor rights & decent work (163.45 S-Pt), community (100.82 S-Pt), and society (92.05 S-Pt), highlighting key social concerns within the harvesting process.

- OFC results

Table 3.8 presents the results for the OFC scenario. Among the different stages, boat maintenance emerges as the most socially impactful process. This is primarily due to the extended duration of maintenance activities, which became more significant during the three-month seaweed growth period. Similar to the WH scenario, the most critical social indicators include *injuries & fatalities* (0.0779 mrheq), *forced labor* (0.0681 mrheq), and *discrimination* (0.0622 mrheq). Fig. 3.21 provides a visualization of the normalized and weighted results, categorized according to each system stage.

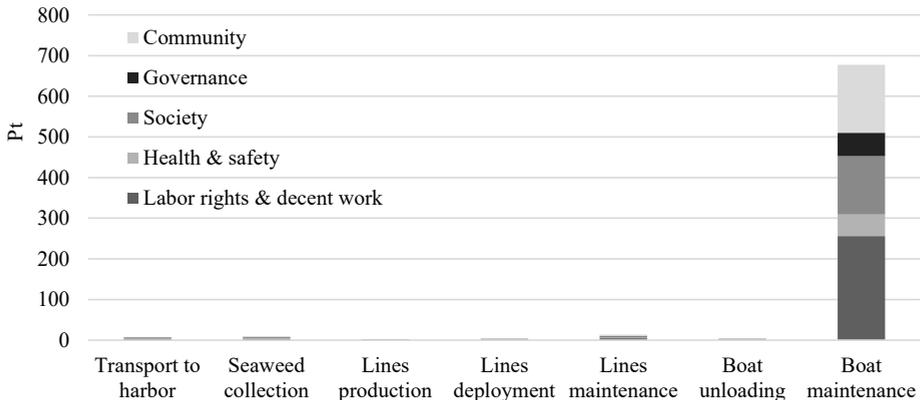


Fig. 3.21. Normalized and weighted results for the OFC scenario.

The individual indicators with the most significant impact in the OFC scenario are *access to electricity* (40.59 S-Pt), *labor law/conventions* (34.66 S-Pt), and *access to sanitation* (32.01 S-Pt). Within the boat maintenance stage, the most critical damage categories are labor rights & decent work (271.99 S-Pt), community (177.06 S-Pt), and society (152.58 S-Pt).

Table 3.8

Sub-category results for the OFC scenario

Impact category	Trans p.	Colle c.	Lines p.	Lines d.	Lines m.	Boat u.	Boat m.	Total	Unit
Wage assessment	0.0008	0.000	0.0001	0.0003	0.0009	0.000	0.0504	0.054	mrheq
Workers in poverty	5	66	3	0	0	27	5		
Child labor	0.0003	0.000	0.0001	0.0002	0.0006	0.000	0.0334	0.035	mrheq
Forced labor	8	50	0	2	7	17	0		
Excessive WkTime	0.0003	0.000	0.0001	0.0002	0.0007	0.000	0.0393	0.042	mrheq
Freedom of assoc	4	56	1	5	6	20	8		
Migrant labor	0.0008	0.000	0.0001	0.0004	0.0012	0.000	0.0639	0.069	mrheq
Social benefits	8	96	9	3	9	37	8		
Labor laws/conv	0.0006	0.000	0.0001	0.0003	0.0009	0.000	0.0478	0.051	mrheq
Discrimination	6	70	4	1	4	27	3		
Unemployment	0.0004	0.000	0.0001	0.0002	0.0007	0.000	0.0344	0.034	mrheq
Occ tox & haz	3	56	2	5	5	25	8		
Injuries & fatalities	0.0003	0.000	0.0000	0.0001	0.0005	0.000	0.0267	0.029	mrheq
Indigenous rights	5	43	9	9	8	16	9		
Gender equity	0.0002	0.000	0.0000	0.0001	0.0004	0.000	0.0210	0.022	mrheq
High conflict zones	0	31	7	4	2	14	6		
Non-Communicable diseases	0.0002	0.000	0.0000	0.0000	0.0002	0.000	0.0132	0.014	mrheq
Communicable diseases	2	17	3	8	3	06	4		
Poverty and inequality	0.0007	0.000	0.0001	0.0003	0.0011	0.000	0.0587	0.062	mrheq
State of env sustainability	0	82	6	7	1	32	3		
Legal system	0.0003	0.000	0.0000	0.0001	0.0004	0.000	0.0182	0.020	mrheq
Corruption	0.0005	0.000	0.0001	0.0003	0.0010	0.000	0.0529	0.056	mrheq
	2	79	7	6	7	33	2		
	0.0012	0.001	0.0002	0.0004	0.0014	0.000	0.0731	0.078	mrheq
	0	08	1	9	6	39	1		
	0.0001	0.000	0.0000	0.0001	0.0003	0.000	0.0159	0.017	mrheq
	3	24	5	1	2	09	2		
	0.0002	0.000	0.0000	0.0001	0.0004	0.000	0.0237	0.025	mrheq
	0	33	7	5	5	12	2		
	0.0003	0.000	0.0001	0.0001	0.0005	0.000	0.0260	0.028	mrheq
	9	42	0	9	7	18	7		
	0.0000	0.000	0.0000	0.0000	0.0001	0.000	0.0064	0.007	mrheq
	8	11	2	5	5	04	0		
	0.0001	0.000	0.0000	0.0001	0.0004	0.000	0.0233	0.025	mrheq
	8	33	7	5	4	12	6		
	0.0004	0.000	0.0001	0.0002	0.0007	0.000	0.0379	0.040	mrheq
	2	56	2	5	6	23	3		
	0.0004	0.000	0.0001	0.0003	0.0009	0.000	0.0486	0.051	mrheq
	1	69	4	1	3	27	8		
	0.0006	0.000	0.0001	0.0002	0.0007	0.000	0.0346	0.037	mrheq
	9	55	1	5	4	22	2		
	0.0003	0.000	0.0000	0.0001	0.0004	0.000	0.0176	0.019	mrheq
	3	30	6	3	0	11	3		

Impact category	Trans p.	Colle c.	Lines p.	Lines d.	Lines m.	Boat u.	Boat m.	Total	Unit
Democracy & freedom of speech	0.0011 0	0.000 77	0.0001 6	0.0003 4	0.0010 3	0.000 34	0.0507 1	0.054	mrheq
Access to drinking water	0.0002 1	0.000 35	0.0000 7	0.0001 6	0.0004 7	0.000 12	0.0235 1	0.024	mrheq
Access to sanitation	0.0003 4	0.000 58	0.0001 1	0.0002 6	0.0007 8	0.000 20	0.0420 0	0.044	mrheq
Children out of school	0.0003 3	0.000 41	0.0000 9	0.0001 8	0.0005 5	0.000 18	0.0264 0	0.028	mrheq
Access to hospital beds	0.0004 1	0.000 52	0.0001 0	0.0002 3	0.0007 0	0.000 17	0.0356 2	0.038	mrheq
Smallholder v commercial farms	0.0000 5	0.000 11	0.0000 4	0.0000 5	0.0001 5	0.000 09	0.0068 7	0.007	mrheq
Access to electricity	0.0000 8	0.000 24	0.0000 5	0.0001 1	0.0003 3	0.000 09	0.0176 6	0.019	mrheq
Property rights	0.0003 8	0.000 61	0.0001 2	0.0002 8	0.0008 3	0.000 21	0.0436 7	0.046	mrheq

Note: Transp. (Transport to harbor), collec. (Seaweed collection), Lines p. (Lines production), Lines d. (Lines deployment), Lines m. (Lines maintenance), Boat u. (Boat unloading), Boat m. (Boat maintenance)

- Comparison results with sensitivity analysis

Fig. 3.22 presents a comparative analysis of the two scenarios, highlighting that OFC exhibits a significantly higher potential social impact than WH. The total impact score for OFC reaches 719.51 S-Pt, whereas WH records a lower score of 432.85 S-Pt. This discrepancy suggests that OC is associated with greater social risks across key categories, including labor rights, community well-being, governance, and health and safety.

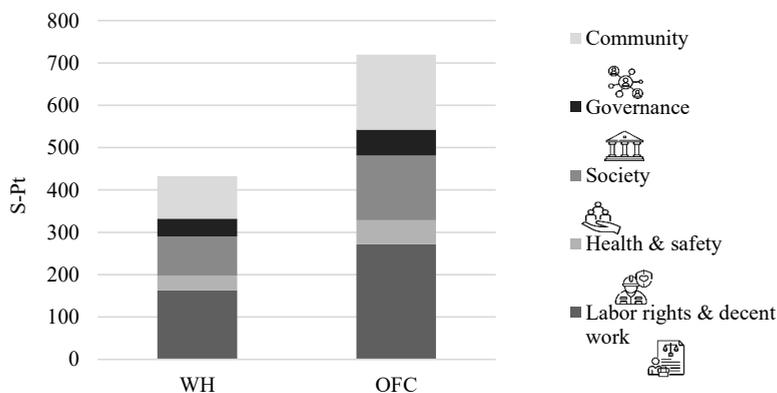


Fig. 3.22. Comparison results for the WH and OFC scenarios.

