A Circular Bioeconomy Solution for the Pulse Industry: Potential of Crude Pea Starch Upcycling through Single Cell Protein Pathway

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Dedication

I dedicate this thesis to two special pillars in my life, my dad, Paul Aidoo, and my mum, Margaret Aggrey, for being outstanding partners in this life's journey and showing me the path to self-discovery.

ABSTRACT

As the generation and underutilization of starch-rich co-products continue to prevail in a dominant linear pea protein extraction industry, deploying sustainable value recovery strategies remains an urgent endeavor. This study explored the potential of Single Cell Protein (SCP) as a circular bioeconomy solution for enhancing the sustainability performance of the pea industry. The first part of the study took a perspective interest in developing a comprehensive circular bioeconomy accounting tool, abbreviated as C-BEAT, as a novel and timely contribution to the circular bioeconomy evolution. The second part of the study ascertained the sustainability performance and sensitivity of a pea-starch-based SCP design as a preliminary analysis for selecting optimal process scenarios for the C-BEAT application. Life Cycle Assessment (LCA) showed a higher relative contribution of the baseline system to the marine ecotoxicity and human non-carcinogenic toxicity, with lower contributions to global warming (3.22 kg CO2 eq/kg SCP) and land use (0.012 m2a crop eq/kg SCP) categories. The harvesting procedure was identified as the hotspot unit process, contributing about 85% and 98% to global warming and marine ecotoxicity. A process scenario assessment demonstrated eliminating the media enrichment process and using organic carbon sources in inoculum production (SCP ONME) as a beneficial scenario for pea starch SCP production, offering about 26% of land use offset. Transportation sensitivity identified air and train freight as sustainable freight options, respective of mileage and mass. Better environmental benefits were achieved when hydropower dominated the selected electricity supply, and low hydropower:wind power ratio appeared as a promising energy supply scenario. Process benchmarking showed closely comparable impact ranges for pea starch SCP, feed products, and chicken, while beef maintained its outrageous performance in most impact categories. Consequentially, it was sustainable to substitute the feed products with pea starch SCP, with a stronger emphasis on fishmeal substitution. These findings from the life cycle assessment portrayed a two-sided inference for pea starch SCP production. One side hinted at a generic limitation to the sustainability contributions of pea starch SCP, informing a need to critically consider regional sustainability trends before recommending its adoption. The other side projects its prospects as a frontline climate and biodiversity action in areas where pea production and processing dominate. The LCA aided in selecting three optimal scenarios for the C-BEAT application. Herein, the economic and social performance of the three optimal scenarios were assessed and further tested with the novel Circular Bioeconomy Index and BWM-CoCoSo multicriteria decision method. The CBI assessment showed no substantial distinction in scenario performance, placing all the process options in a common sustainability region. Nonetheless, the BWM-CoCoSo methodology revealed an explicit distinction between the scenarios, identifying the process scenario which considered no media enrichment and utilized organic carbon source in the inoculum production (labeled as SCP ONME) as a relatively sustainable option. In conclusion, pea starch SCP production with less reliance on synthetic enrichment is emphasized as a sustainable upcycling option for enhancing co-product management in the pea protein extraction industry, establishing its viability for local and global exploration. It is also emphasized as an opportunity to be leveraged in the aquaculture industry for sustainable feed production. Finally, the C-BEAT is recommended as an advanced, robust, and sector-adaptable framework for circular bioeconomy practice at all levels of decision-making.