To enhance the reliability of these findings and evaluate their robustness, a Monte Carlo simulation was performed. This probabilistic analysis accounts for variability in key parameters (Table 2.25), offering a more comprehensive understanding of potential social impact fluctuations. The results of this simulation are illustrated in Fig. 3.23.

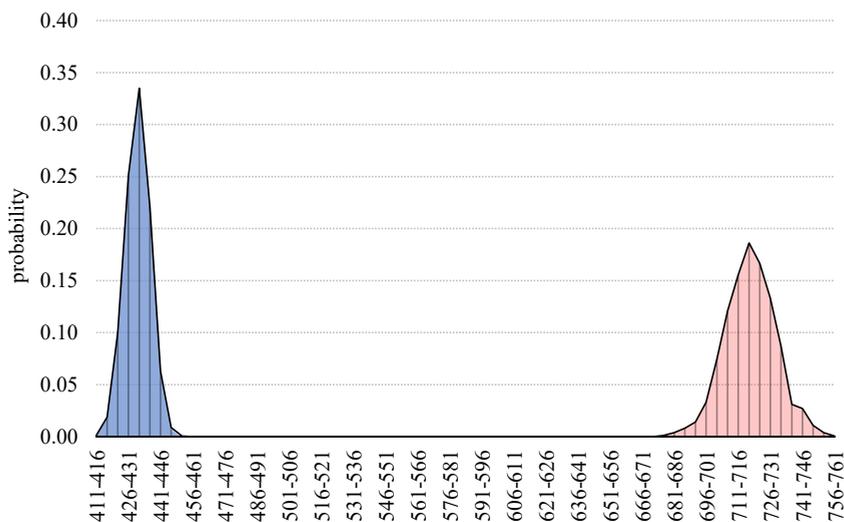


Fig. 3.23. Monte Carlo simulation results for the WH and OFC scenarios. The red chart is OFC, and the blue one is the WH.

The Monte Carlo simulation, conducted over 5,000 iterations, revealed distinct ranges of social impact values for each scenario. For WH, the impact varied between 411.5 S-Pt in the most favorable scenario and 454.5 S-Pt in the most unfavorable scenario. OFC exhibited a broader range, with values spanning from 678.1 S-Pt to 753.3 S-Pt. Notably, there is no overlap between these ranges, indicating a clear and consistent trend favoring WH over OFC in terms of lower social impact. This distinction strongly supports the conclusion that, based on the current assumptions and available data, OFC does not outperform WH from a social sustainability perspective. These findings reinforce the need to prioritize WH practices while further exploring ways to reduce the social risks associated with OFC.

- Take-home messages

This study encounters several limitations, primarily due to the challenges in applying the S-LCA framework to the macroalgae sector. The absence of standardized guidelines and limited case studies in this field creates obstacles in defining a clear methodological approach. Developing the assessment required overcoming various challenges, particularly in impact allocation. This study adopts a working-hour-based allocation to the FU, following methodologies suggested by Martinez-Blanco et al. (2014) [182] and Zamani et al. (2018) [186]. While this was deemed the most appropriate method given the available data, alternative approaches, such as monetary-based allocations [235], could yield different results, highlighting the sensitivity of outcomes to methodological choices.

A key limitation is the reliance on the SHDB, which uses national-level data to estimate social risks. This broad scope makes it difficult to capture localized impacts, such as those affecting small communities engaged in macroalgae harvesting or cultivation. Although the analysis provides useful insights through hotspot identification, it lacks the specificity needed to reflect the socio-economic realities of individual regions.

Additionally, the modeled systems introduce varying degrees of uncertainty. While the WH system is based on real-world data, the OFC system is hypothetical. This results in a higher degree of uncertainty when estimating social impacts for OFC, as certain processes and their consequences remain speculative. As more data becomes available, these models can be refined for greater accuracy.

Other uncertainties stem from assumptions and data inputs used in the study. Time allocations for activities like harvesting and maintenance, as well as the weighing of processes, are influenced by external variables such as weather conditions and operational variability. Sensitivity analyses, including Monte Carlo simulations, were conducted to mitigate these uncertainties by testing a range of potential outcomes. However, the results remain contingent on the initial assumptions, particularly in how data is normalized to the FU.

Moreover, the IPA S-LCA methodology is typically more applicable to well-established industries [185]. Its application to emerging fields like large-scale macroalgae cultivation presents challenges, as limited empirical data makes impact assessments more complex. Despite these limitations, these results provide a starting point for understanding the social dimensions of macroalgae production in the Baltic Sea Region, offering a framework for future research. Expanding datasets, refining allocation techniques, and integrating localized data will be essential for advancing S-LCA applications in this context.

Findings indicate that WH has lower social risks across all assessed categories, making it the more sustainable option under current conditions. Conversely, OFC, while innovative, presents higher social risks, mainly due to the extended maintenance activities required over its three-month cultivation period.

In the WH scenario, macroalgae harvesting was identified as the stage with the greatest social impact, with significant contributions from indicators such as *injuries & fatalities*, *forced labor*, and *discrimination*. The total normalized social impact score for WH was 432.85 Pt, with *labor rights & decent work* and *community* emerging as the dominant damage categories. WH also provides cultural and economic benefits for coastal communities, supporting community engagement and cultural preservation, though occupational safety remains an area needing improvement. Additionally, gaps in labor rights protections highlight the need for stronger regulatory oversight.

A notable observation in WH emerges from the SHDB analysis, which shows that the highest impact indicator, *access to electricity*, does not primarily affect Estonia but instead impacts supply chain countries such as the Ivory Coast and Ghana, see Fig. 3.24. Similarly, other indicators, such as *occupational toxicity & hazard*, show China as one of the most affected regions. This underscores the dispersed nature of social risks in global supply chains, demonstrating that social impacts can be generated far from the system's geographical location.

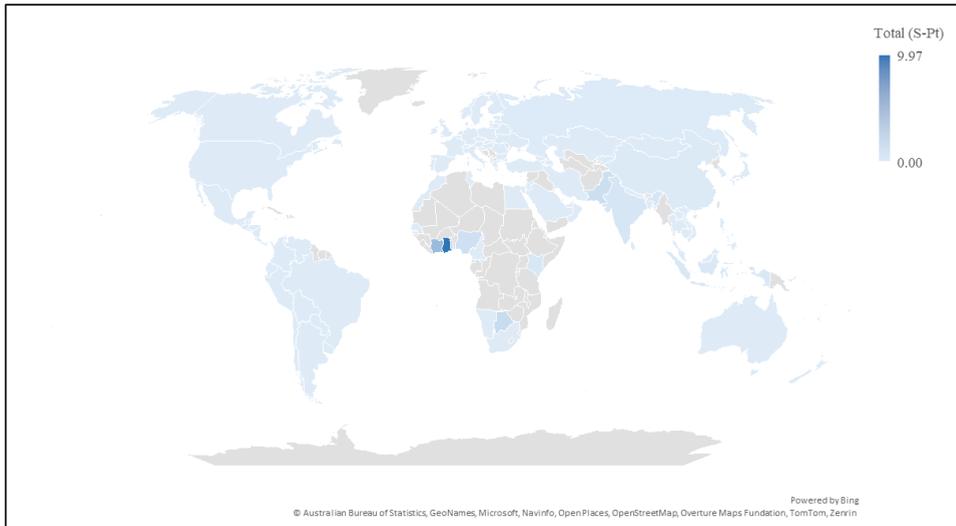


Fig. 3.24. Countries' hot spot analysis results for the normalized *Access to electricity* indicator.

In contrast, the OFC scenario had a higher total impact score of 719.51 S-Pt, mainly due to the extended maintenance and labor-intensive nature of line deployment and upkeep. Similar to WH, *injuries & fatalities* and *forced labor* were significant contributors, but OFC's more complex and prolonged operations amplified these risks. This highlights the challenges of industrialized systems in ensuring fair labor practices, reducing occupational hazards, and implementing strong governance structures.

The comparative analysis underscores the trade-offs between traditional and industrialized approaches to macroalgae production. WH remains the more socially sustainable choice within the study's assumptions, as it integrates well with existing socio-economic frameworks while presenting fewer risks. In contrast, OFC holds potential for scalability but requires substantial mitigation efforts to address its higher social risks.

To enhance WH sustainability, interventions should focus on improving occupational safety, particularly in reducing workplace injuries. Strengthening labor rights frameworks is also crucial to ensure fair wages and working conditions. For OFC, pilot studies are essential to refine operational parameters, reduce uncertainties, and validate key assumptions. Implementing training programs for cultivation workers can mitigate health and safety risks, while policy initiatives can promote sustainable industry practices.