Keywords: Pea Starch, Circular Bioeconomy, Single Cell Protein, Sustainability Assessment, Biocircular Decision

RÉSUMÉ

Comme la génération et la sous-utilisation de co-produits riches en amidon continuent de prévaloir dans une industrie dominante d'extraction linéaire de protéines de pois, le déploiement de stratégies durables de valorisation reste une entreprise urgente. Cette étude a exploré le potentiel des Protéines à Cellule Unique (SCP) en tant que solution circulaire de bioéconomie pour améliorer la performance en matière de durabilité de l'industrie du pois. La première partie de l'étude s'est intéressée au développement d'un outil de comptabilité de bioéconomie circulaire complet, abrégé en C-BEAT, en tant que contribution nouvelle et opportune à l'évolution de la bioéconomie circulaire. La deuxième partie de l'étude a évalué la performance en matière de durabilité et la sensibilité d'une conception de SCP à base d'amidon de pois, en tant qu'analyse préliminaire pour sélectionner des scénarios de processus optimaux pour l'application du C-BEAT. L'Analyse de Cycle de Vie (LCA) a montré une contribution relative plus élevée du système de référence à l'écotoxicité marine et à la toxicité non carcinogène pour l'homme, avec des contributions plus faibles aux catégories du réchauffement climatique (3,22 kg CO2 éq/kg SCP) et de l'utilisation des terres (0,012 m²a éq de culture/kg SCP). La procédure de récolte a été identifiée comme le processus unitaire critique, contribuant à environ 85 % et 98 % du réchauffement climatique et de l'écotoxicité marine. Une évaluation des scénarios de processus a démontré que l'élimination du processus d'enrichissement du milieu et l'utilisation de sources de carbone organique dans la production d'inoculum (SCP ONME) constituaient un scénario bénéfique pour la production de SCP à base d'amidon de pois, offrant environ 26 % de réduction de l'utilisation des terres. La sensibilité au transport a identifié le fret aérien et ferroviaire comme des options de fret durables, en fonction de la distance et de la masse. De meilleurs avantages environnementaux ont été obtenus lorsque l'approvisionnement en électricité était dominé par l'hydroélectricité, et un faible rapport hydroélectricité : éolien est apparu comme un scénario prometteur d'approvisionnement en énergie. La comparaison des performances a montré des plages d'impact étroitement comparables pour les SCP à base d'amidon de pois, les produits alimentaires pour animaux et le poulet, tandis que le bœuf a maintenu ses performances exceptionnelles dans la plupart des catégories d'impact. En conséquence, il était viable de substituer les produits alimentaires pour animaux par des SCP à base d'amidon de pois, en mettant davantage l'accent sur la substitution de la farine de poisson. Ces résultats de l'analyse du cycle de vie ont donné une inférence à double facette pour la production de SCP à base d'amidon de pois. D'un côté, ils ont indiqué une limitation générique des contributions en matière de durabilité des SCP à base d'amidon de pois, soulignant la nécessité de tenir compte des tendances régionales de durabilité avant de recommander leur adoption. D'un autre côté, ils ont projeté ses perspectives en tant qu'action climatique et de biodiversité de premier plan dans les régions où la production et la transformation de pois dominent. L'LCA a aidé à sélectionner trois scénarios optimaux pour l'application du C-BEAT. Ici, les performances économiques et sociales des trois scénarios optimaux ont été évaluées et testées avec le nouvel Indice de Bioéconomie Circulaire (CBI) et la méthode de décision multicritère BWM-CoCoSo.

L'évaluation de l'CBI n'a montré aucune distinction substantielle dans les performances des scénarios, plaçant toutes les options de processus dans une région de durabilité commune. Néanmoins, la méthodologie BWM-CoCoSo a révélé une distinction explicite entre les scénarios, identifiant le scénario de processus qui ne considérait pas d'enrichissement du milieu et utilisait une source de carbone organique dans la production d'inoculum (étiqueté comme SCP ONME) comme une option relativement durable. En conclusion, la production de SCP à base d'amidon de pois avec moins de dépendance à l'enrichissement synthétique est mise en avant comme une option de surcyclage durable pour améliorer la gestion des co-produits dans l'industrie d'extraction de protéines de pois, établissant sa viabilité pour l'exploration locale et mondiale. Elle est également mise en avant comme une opportunité à exploiter dans l'industrie de l'aquaculture pour la production d'aliments durables. Enfin, le C-BEAT est recommandé comme un cadre avancé, robuste et adaptable au secteur pour la pratique de la bioéconomie circulaire à tous les niveaux de prise de décision.

Mots-clés : Amidon de pois, Bioéconomie circulaire, Protéine à cellule unique, Évaluation de la durabilité, Décision biocirculaire

STATEMENT FROM THE THESIS OFFICE

According to the regulations of the Faculty of Graduate Studies and Research of McGill University, Guidelines for Thesis Preparation include:

Candidates have the option of including, as part of the thesis, the text of one or more papers submitted, or to be submitted, for publication, or the clearly duplicated text of one or more published papers. These texts must conform to the "Guidelines for Thesis Preparation" and must be bound together as an integral part of the thesis.

The thesis must be more than a collection of manuscripts. All components must be integrated into a cohesive unit with a logical progression from one chapter to the next. In order to ensure that the thesis has continuity, connecting texts that provide logical bridges between the different papers are mandatory.

The thesis must conform to all other requirements of the "Guidelines for Thesis Preparation" in addition to the manuscripts.

As manuscripts for publication are frequently very concise documents, where appropriate, additional material must be provided in sufficient detail to allow a clear and precise judgement to be made of the importance and originality of the research reported in the thesis.

In general, when co-authored papers are included in a thesis, the candidate must have made a substantial contribution to all papers included in the thesis. In addition, the candidate is required to make an explicit statement in the thesis as to who contributed to such work and to what extent. This statement should appear in a single section entitled "Contribution of Authors" as a preface of the thesis.

When previously published copyright material is presented in a thesis, the candidate must obtain, if necessary, signed waivers from the co-authors and publishers and submit these to the Thesis Office with the final deposition.

PREFACE AND CONTRIBUTION OF AUTHORS

This thesis includes six chapters.

Chapter 1 broadly discusses the food system and how agro-industrial waste generation affects its sustainability performance. It describes circular bioeconomy as an emerging sustainability concept, underscoring the need for conceptual remodeling to enhance practice. It then considers the potential of circular biotechnological solutions like single cell protein in augmenting value recovery in the pea industry, particularly for pea starch. It also portrays the relevance of advanced circular bioeconomy tools in enhancing such pursuits. The chapter concludes by defining the thesis's primary goal, objectives, and activities.

Chapter 2 comprises a literature review on relevant topics surrounding the production, commercial prospects, and sustainability potentials of single cell protein, which highlights second-generation substrates as sustainable feedstock alternatives for single cell protein and identifies crude pea starch as an untapped potential substrate requiring urgent exploration. Furthermore, it underpins the integration of life cycle sustainability assessment and decision tools as a profound pathway for augmenting the sustainability benefits of single cell protein designs.

Chapter 3 takes on the recommendation from the literature review to design a comprehensive circular bioeconomy framework, which leverages stakeholder engagement, life cycle sustainability assessment, and multicriteria decision analysis in modeling a conceptual circular bioeconomy accounting tool for agro-industrial practice.

Chapter 4 then designs and conducts a life cycle assessment of a baseline pea-starch single cell protein production system, determines the hotspots of the design, and performs a process sensitivity analysis to identify optimal design alternatives for further analysis.

Chapter 5 applies the circular bioeconomy accounting tool described in chapter 3 to ascertain the circular bioeconomy potentials of the optimal pea-starch single cell protein designs identified in chapter 4. Here, the social and economic performance of the designs are ascertained, and the novel circular bioeconomy index (CBI) and Best Worst Method-Combined Comprised Solution (BWM-CoCoSo) multicriteria decision analysis methodology are used to determine the ideal optimal design.

Finally, **Chapter 6** summarizes the thesis, highlighting relevant insights and recommendations for industry and public policy.

Raphael Aidoo was responsible for conceptual design, computational modeling, data curation, analysis, manuscript writing, and final editing.

Dr. Ebenezer Miezah Kwofie, the principal supervisor, was responsible for the study conception, revision, and supervision of the study.