Beyond these specific improvements, refining the S-LCA methodology is necessary to increase the accuracy of future studies. Expanding social indicator coverage to include community-specific factors would provide a more detailed understanding of socio-economic impacts. Conducting collaborative industry surveys could complement SHDB data, reducing reliance on broad proxy-based modeling and improving the representation of specific production systems.

Ultimately, these findings highlight the importance of balancing environmental and social considerations when developing macroalgae production systems. While WH remains the more socially sustainable choice under current conditions, advancements in OFC technology, governance, and operational frameworks could shift this balance, creating opportunities for more equitable and inclusive practices within the macroalgae industry.

Results of the RSA approach

The RSA approach provides a more targeted S-LCA by utilizing stakeholder-specific data gathered through questionnaires shared with 8 experts. These questionnaires were designed to assess the social risks associated with macroalgae harvesting and cultivation operations (upstream) as well as macroalgae processing operations (down-stream), based on the stakeholder groups and social indicators outlined in Table 2.26.

- Harvesting&cultivation social risk results

The results for the harvesting and cultivation phase are presented in Table 3.9, offering insights into the key social risks identified within these activities. The assessment captures various concerns, including labor conditions, occupational safety, and socio-economic impacts on workers and communities involved in the macroalgae sector. By integrating direct stakeholder input, this approach refines the evaluation of social sustainability, ensuring a more context-specific understanding of potential risks compared to broader database-driven assessments.

Table 3.9

Analysis of the social risk for the macroalgae harvesting&cultivation operations

Indicators	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 7	No. 8	Average
<i>Workers</i>									
Health and safety	0	2	-2	2	1	-2	-2	-2	-0.43
Working hours	0	2	2	2	-1	-2	-1	-1	0.14
Social benefits/social security	0	2	1	2	0	0	2	0	1.75
<i>Local community</i>									
Safe and healthy living conditions	0	2	-1	2	-2	-2	-1	2	0.00
Local economic development	2	2	2	2	2	2	2	2	2.00
<i>Consumers</i>									
Health and safety	0	0	-1	2	0	1	0	1	0.75
Transparency	0	1	2	2	-1	-2	0	2	0.67
<i>Value chain</i>									
Fair competition	0	0	-1	2	0	0	1	0	0.67
Promoting social responsibility	0	1	-1	1	0	0	0	0	0.33
Wealth distribution	0	-1	1	1	0	0	0	0	0.33

The analysis reveals notable concerns in specific areas, particularly regarding workers' *health and safety*, which received an average score of -0.43. This negative rating suggests a moderate level of risk, indicating that worker well-being is compromised and requires urgent attention.

On a more positive note, the local community's *economic development* emerged as a strong area, consistently scoring high with an average of 2.00. This indicates a very low

risk and confirms widespread agreement among experts on the significant economic benefits that macroalgae harvesting operations contribute to the local economy.

However, several other aspects present moderate concerns. *Working hours* received an average score of 0.14, reflecting inconsistencies that may require intervention to ensure standardized labor conditions. *Safe and healthy living conditions* within the local community had an average score of 0.00, indicating considerable variability, suggesting that while some communities are benefiting, others face notable challenges that need to be addressed.

Consumers' *health and safety* were assessed with a moderately positive average score of 0.75, implying a generally low risk but also highlighting room for improvement. *Transparency* received an average score of 0.67, indicating an overall low-to-moderate risk. However, the presence of negative responses suggests that transparency measures could be strengthened in certain areas.

The value chain was the stakeholder group with the highest number of null responses, suggesting a lack of expertise in this area, which could limit a comprehensive assessment. *Fair competition* was rated with an average score of 0.67, indicating a low overall risk, though some high-risk responses highlight the need to address disparities in market competition. Similarly, *promoting social responsibility* and *wealth distribution* each scored 0.33, signaling moderate risks and emphasizing the need for stronger efforts to enhance social responsibility practices and tackle economic inequalities.

A comprehensive overview of expert responses is visualized in Fig. 3.25, providing further insight into the social risk distribution across different categories.

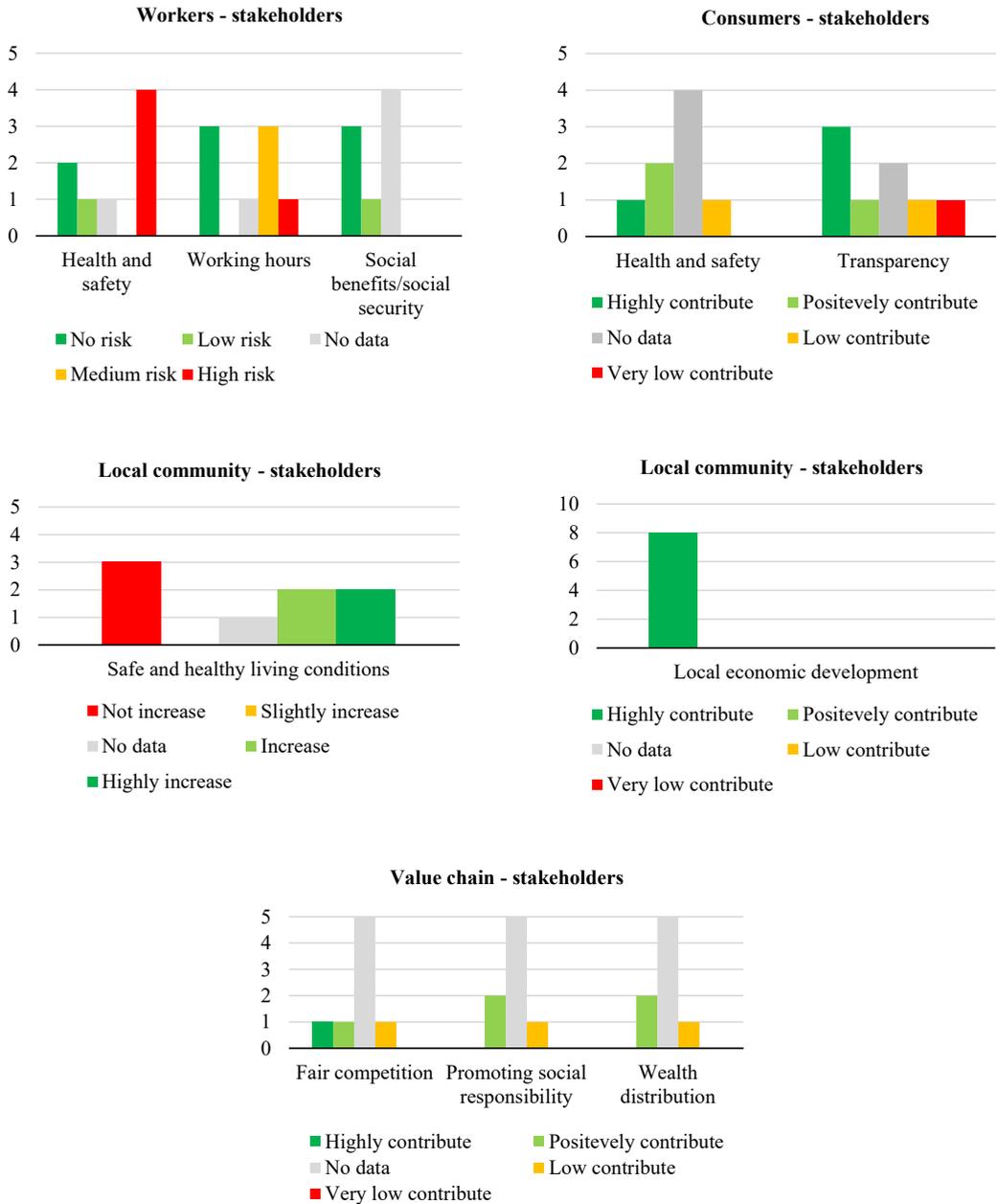


Fig.3.25. Summary of all experts' answers translated into a potential risk scale for the harvesting&cultivation operations.

- Macroalgae processing social risk results

Table 3.10 presents the results for the seaweed processing stage, highlighting key social indicators across various stakeholder categories.

Table 3.10

Analysis of the social risk for the macroalgae processing operations

Indicators	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 7	No. 8	Average
<i>Workers</i>									
Health and safety	0	2	-1	2	1	-1	-1	-1	0.14
Working hours	0	2	-1	2	-1	-1	-1	2	0.29
Social benefits/social security	0	2	1	2	0	0	2	0	1.75
<i>Local community</i>									
Safe and healthy living conditions	0	2	-1	2	0	-1	2	1	0.83
Local economic development	2	2	2	2	2	2	2	2	2.00
<i>Consumers</i>									
Health and safety	0	0	-1	2	0	1	0	1	0.75
Transparency	0	1	-2	2	0	-2	0	2	0.20
<i>Value chain</i>									
Fair competition	0	0	0	2	0	0	2	0	2.00
Promoting social responsibility	0	1	-2	1	0	0	0	0	0.00
Wealth distribution	0	-1	-2	1	0	0	0	0	-0.67

Within the workers category, the *health and safety* indicator received an average score of 0.14, indicating a moderate risk. While some areas demonstrate satisfactory conditions, concerns remain that require attention to improve the workers' well-being. However, this score is notably lower compared to the macroalgae harvesting stage, suggesting that harvesting poses a greater occupational hazard. The *working hours* indicator scored an average of 0.29, reflecting similar moderate risks, with inconsistencies in work schedules that may require corrective actions to ensure standardization across the industry. In contrast, the *social benefits/social security* indicator achieved a high positive average of 1.75, signifying low risk and suggesting that workers generally have reliable access to these benefits.