Dr. Peter Adewale, the third author of chapters 2, 4, and 5, was responsible for the study conception, revision of manuscripts, and supervision of the study.

Dr. Edmond Lam, the fourth author of chapters 2, 4, and 5, was responsible for revising manuscripts and supervising the study.

Prof. Michael Ngadi, the fifth author of chapters 2, 4, and 5, supervised the study and gave expert inputs.

RESEARCH CONTRIBUTIONS

PUBLICATIONS

A. Peer-reviewed journal articles

1. Aidoo, Raphael, Kwofie, Ebenezer Miezah, Adewale, Peter, Lam, Edmond, and Ngadi, Michael (2023), Overview of Single Cell Protein: Production Pathway, Sustainability Outlook, and Digital Twin Potentials, *Trends in Food Science and Technology,* 138 (2023), 577-598, https://doi.org/10.1016/j.tifs.2023.07.003

2. Aidoo, Raphael and Kwofie, Ebenezer Miezah (2023), Circular Bioeconomy Accounting Tool (C-BEAT): Circular Bioeconomy Accounting Tool (C-BEAT): A Comprehensive Framework for Improving Agro-Industrial Circularity Practice (Manuscript being revised for submission)

3. Aidoo, Raphael, Kwofie, Ebenezer Miezah, Adewale, Peter, Lam, Edmond, and Ngadi, Michael (2023), Designing Sustainable Circular Bioeconomy Solutions for the Pulse Industry: The Case of Pea-Starch Based Single Cell Protein, Science of the Total Environment, https://dx.doi.org/10.2139/ssrn.4510778 (Manuscript Preprint)

4. Aidoo, Raphael, Kwofie, Ebenezer Miezah, Adewale, Peter, Lam, Edmond, and Ngadi, Michael (2023), Consequential LCA-LCC of Pea Starch Single Cell Protein: Modelling Optimal Decisions for the Aquacultural Industry (Manuscript under internal review)

5. Aidoo, Raphael, Kwofie, Ebenezer Miezah, Adewale, Peter, Lam, Edmond, and Ngadi, Michael (2023), Application of the C-BEAT in Agro-industrial System Modelling Modelling: A Case Study for Pea Starch Based Single Cell Protein. (Manuscript under internal review)

B. Conference presentation

1. Aidoo, Raphael and Kwofie, Ebenezer Miezah (2023), Circular Bioeconomy Accounting Model for Food and Bioprocesses Presented at ASABE Conference 2023, Omaha, Nebraska, USA, Oral Presentation by Ebenezer Miezah Kwofie, 8th July, 2023

2. Aidoo, Raphael, Kwofie, Ebenezer Miezah, Adewale, Peter, Lam, Edmond, and Ngadi, Michael (2023), Value Recovery from Pea-Protein Co-Product: Computational Design and Sustainability Assessment of Single-Cell Protein Production from Pea Starch, Presented at CSBE/SCGAB AGM Conference 2023, Lethbridge, Alberta, Canada, Oral Presentation, 25th July, 2023

3. Aidoo, Raphael, Kwofie, Ebenezer Miezah, Adewale, Peter, Lam, Edmond, and Ngadi, Michael (2023), Exploring the Circular Bioeconomy Potentials of Industrial Pea Starch: A Case Study for Second-Generation Single-Cell Protein Production, Presented at NABEC 2023, Guelph, Ontario, Canada, Oral Presentation, 1st August, 2023.