In the local community category, the *safe and healthy living conditions* indicator received an average score of 0.83, reflecting a lower level of risk and generally satisfactory conditions, though some areas may still require further attention. Meanwhile, the *local economic development* indicator remained consistently high at 2.00, indicating very low risk and robust economic contributions from the seaweed processing industry.

For consumers, the *health and safety* indicator recorded a positive average score of 0.75, signifying a low risk in this category. However, the presence of a negative score highlights the need for ongoing improvements to maintain high safety standards. The *transparency* indicator had an average score of 0.20, reflecting a moderate risk. Negative responses suggest that transparency practices could be significantly improved to strengthen consumer trust and confidence in the sector.

In the value chain category, the *fair competition* indicator received a strong positive score of 2.00, indicating minimal risk and a well-functioning competitive market. However, the *promoting social responsibility* indicator scored 0.00, suggesting a neutral risk level.

While some efforts to encourage social responsibility exist, they are not widespread, indicating room for improvement. The *wealth distribution* indicator, however, recorded a negative average score of -0.67, pointing to a moderate to high risk. This suggests that economic disparities remain significant, requiring targeted measures to promote more equitable wealth distribution within the industry.

A comprehensive breakdown of expert responses is visualized in Fig. 3.26, providing deeper insights into the distribution of social risks across these key indicators.

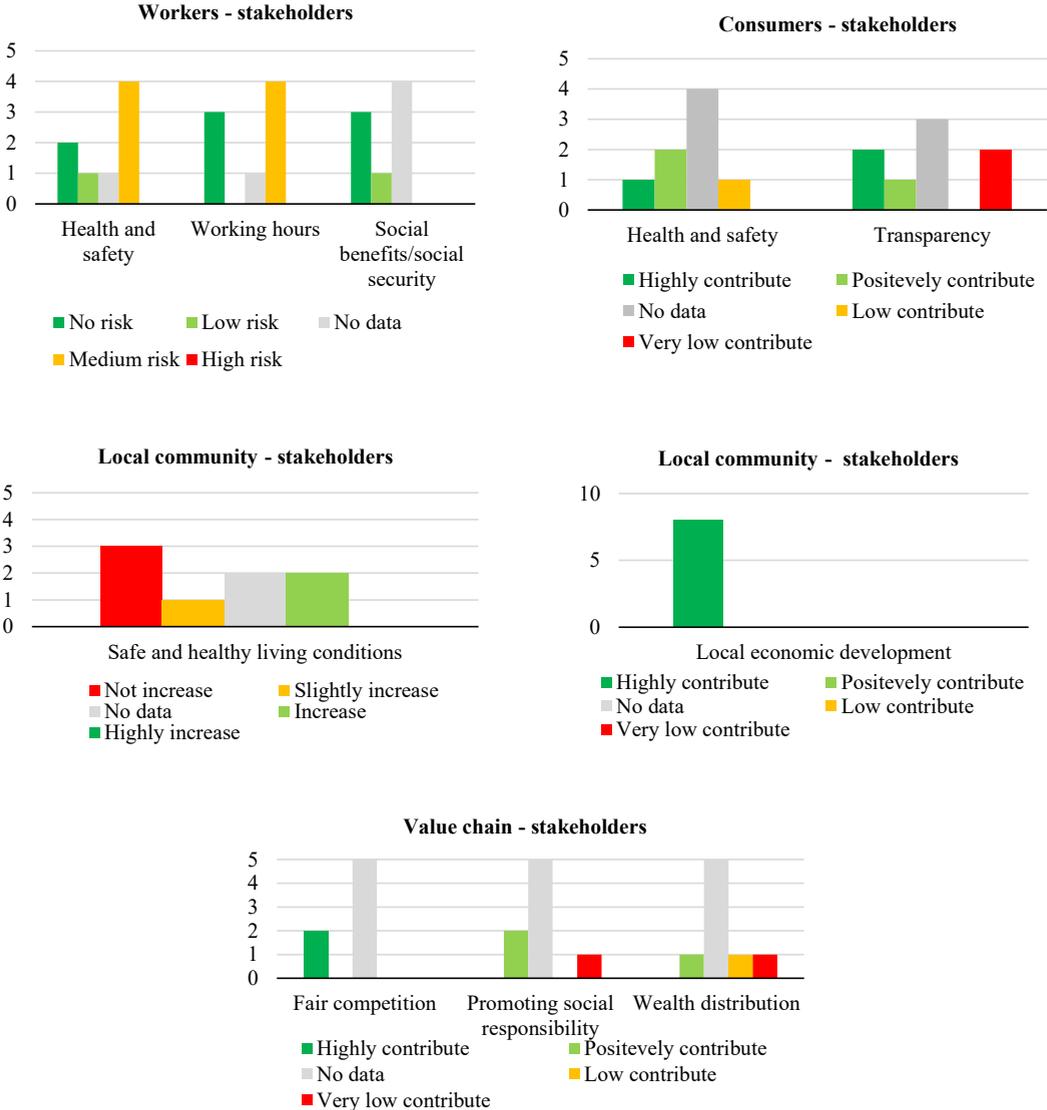


Fig. 3.26. Summary of all experts' answers translated into a potential risk scale for the macroalgae processing operations.

- Take-home messages

Based on the findings, experts widely agreed on the absence of risk for the *local economic development* indicator, consistently assigning it a score of +2 for both the harvesting & cultivation and processing phases. This positive outcome is attributed to improvements in infrastructure, such as additional services supporting macroalgae-related operations, and the potential for increased tourism interest, as these activities may attract visitors. Furthermore, seaweed-related employment can sustain economic activities during off-seasons when harvesting is not possible. In small, remote locations like islands, jobs in the macroalgae sector could be among the few available employment opportunities, reinforcing the economic importance of the industry. A similar trend was observed for the *social benefits/social security* indicator, which received an average score of +1.75. However, only 50 % of the surveyed experts provided responses for this indicator, introducing some uncertainty. The seasonal nature of macroalgae harvesting raises concerns about disparities in worker benefits, as seasonal employees may not receive the same social protections as full-time workers, potentially leading to inequities in worker rights.

For the fair competition indicator, results suggest no risk in the processing phase, whereas a low level of risk was identified during the biomass preparation stage. Moderate risks were identified in two key areas: worker *health and safety* during the cultivation and harvesting phase (-0.43) and *wealth distribution* in the processing phase (-0.67). Occupational risks in harvesting and cultivation primarily stem from offshore operations, where unpredictable weather conditions and machinery failures pose significant hazards. Additionally, macroalgae harvesting is performed within short operational windows, requiring intense labor efforts that increase workplace injury risks. The physically demanding nature of the work also raises concerns about long-term health impacts, particularly musculoskeletal disorders. Implementing electric loaders and other mechanized equipment could reduce these risks by minimizing manual labor. In processing operations, potential hazards include exposure to cutting and milling machinery, which requires specialized handling and training. Chemical exposure during macroalgae processing further underscores the need for safety measures and protective equipment.

Regarding *wealth distribution*, findings suggest an imbalance in the allocation of economic benefits within the supply chain, raising concerns that financial gains from the biorefinery process may not be equitably shared among stakeholders.

For the *safe and healthy living conditions* indicator, macroalgae-related activities are perceived as posing lower risks than aquaculture operations, such as fish and mussel farming, which can contribute to nutrient accumulation, storm-related damages, and ecological disturbances, including impacts on duck populations. However, intensive harvesting using diesel-powered boats can introduce oil and polycyclic aromatic hydrocarbon emissions, negatively affecting local marine ecosystems and communities dependent on these resources. The use of antifouling agents on boats also presents a risk of leaching, potentially causing significant environmental harm.

The *transparency* indicator for consumers revealed concerns about misleading or unverified claims regarding the climate benefits of macroalgae biomass. These claims are

often used for marketing purposes without scientific backing, leading to potential misinformation. To mitigate this risk, publicly accessible and scientifically validated composition analyses should be provided to ensure consumers receive accurate, evidence-based information.

A major challenge in conducting this S-LCA was data collection. Difficulties encountered were both specific to this study and common in S-LCA research. Standard S-LCA methodologies, which rely on international databases such as SHDB and PSILCA, struggle to capture the full social dynamics of an *F. lumbricalis* biorefinery, as reported in the previous sub-chapter, particularly given the small community scale of operations. To address this limitation, a stakeholder engagement approach was pursued through questionnaires. However, participation was limited, with only eight responses out of more than 20 experts contacted. Despite the small sample size, these responses provided valuable insights and represented an important first step toward improving the understanding of social sustainability in the macroalgae sector.

One key limitation identified in the social analysis was the lack of data on "other value chain actors." The high proportion of missing responses for this stakeholder group suggests that future studies should focus more on engaging these actors. Social indicators such as *fair competition*, *promotion of social responsibility*, and *wealth distribution* remain underexplored, highlighting the need for further efforts to bridge these gaps. A more engaged and willing-to-participate expert community is critical for improving the reliability and depth of social sustainability assessments in this field. Nonetheless, the preliminary social mapping conducted in this study has provided valuable insights and serves as a foundation for future research in this area.

3.4. Final sustainability index

The final sustainability index in the context of LCSA has been determined using the TOPSIS method, whose framework is outlined in Chapter 2.5. This approach integrates results from LCA, LCC, and S-LCA presented in previous chapters to identify the most sustainable biorefinery design among CB, SPE, and TLE. By applying TOPSIS, the analysis ranks the biorefinery configurations based on their relative closeness to an ideal sustainable solution while maximizing environmental, economic, and social performance.

TOPSIS results

In the first step of the TOPSIS approach, each selected indicator within the three sustainability dimensions, environmental, economic, and social, was classified based on whether it should be maximized or minimized relative to an ideal sustainability scenario. This classification is crucial to ensure a consistent and systematic assessment of sustainability performance across all criteria. Indicators related to environmental impact were categorized as criteria to be minimized, as lower values indicate better sustainability outcomes. Conversely, economic indicators such as NPV and ROI were set to be maximized, reflecting higher profitability as a more favorable outcome. For the social

dimension, indicators were classified as maximization criteria, as in the RF scales, the +2 was the value of no risk. The details of this classification process are systematically outlined in Table 3.11, together with the initial weight assigned, ensuring a transparent evaluation methodology that allows for an objective ranking of the biorefinery designs.

Table 3.11

Indicators and criteria decision. MIN and MAX indicate if each has been minimized or maximized, respectively, in comparison to an ideal scenario

Dimension	Indicator	Unit of measure	Type	MIN/MAX	Weights
Environmental	Global warming potential	kg CO ₂ eq.	Quantitative	MIN	0.083
	Fine particulate matter formation	kg PM _{2.5}	Quantitative	MIN	0.083
	Human carcinogenic and non-carcinogenic toxicity	kg 1.4-DCB	Quantitative	MIN	0.083
	Water consumption	m ³	Quantitative	MIN	0.083
Economic	Return of investment	%	Quantitative	MAX	0.083
	Net present value	EUR	Quantitative	MAX	0.083
	Internal rate of return	%	Quantitative	MAX	0.083
	Profitability index	-	Quantitative	MAX	0.083
Social	Health & safety of the worker	-	Semi-quantitative	MAX	0.083
	Working hours	-	Semi-quantitative	MAX	0.083
	Local economy development	-	Semi-quantitative	MAX	0.083
	Wealth distribution	-	Semi-quantitative	MAX	0.083

The normalization matrix standardizes the values of each indicator, ensuring comparability across different units and scales. This step is essential in the TOPSIS method, as it transforms the raw data into a uniform format, allowing for a consistent evaluation of sustainability performance among the three scenarios (CB, SPE, and TLE). The normalized values in Table 3.12 provide a balanced representation of each criterion, ensuring that the impact of different sustainability indicators is appropriately weighted in the final assessment.

Table 3.12

Normalized matrix for the assessment indicators and the different biorefinery designs

Dimension	Indicator	Unit of measure	Scenarios		
			CB	SPE	TLE
Environmental	GWP	kg CO ₂ eq.	0.68	0.69	0.23
	PM	kg PM _{2.5}	0.54	0.80	0.27
	HCCT	kg 1.4-DCB	0.49	0.83	0.28
	WC	m ³	0.74	0.63	0.21
Economic	ROI	%	1.00	-0.07	-0.09
	NPV	EUR	0.99	-0.09	-0.05
	IRR	%	0.78	0.62	0.00

Dimension	Indicator	Unit of measure	Scenarios		
			CB	SPE	TLE
Social	PI	-	0.99	-0.09	-0.05
	HS	-	0.51	-0.69	0.51
	WH	-	0.58	-0.58	0.58
	LED	-	0.69	0.69	0.23
	WD	-	-0.52	-0.52	-0.69

The weighted normalization matrix was finalized, and the Euclidean distance to both the optimal and least favorable scenarios was computed to identify the most sustainable biorefinery design among the three proposed configurations. Fig. 3.27 visually depicts the results, showcasing the comparative sustainability performance of CB, SPE, and TLE.

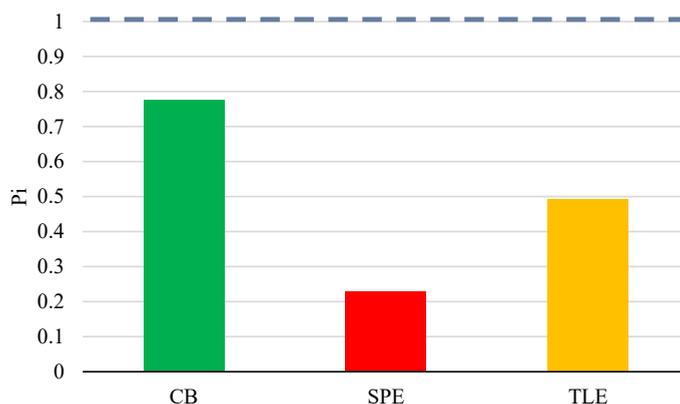


Fig. 3.27. Results of the LCSA following the TOPSIS approach. The blue line represents the ideal scenario benchmark.

The CB design was identified as the most sustainable configuration for the *F. lumbricalis* biorefinery, achieving a Pi value of 0.776, positioning it closest to the ideal scenario ($P_i = 1$). TLE followed with a Pi value of 0.492, representing an intermediate option, while SPE, with a Pi value of 0.229, ranked as the least sustainable design, further highlighting the constraints of a single-product approach.

Sensitivity analysis results

To ensure the reliability of the findings, a sensitivity analysis was conducted by adjusting the weight assigned to each of the three sustainability dimensions (i.e., environmental, economic, and social). This analysis assesses how variations in weight distribution influence the overall ranking of the biorefinery designs. The outcomes of this evaluation are illustrated in Fig. 3.28.

Even when accounting for the sensitivity analysis, CB consistently emerges as the most sustainable design. Across 15 simulations, it ranked as the most favorable scenario in 80% of cases. The environmental dimension plays a crucial role in influencing the ranking, as TLE surpasses CB in two instances when environmental sustainability is given the highest

weight. This underscores the need for increased focus on environmental factors in the future development of *F. lumbricalis* biorefineries.

From an economic standpoint, the CB proves advantageous, as the ability to generate multiple products strengthens market positioning and enhances economic viability. Conversely, TLE struggles in this area. Due to the initial biomass fractionation, TLE cannot achieve the same product yield extraction as CB, limiting its financial feasibility. Notably, when the economic dimension receives the highest weighting, TLE falls below SPE, further highlighting its constraints.

Regarding social sustainability, no scenario crossovers were observed, likely due to the semi-quantitative nature of the input values, which restrict differentiation in social performance.

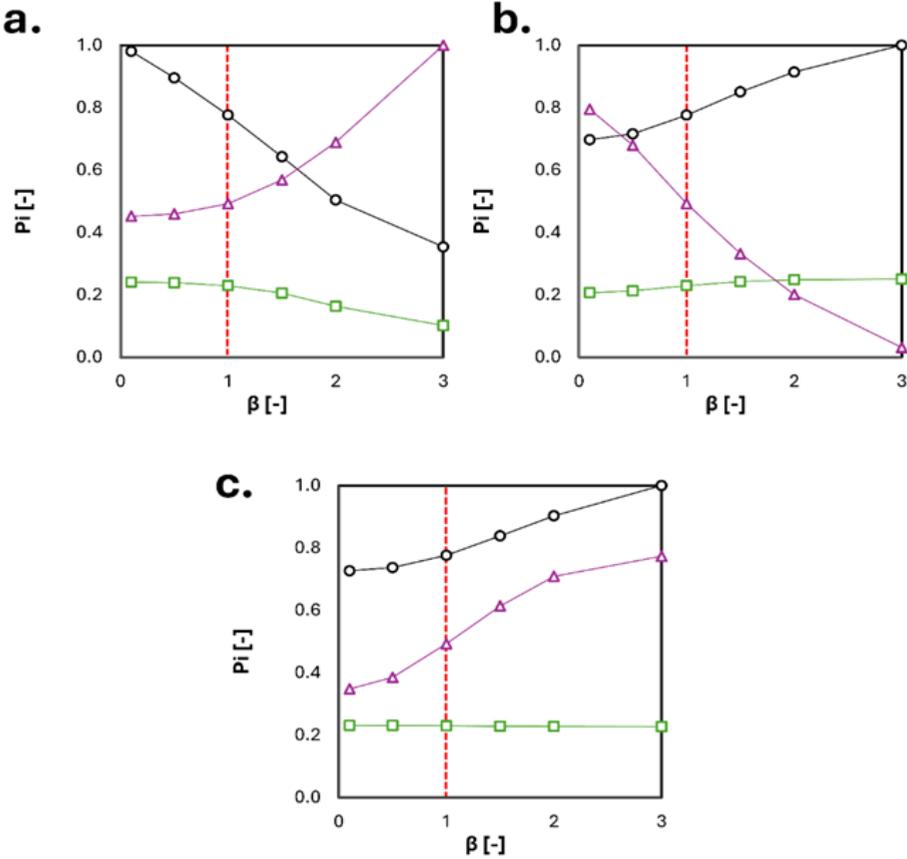


Fig. 3.28. Results of the sensitivity analysis for weights to (a) environmental, (b) economic, and (c) social criteria. Closeness coefficient (P_i) as a function of the variation ratio (β). The black line represents the CB, the violet line SPE, and the green line TLE. The Red dashed line indicates the results of the LCSA before the sensitivity analysis.