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NOMENCLATURE/LIST OF ABBREVIATIONS

Name	Abbreviation	Reference unit
Association of Southeast Asian Nations	ASEAN	
Best Worst Method	BWM	
Circular Bioeconomy Accounting Tool	C-BEAT	
Circular Bioeconomy Index	CBI	
Combined Compromise Solution	CoCoSo	
Deep learning	DL	
Employment Index	EI	
Food and Agriculture Organization	FAO	
Fuzzy Logic	FL	
Fuzzy Neural Network	FNN	
Least Square	LS	
Life Cycle Assessment	LCA	
Life Cycle Cost Assessment	LCCA	
Life Cycle Costing	LCC	
Life Cycle Sustainability Assessment	LCSA	
Multicriteria Decision Analysis	MCDA	
Multiple Output Variable Least Square	MLO	
Net Present Value	NPV	
Net Profit	NP	
No Media Enrichment	NME	
Organic and No Media Enrichment	ONME	
Organic with Media Enrichment	OME	
Particle Swarm Optimization	PSO	
Polyhydroxyalkanoate	PHA	
Polyhydroxybutyrate	PHB	
Power-to-food	PtF	
Recurrent Neural Network	RNN	
Single Cell Protein	SCP	

Social Life Cycle Assessment	SLCA	
Support Vector Model	SVM	
Sustainability Accounting Tools	SAT	
Technoeconomic Assessment	TEA	
Fine particulate matter formation	FPMF	kg PM2.5 eq
Fossil resource scarcity	FRS	kg oil eq
Freshwater ecotoxicity	FEC	kg 1,4-DCB
Freshwater eutrophication	FET	kg P eq
Global warming	GW	kg CO2 eq
Human carcinogenic toxicity	HCT	kg 1,4-DCB
Human non-carcinogenic toxicity	HNCT	kg 1,4-DCB
Ionizing radiation	IR	kBq Co-60 eq
Land use	LU	m2a crop eq
Marine ecotoxicity	MEC	kg 1,4-DCB
Marine eutrophication	MET	kg N eq
Mineral resource scarcity	MRS	kg Cu eq
Ozone formation, Human health	OZHH	kg NOx eq
Ozone formation, Terrestrial ecosystems	OZTE	kg NOx eq
Stratospheric ozone depletion	SOD	kg CFC11 eq
Terrestrial acidification	TA	kg SO2 eq
Terrestrial ecotoxicity	TEC	kg 1,4-DCB
Water consumption	WC	m3

1 CHAPTER ONE: GENERAL INTRODUCTION

1.1 Background

The food system is a complex entity of diverse sub-systems. Its ever-increasing role in buffering economic growth has driven the consistent development of innovative and technological solutions to reinforce resilience and promote sustainability (Agyemang, P. et al., 2022; Agyemang, Prince et al., 2022). Despite these, the food system still faces crucial sustainability challenges, partially but significantly driven by the persistent generation of voluminous waste. For instance, several reports have highlighted the aggravating impacts on planetary boundaries accompanying agricultural and agro-industrial waste, wherein global warming and land occupancy impacts stand out among the longlist of impact categories (Aidoo, Raphael et al., 2022; Areniello et al., 2022; Balanay et al., 2022). Moreover, these wastes pose significant socioeconomic threats, with extant reports establishing the impact of food waste on global food security. According to FAO, addressing current food waste could provide food to about 3 billion people, addressing the food needs of about a third of the global population (FAO, 2023). Though mitigative strategies are gradually emerging, sustainability actions have not adequately advanced, indicating the necessity to double current efforts toward a sustainable food system. The circular bioeconomy framework is quickly gaining traction as a supportive framework to enhance these efforts (Mabee, 2022; Navare et al., 2021). It is shifting focus to the standing opportunity to promote food system sustainability and minimize the impacts of the dominant linear economic model through its regenerative thinking strategy. It further suggests an opportunity to maximize value recovery in the food system to close sustainability loops, hinting at a vast potential in the agro-industry (Navare et al., 2021).