Table 3.13 summarizes the probability distribution of rankings for each scenario based on the sensitivity analysis simulations.

Table 3.13

Probability ranking of the different design approaches

Design	Probability ranking		
	Best scenario	Mid scenario	Worst scenario
CB	80 %	20 %	0 %
SPE	0 %	47 %	53 %
TLE	20 %	33 %	47 %

Take-home messages

The integration of the three sustainability pillars within the LCSA framework using the TOPSIS methodology has proven to be an effective approach, enabling the development of a comprehensive sustainability index. Future research should further refine this method to ensure well-balanced assessments across environmental, economic, and social dimensions. While the CB design shows a slightly higher environmental impact than TLE, its overall sustainability performance is superior when considering economic and social factors. The marginal increase in environmental burden is offset by significant gains in the other two dimensions, reinforcing that a circular, multi-product biorefinery model is the most viable strategy for fostering a sustainable blue bioeconomy based on *F. lumbricalis*. Conversely, an SPE approach leads to substantial trade-offs across all sustainability metrics. Although the development of a multi-product system requires more resources and time, the long-term sustainability benefits strongly support its adoption.

A limitation of this study is the selection of the TOPSIS method as the multi-criteria decision-making approach, despite the availability of alternative methodologies. While TOPSIS was deemed appropriate due to its distance-based ranking capabilities, other approaches such as value/utility functions (e.g., MAVT, MAUT, SAW), pairwise comparison methods (e.g., AHP, ANP), or outranking techniques (e.g., PROMETHEE, ELECTRE) could potentially yield different rankings. Future research should explore these alternative decision-support tools to assess how methodological choices impact sustainability evaluations. Additionally, the exploratory nature of this study presents certain limitations, as the LCA, LCC, and S-LCA results are based on a pre-industrial system where the final technological configurations are not yet fully established. As a result, the reliance on a combination of primary data, literature sources, and proxies may introduce uncertainties. Further studies should aim to refine data accuracy as the biorefinery concept progresses toward industrial-scale implementation.

Despite these constraints, this Thesis marks an important step toward the sustainable valorization of *F. lumbricalis*, demonstrating the importance of an integrated, circular biorefinery model. The findings provide a robust foundation for future advancements in the macroalgae sector and highlight the necessity of adopting multi-product strategies to achieve long-term sustainability in the emerging blue bioeconomy.

CONCLUSIONS AND RECOMMENDATIONS

Conclusions

This Doctoral Thesis presented a novel approach for the development and assessment of a cascade biorefinery system for the red macroalga *F. lumbricalis* in the Baltic Sea Region, underpinned by an LCSA framework. By integrating LCA, LCC, and S-LCA, the study provides a holistic evaluation of sustainability across three distinct biorefinery designs: CB, SPE, and TLE.

A general conclusion, the findings demonstrate that integrating environmental, economic, and social dimensions through the LCSA approach represents a robust and comprehensive strategy for informing industrial planning and policy-making in the bioeconomy sector. As one of the first complete LCSA applications for macroalgae biorefineries built upon primary data, the study confirms that a multi-product cascade configuration is a promising and viable pathway for supporting the sustainable development of the blue bioeconomy. This research offers a sound basis for future investigations and provides practical guidance for the industrial implementation of macroalgae-based biorefineries, identifying critical parameters and technological considerations to enhance sustainability performance.

More specific conclusions are drawn below:

- State-of-the-art review. The literature analysis revealed that macroalgae biorefinery studies remain predominantly at the experimental stage. Brown macroalgae dominate existing research, while red macroalgae, including *F. lumbricalis*, remain significantly understudied, particularly in LCA and S-LCA contexts. This highlights a critical research gap addressed by the present Thesis.
- Technological Development of Cascade Biorefinery. A complete cascade biorefinery model was developed for *F. lumbricalis*, combining pigment, protein, and carrageenan extraction. The model maximizes biomass valorization and introduces optional residual biomass treatment through either fertilizer production or biogas generation. Compared to conventional SPE and TLE designs, the cascade model achieves significantly higher resource efficiency and output yield from a fixed biomass input.
- Critical role of process design. A complementary assessment of three pigment extraction strategies, EAE, UAE, and water extraction, demonstrated the critical role of process design in overall sustainability. EAE showed the best trade-off between productivity, economic feasibility, and environmental performance, achieving a favorable Green Index and the lowest waste-to-product ratio. Despite its higher energy demands, the reuse of solvents and higher yield positioned it as the most sustainable option. In contrast, the UAE, while avoiding chemical inputs, suffered from high electricity consumption, and water extraction was hindered by poor extraction yield, leading to environmental and economic inefficiency. These findings reinforce the importance of optimizing both yield and energy profile in future cascade biorefinery developments.

- Environmental analysis of the upstream processes. The LCA assessed three biomass sourcing strategies (i.e., WH, OFC, and ONC) to evaluate the environmental footprint and the most sustainable operation. Among them, ONC showed the highest impact (56.21 mPt), followed by OFC (22.76 mPt) and WH (11.10 mPt). Alternative scenarios revealed several improvement opportunities. Replacing LED lighting with natural sunlight in the ONC system reduced environmental impacts by 78.25 %, while adding solar panels had a smaller benefit. For WH, the antifouling agent was the most critical parameter; switching to an eco-friendly alternative led to a 28.02 % reduction, lowering the total impact to 7.99 mPt, the best result among all systems. In contrast, no significant improvements were observed for OFC under the tested alternatives. These findings provide practical guidance for selecting low-impact biomass acquisition strategies and suggest that further integrating cultivation or harvesting systems into biorefinery designs could enhance overall sustainability.
- Environmental analysis of the full biorefinery system. The LCA of the complete cascade biorefinery system for *F. lumbricalis* in the Baltic Sea Region quantified the impacts associated with the production of pigments, proteins, and carrageenan. Two Functional Units (FUs) were applied to evaluate system performance. Under FU1, which captures the full process, the total environmental impact reached 140.08 Pt when macroalgae residues were repurposed as fertilizer and 153.22 Pt when utilized for biogas production. Carrageenan extraction was identified as the most critical stage (63.28 Pt), mainly due to its significant energy requirements, underscoring the need for targeted optimization and the adoption of more energy-efficient technologies. Reuse of residues as biostimulant fertilizers was found to be the more favorable end-of-life option, offering an avoided impact of -8.82 Pt. Moreover, the integration of a drying step, as tested in the alternative scenarios, proved to have a notable influence on overall sustainability and should, therefore, be carefully considered in future system designs. Under FU2, which allows comparison between different process configurations, the CB showed superior performance over the SPE, which resulted in the highest impact at 144.60 Pt. Interestingly, the TLE system, when coupled with commercially available product substitutes, achieved the lowest impact at 78.39 Pt. While TLE is less efficient in biomass utilization, this hybrid model may represent a promising and pragmatic pathway for sustainable market development.
- Economic sustainability. The CB scenario demonstrated the strongest performance, driven by product diversification and full biomass valorization. Particularly under favorable market conditions, its economic viability was closely linked to the high market value of pigments. However, the CB system also presented high operational and capital costs, highlighting the need for improved process efficiency and strategies to manage upfront investment burdens. In contrast, the SPE and TLE systems showed limited profitability due to lower product yields and less efficient resource use, making them less competitive. The

analysis also identified several critical factors affecting economic sustainability, such as the dependence on high-value compounds, raw material and logistics costs, and exposure to market volatility. Integrating monetized environmental externalities into the assessment offered a more comprehensive perspective, reinforcing the CB's dual environmental and economic advantage.