The agro-industry is a global economic backbone, offering substantial benefits in satisfying food demands for economic success. However, it is a source of substantial quantities of solid and liquid residues with a high potential for high-value upcycling. For instance, the biomolecular extraction industry, including fat, starch, and protein extraction from grains, seeds, and pulses, generates enormous amounts of valuable yet underutilized by/co-products. This has instigated regenerative actions to maximize sustainability benefits. Consequently, interest in biotechnological solutions has gained significant attention with reports highlighting the sustainability disposition of biotechnological solutions, especially in offsetting land use and global warming potential associated with industrial wastes by consuming them as feedstocks for high-value pathways (Raziq, 2020; Sakarika et al., 2022). Typical examples are biofuel production, fine chemicals extraction, and the production of microbial foods from biomass (Agyemang, Prince et al., 2022; Areniello et al., 2022; Bonan et al., 2021). The place of microbial foods is particularly resounding, looking at their multidirectional benefits, especially in providing sustainable food and feed materials to satisfy growing needs and driving sustainable waste management. In this, Single Cell

Protein (SCP) is rapidly surging for its potential to augment protein supply in the face of exacerbating protein insecurity. It is tagged with benefits in improving the sustainability performance of bioindustrial systems through integrative upcycling approaches. Recent exploration includes bioindustrial wastewater upcycling for protein-rich microbial biomass, which is being expansively utilized in the aquacultural industry for feed production due to its appreciable nutritional composition. In recent explorations, several SCPs have been explored as substitutes for fishmeal and soybean meal, with significant benefits attached to carbon offsets and land use savings (Spiller et al., 2020; Türker et al., 2022).

This evolving interest in circular bioeconomy as a decision-support framework, coupled with the resurging interest in SCP as an industrial biotechnological solution for waste and co-product management, prompted the relevance of exploring SCP as circular bioeconomy solution in the pea industry. The pea industry is rapidly expanding due to the increasing need to satisfy raw material demand for sustainable protein production. However, reports attach a substantial generation of underutilized starch-rich co-products to the pea processing industry due to the siloed focus on protein extraction, denting the overall sustainability goal of plant-protein extraction (Ren et al., 2021; Zhou et al., 2021). Previous practices have explored transforming these co-products into commercially viable pure and modified starch for food and non-food application (Leite et al., 2017; Olagunju et al., 2020; Zhou et al., 2019). However, this has not yielded significant commercial success, impeded by the availability of cost-effective, highly competitive, high-quality starches from staples like corn, wheat, and rice, among other sustainability implications related to the high demand for harsh chemicals and energy for these processes. This has prompted the need to engage other commercially feasible and sustainable pathways. In supposition, a biocircular path that consumes the co-products and produces useable protein products seems a convenient and sustainable option for rapidly achieving commercial success, hinging on the rising need to address protein insecurity issues amidst rapid population growth. Herein, SCP production is a feasible choice amongst the list of alternatives.

As this “circular bioeconomy in the pea industry” remains a promising co-product management approach, engaging suitable methodologies to facilitate sustainable and optimal upcycling designs is also vital. This is to sustain the alignment of circularity practice with global sustainability goals. Circular bioeconomy has previously followed developments in the general circular economy model, applying the ReSOLVE framework as an adaptable guideline for identifying circularity hotspots within biosystems. Though this has significantly improved practice, recent discussions underlie an urgent need to go beyond simply identifying and implementing biocircular pathways. Experts advise the relevance of developing robust decision-support models that prioritize stakeholder inclusion, life cycle sustainability assessment, and multi-objective modeling in every decision-making process and how these would reinforce the precision, applicability, and reliability of biocircular decisions (Engels &

Jonker, 2022; Khitous et al., 2022). In justification, the involvement of multiple stakeholders enables the identification of feasible and market-ready pathways with optimal commercial success rates, notable for businesses with strategic stakeholder and market inclusion actions (Agyemang, Prince et al., 2022).

Moreover, life cycle sustainability assessment provides insights into the sustainability placement of identified pathways across the sustainability tenets, which may inform further optimization for maximum performance. Furthermore, businesses or industries are primarily profit-making entities that take on other environmental and social responsibilities, usually less rated than their grand economic ambitions. As such, for biocircular pathways to gain substantial industrial traction and subsequent economic success, associated models must facilitate decisions supporting economic goals while rendering substantial environmental and social benefits. This could be achieved through multi-criteria decision-making, which allows optimal decisions that satisfy weighted criteria. The above discussion triggered the question of how circular bioeconomy can be improved for advancing practice in the agro-industry, focusing on its potential in the pea industry. This informed the study's general goal of exploring circular bioeconomy as a sustainable pathway for promoting crude pea starch upcycling, focusing on its potential for Single Cell Protein production. Three objectives with related activities were formulated to achieve this goal, as described in the subsequent subsection.