While the result confirms the potential of *F. lumbricalis* biorefineries to contribute to a sustainable bioeconomy, their practical realization will depend on overcoming challenges related to production costs, market dynamics, and technological scalability. Targeted actions, including public funding, process optimization, and the adoption of innovative extraction methods, will be key to enhancing cost-effectiveness and accelerating market integration.

- Social analysis with the IPA approach on upstream processes. The results confirm that WH is presently the more socially sustainable option, with lower risks across all evaluated categories. WH aligns well with existing socio-economic structures, supporting local livelihoods and cultural continuity. However, occupational safety remains a key concern and requires focused attention to improve working conditions. In contrast, OFC shows higher social risks due to its labor-intensive nature and extended maintenance demands. For OFC to become a socially viable alternative, significant advancements in technology, labor practices, and governance frameworks are needed. Looking ahead, improving the robustness of S-LCA for macroalgae systems will require more localized and stakeholder-informed data. Future research should prioritize direct engagement with communities and pilot-scale implementations of OC systems to refine assessments and better understand context-specific social dynamics. Enhancing the depth and coverage of social indicators will be essential to support more accurate and comprehensive evaluations of social sustainability in macroalgae production.
- Social analysis with the RFS approach on full biorefinery processes. The S-LCA revealed that the cascade biorefinery approach for *F. lumbricalis* offers meaningful social benefits, particularly in supporting local economic development and employment. However, the analysis also identified key social risks, including occupational *health and safety* concerns in harvesting and processing stages, and imbalances in wealth distribution across the value chain. The expert-based evaluation highlighted limited stakeholder awareness and data availability, especially regarding “other value chain actors,” underscoring the need for broader engagement and more granular data collection in future studies. Despite these limitations, the cascade model emerged as the socially most favorable option among the assessed configurations, reinforcing the importance of integrating social dimensions into sustainability assessments. Continued development of socially responsible practices, transparent communication, and equitable value sharing will be essential for the successful and inclusive implementation of macroalgae-based biorefineries.

- Sustainability index. The TOPSIS comparison of the CB with the alternative SPE and TLE systems, integrating LCA, LCC, and S-LCA results, clearly demonstrates that the CB model offers the most balanced sustainability performance. It improves resource efficiency, enhances economic viability, and delivers positive social outcomes, particularly in job creation and support for local economies.

Recommendations

- **Pilot and Scale-Up:** Translating the CB concept from theoretical modeling and lab-scale tests to industrial-scale operations is a critical next step. Pilot projects should aim to validate process yields, equipment reliability, and scalability under real-world conditions. These initiatives will also provide insight into supply chain logistics, workforce needs, and site-specific variables that influence system performance. Collaboration between academic institutions, technology developers, and industry stakeholders will be essential for successful scale-up.
- **Energy Optimization:** Environmental and economic assessments identified energy-intensive operations, particularly drying and heating steps, as major hotspots. Investing in energy-efficient technologies, such as low-temperature drying systems, waste heat recovery, and advanced process integration, can significantly reduce impacts. Furthermore, integrating renewable energy sources, such as solar or bio-based electricity, is strongly recommended to improve overall sustainability and align with carbon neutrality goals.
- **Market Strategy:** Enhancing the marketability of macroalgae-derived products is crucial for economic viability. Efforts should focus on improving product purity, safety, and regulatory compliance to access high-value markets such as food, cosmetics, pharmaceuticals, and agriculture. Diversifying the product portfolio (e.g., expanding into nutraceuticals or bio-stimulants), can buffer against market volatility and foster resilience. Co-development of product standards and certification schemes would support consumer trust and market growth.
- **Data Improvement in S-LCA:** S-LCA assessment in the macroalgae sector is still in its infancy. Future research should work toward improving the robustness and representativeness of social impact data by incorporating site-specific, community-level, and gender-sensitive indicators. Expanding stakeholder participation through interviews, co-design workshops, and citizen science initiatives can enhance data quality and promote social inclusiveness in biorefinery development.
- **Policy Incentives:** To unlock the potential of macroalgae biorefineries, enabling policies are needed. Governments and EU institutions should introduce targeted funding programs, tax incentives, and fast-track permitting schemes that lower barriers to entry for sustainable macroalgae valorization projects. Developing clear regulatory pathways for novel macroalgae-based products, including food-grade and pharmaceutical applications, will further stimulate investment and innovation.
- **Extension to Other Macroalgae:** The LCSA framework applied in this study should be replicated for other macroalgae species and geographical regions to validate its

robustness and transferability. Comparing different biomass types, cultivation systems, and local socio-economic contexts can generate best practices for the broader application of sustainable biorefinery models across the blue bioeconomy sector.

- **Circular Integration with Other Sectors:** There is significant potential to enhance sustainability by integrating macroalgae biorefineries with other circular economy sectors, such as aquaculture, wastewater treatment, or agriculture. For instance, co-locating biorefineries with fish farms or greenhouse operations could enable nutrient recovery, energy sharing, and cross-sector synergies that lower costs and environmental burdens.
- **Capacity Building and Education:** Investments in workforce development, vocational training, and educational outreach will be essential to support the growing macroalgae sector. Establishing interdisciplinary training programs and knowledge-sharing platforms can build the human capital needed for sustainable biorefinery operations and foster innovation across the value chain.
- **Comparative Assessment with Crop-Based Biorefineries:** To further substantiate the findings, comparative studies should be carried out between seaweed-based and crop-based biorefineries. These comparisons would enhance the strength of the sustainability narrative and help position macroalgae as a viable alternative in the bioeconomy landscape.
- **Critical Final Remark - The Marked Demand Analysis:** The most urgent recommendation emerging from this work is the need for a detailed market-demand analysis of the CB-derived products. Although this study confirms the technical and sustainability validity of the cascade biorefinery approach, its real-world viability depends entirely on whether there is sufficient market demand. If the final products are not needed, even the most sustainable system becomes economically and socially unfeasible. Therefore, future studies must shift from theoretical modeling to applied market scenarios, stakeholder consultations, and policy forecasting to gauge the actual necessity and competitiveness of the system.

This Thesis demonstrates that the development of a tailored cascade biorefinery for *F. lumbricalis* can be environmentally sound, economically viable, and socially responsible. The LCSA approach provides a powerful decision-support tool that can bridge the gap between laboratory innovation and real-world sustainable implementation.

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ANNEXES

ANNEX 1.

Questionnaire 1 for the RSA S-LCA approach.



Riga Technical University
Institute of Energy System and Environment

Questionnaire for

SOCIAL LIFE CYCLE ASSESSMENT FRAMEWORK

FOR THE TACO ALGAE PROJECT

1. INTRODUCTION: What is the Social Life Cycle Assessment?

The Social Life Cycle Assessment (S-LCA) is one of three methodologies that have been developed to assess the sustainability of the three Pillars of organizations, products, and services, focusing on the People Pillar. Thus, S-LCA is the framework to assess the social impacts of products and services across their life cycle. The S-LCA aims to assess social consequences (positive or negative) on different stakeholder categories:



Workers, the local community, society, consumers, value chain actors, and children are the main stakeholder categories reported in the UNEP "Guidelines for Social Life Cycle Assessment." However, based on system characteristics, it is also possible to expand this list to include more specific stakeholder categories.

The social effects of the stakeholders can be measured on impact categories:



These impact categories summarized the list of social indicators based on the stakeholders:

- | | |
|---|--|
| <ul style="list-style-type: none"> - Worker 1. Freedom of association and collective bargaining 2. Child labor 3. Fair salary 4. Working hours 5. Forced labor 6. Equal opportunities/discrimination 7. Health and safety 8. Social benefits/social security 9. Employment relationship 10. Sexual harassment 11. Smallholders, including farmers | <ul style="list-style-type: none"> - Local community 1. Access to material resources 2. Access to immaterial resources 3. Delocalization and migration 4. Cultural heritage 5. Safe and healthy living conditions 6. Respect for indigenous rights 7. Community engagement 8. Local employment 9. Secure living conditions |
|---|--|

- **Society**

1. Public commitment to sustainability issues
2. Contribution to economic development
3. Prevention and mitigation of armed conflict
4. Technology development
5. Corruption
6. Ethical treatment of animals
7. Poverty alleviation

- **Value chain actors (not including consumers)**

1. Fair competition
2. Promoting social responsibility
3. Supplier relationship
4. Respect for intellectual property rights
5. Wealth distribution

- **Consumer**

1. Health and safety
2. Feedback mechanism
3. Consumer privacy
4. Transparency
5. End-of-life responsibility

2. INSTRUCTION FOR THE QUESTIONNAIRE COMPILATION

We would like to ask for your cooperation in filling out the following questionnaire.

The goal is to acquire opinions and information about potential social impact in the case of a seaweed biorefinery. We would be asked:

- To select which are the most representative type of stakeholder based on your experience and opinion;
- Select the most representative impact categories for each of the stakeholders.

3. THE QUESTIONNAIRE

1- Please select, based on opinion and expertise, which are the most representative **stakeholders** (you can select more than 1) for a seaweed biorefinery system from the list below:

- Worker
- Local community
- Society
- Consumers
- Value chain actors
- Children
- Others

Please, if you select others, specify here what:

2- Please select, based on opinion and expertise, which are the most representative **social indicators** (you can select more than 1) for the **Worker** stakeholder:

- Freedom of association and collective bargaining
- Child labor
- Fair salary
- Working hours
- Forced labor
- Equal opportunities/discrimination
- Health and safety
- Social benefits/social security
- Employment relationship
- Sexual harassment
- Smallholders including farmers
- Others

Please, if you select others, specify here what:

3- Please select, based on opinion and expertise, which are the most representative **social indicators** (you can select more than 1) for the **Local community** stakeholder:

- Access to material resources
- Access to immaterial resources
- Delocalization and migration
- Cultural heritage
- Safe and healthy living conditions
- Respect of indigenous rights
- Community engagement
- Local employment
- Secure living conditions
- Others

Please, if you select others, specify here what:

4- Please select, based on opinion and expertise, which are the most representative **social indicators** (you can select more than 1) for the **Society** stakeholder:

- Public commitment to sustainability issues
- Contribution to economic development
- Prevention and mitigation of armed conflict
- Technology development
- Corruption
- Ethical treatment of animals
- Poverty alleviation
- Others



Please, if you select others, specify here what:

5- Please select, based on opinion and expertise, which are the most representative **social indicators** (you can select more than 1) for the **Consumer stakeholder**:

- Health and safety
- Feedback mechanism
- Consumer privacy
- Transparency
- End-of-life responsibility
- Others

Please, if you select others, specify here what:

6- Please select, based on opinion and expertise, which are the most representative **social indicators** (you can select more than 1) for the **Value chain actors stakeholder**:

- Fair competition
- Promoting social responsibility
- Supplier relationship
- Respect of intellectual property rights
- Wealth distribution

Please, if you select others, specify here what:

7- Please, if you have selected **another stakeholder in question 1**, add here what kind of social indicators could be related to it:

Questionnaire 2 for the RSA S-LCA approach.



Riga Technical University
Institute of Energy System and Environment

**Questionnaire for
THE ASSESSMENT OF THE POTENTIAL SOCIAL IMPACTS
RELATED TO THE SEAWEED HARVESTING AND PROCESSING
PROCEDURES.**

1. INTRODUCTION

We would like to ask your cooperation to fill in the following questionnaire.

The goal is to acquire opinions and information about potential social impacts on stakeholders that can be directly involved in the seaweed harvesting/collection/cultivation and seaweed refining sector.

The S-LCA aims to assess social consequences (positive or negative) on different stakeholder categories:



Workers



Local
community



Consumers



Value chain
actors

2. THE QUESTIONNAIRE FOR "WORKERS" STAKEHOLDERS



Social indicator: Health and Safety

Q1. Could the workers of the processes listed below be exposed to risks of accidents/damages to human health, assuming complete compliance with all the health and safety standards and laws?				
Processes	YES	If YES, which kind of risk (high, medium, low)? You can also describe it.	NO	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation				
Seaweed processing				

Q2. Could the workers of the processes listed below carry out strenuous activities?				
Processes	YES	If YES, which kind of risk (high, medium, low)? You can also describe it.	NO	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation				
Seaweed processing				

Social indicator: Working hours

Q3. Could the workers of the processes listed below exceed the 40-48 hours worked per week?					
Processes	YES	If YES, which kind of risk:		NO	I DO NOT KNOW (I have not appropriate info and/or data)
		- High (>50% of workers exceed)	- Medium (<50% of workers exceed)		
Seaweed harvesting/collection/cultivation					
Seaweed processing					

Social indicator: Social benefits/social security

Considering this list of social benefits:

- Medical insurance
- Dental Insurance
- Paramedical insurance (i.e., preventive medicine)
- Medicine Insurance
- Wage insurance
- Paid maternity and paternity leave
- Paid sick leave
- Education and training
- Meal voucher
- Agreement with gyms, kindergartens, etc.
- Others

Q4. Do the workers involved in the processes listed below have access to the majority of these social benefits as part of their employment perks?					
Processes	YES, If they can obtain more than 6 social benefits. If you select others, please specify	NO	If NO, which kind of risk level:		I DO NOT KNOW (I have not appropriate info and/or data)
			- High (no social benefits)	- Medium (1-2 social benefits)	
Seaweed harvesting/collection/cultivation					
Seaweed processing					

3. THE QUESTIONNAIRE FOR "LOCAL COMMUNITY" STAKEHOLDERS



Social indicator: *Safe and healthy living conditions*

Q5. Could the presence of the processes listed below increase the perception of a higher risk of affecting the living conditions of local communities? For example, due to occasional events such as accidents or exposure to emissions in the environmental matrices.				
Processes	YES	If YES, which kind of risk level (High, medium, low)? You can also describe it.	NO	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation				
Seaweed processing				

Social indicator: *Local economic development*

Q6. Could the presence of the processes listed below contribute positively to the development of the local economy?				
Processes	YES	If YES, could you explain how?	NO	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation				
Seaweed processing				

Q7. Could the presence of the processes listed below contribute positively to the employment of local workers?				
Processes	YES.	NO or only to a rather extent	NO	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation				
Seaweed processing				

Q8. Could the presence of the processes listed below enhance the activity of local suppliers (e.g., for providing materials, components, or services)?

Processes	YES	NO or only to a rather extent	NO	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation				
Seaweed processing				

Q9. Could the presence of the processes listed below enhance the development of local "satellite activities"?

Processes	YES	If YES, which kind of activity	NO	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation				
Seaweed processing				

4. THE QUESTIONNAIRE FOR "CONSUMER" STAKEHOLDERS



Social indicator: *Health and safety*

Q10. Are the processes listed below considering the health and safety of the consumers?

Processes	YES	If YES, which kind of activity (i.e., the presence of specific management measures, labels of health and safety requirements, an adaptation of ISO 9001 standard, etc....)	NO	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation				
Seaweed processing				

Social indicator: *Transparency*

Q11. Are the processes listed below targeting transparency for the consumers?				
Processes	YES	If YES, which kind of activity (i.e., certification standards, environmental labels, special indices, sustainability reports)	NO	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation				
Seaweed processing				

5. THE QUESTIONNAIRE FOR "VALUE CHAIN" STAKEHOLDERS



Social indicator: *Fair competition**

Q12. Are the "actors" involved in the processes listed below contributing to the fair competition among the other stakeholders in the value chain?			
Processes	YES	If YES, what kind of activity and to what extent (highly, positively, low, very low) do they contribute?	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation			
Seaweed processing			

* Fair competition regards anti-competitive behavior: actions of the reporting "actor" that may result in collusion with potential competitors to fix prices, coordinate bids, create market or output restrictions, impose geographic quotas, or allocate customers, suppliers, geographic areas, and product lines with the purpose of limiting the effects of market competition.

Social indicator: *Promoting social responsibility**

Q13. Are the "actors" involved in the processes listed below promoting social responsibility among its suppliers and through their own actions?			
Processes	YES	If YES, what kind of activity and to what extent (highly, positively, low, very low) do they contribute?	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation			
Seaweed processing			

* Social responsibility is an "actor's" obligation to consider the interests of its stakeholders as customers, employees, shareholders, or consumers. This indicator can be evaluated by considering if the "actor" manages its suppliers in a socially responsible way, including monitoring, auditing, and training efforts. And the possibility of taking action toward suppliers when warranted.

Social indicator: *Wealth distribution**

Q14. Are the "actors" involved in the processes listed below promoting wealth distribution in the supply chain? Some examples could be contract instruments, interbranch organization, and the definition of a fair price for the products.			
Processes	YES	If YES, what kind of activity and to what extent (highly, positively, low, very low) do they contribute?	I DO NOT KNOW (I have not appropriate info and/or data)
Seaweed harvesting/collection/cultivation			
Seaweed processing			

* Wealth distribution focuses on how the value is distributed among all the actors of the value chain. An equal distribution is obtained when a fair selling price for a product or service is established, i.e., when the price is such that it covers all the production costs and everyone returns an acceptable profit margin.

Name and Surname of the member of the panel of experts:

Affiliation:

Date:



Riccardo Paoli was born in 1996 in Varese, Italy. He earned a Bachelor's degree in Environmental and Workplace Safety Engineering (2018) and a Master's degree in Environmental and Workplace Sustainability Engineering (2021) from the University of Insubria. Since 2021, he has been a researcher at the Institute of Energy Systems and Environment at Riga Technical University and is currently working as a Sustainability Engineer at ABB. During his doctoral studies, Riccardo was a visiting student at the University of Texas at Arlington and received the Young Scientist Award at the Baltic Sea Science Day in 2022.

His research focuses on cleaner production, circular economy approaches, and sustainability strategies, particularly in complex value chains and emerging technologies.