1.2 General Objectives and Activities

Objective 1: To develop a comprehensive and adaptable circular bioeconomy accounting tool (C-BEAT) to enhance circular bioeconomy practice in the agriculture and agri-food industry.

a. Activity 1a: Design and describe a theoretical circular bioeconomy accounting tool (C-BEAT) considering stakeholder engagement and sustainability assessment. This model is framed to facilitate biocircular decisions and place optimal scenarios within an attainable region of maximum economic and social impacts and minimum environmental impact.
b. Activity 1b: Develop a unified circular bioeconomy index (CBI) to enhance the aggregation of sustainability results. This is to provide a relatable yet relative score for biocircular communications.
c. Activity 1c: Identify and describe a multi-criteria decision method for trade-off analysis of circular bioeconomy scenarios or pathways for optimal decision.

Objective 2: To ascertain dynamics in the sustainability performance of a pea-starch-based Single Cell Protein production system in response to systemic variation in the baseline design.

a. Activity 2a: Computational design of a solid-state fermentation-based Single Cell Protein Production System using pea starch as a carbon substrate.
b. Activity 2b: Conduct a life cycle assessment (LCA) to determine the environmental impacts of the baseline system.

c. Activity 2c: Design and assess varying scenarios to identify system sensitivity to variations in selected system elements for optimal design selection.
d. Activity 2d: Conduct a consequential LCA to ascertain the variation in impact for applying pea-starch SCP as a substitute for fishmeal and soybean with varying substitution ratios

Objective 3: Apply the C-BEAT model for assessing the circular bioeconomy potentials of pea-starch SCP process scenarios

a. Activity 3a: Determine the life cycle cost and employment index of the various process scenarios described in objective 2.
b. Activity 3b: Calculate the Circular Bioeconomy Index of the various scenarios based on the steps described in Objective 1.
c. Conduct a multicriteria decision analysis of the various process alternatives using the environmental, economic, and social metrics as decision criteria.

CONNECTING TEXT I

The introductory chapter (Chapter 1) gave a broad overview of the background of the study, emphasizing the remodeling of the circular bioeconomy framework and exploration of single cell protein in the pea industry as the primary goal. Furthermore, it emphasized how a comprehensive circular bioeconomy accounting tool could augment agro-industrial practices in ensuring sustainable design and selection of an optimal pea starch SCP scenario. The following chapter (Chapter 2) conducts a literature review of single cell proteins, broadly looking at the production steps, sustainability outlook, and opportunities for improving performance. It serves as the basis for identifying the process flow for single cell protein production and consolidating knowledge and data for subsequent simulations and analysis.

2 CHAPTER TWO: LITERATURE REVIEW

Overview of Single Cell Protein: Production Pathway, Sustainability Assessment, and Digital Twin Potentials

Abstract

Single-cell protein (SCP) is an evolving biotechnological concept that can potentially align protein production with the global sustainability commitment. This review presents an overview of SCP's current outlook, including production, commercial prospects, and sustainability status. It also elaborates on the potential of the evolving digital twin concept in improving SCP production's efficiency and sustainability performance. An expanding body of work was identified, with well-explored fermentative approaches and varying substrates and microbes. Whereas interest in first-generation substrates such as methane is gradually fading due to their high competitiveness and acquisition cost, second-generation substrates such as lignocellulosic materials and agro-industrial wastes are rapidly evolving due to their availability, low cost, appreciable nutrient density, and alignment with the circular bioeconomy path. Sustainability assessment of current production attaches substantial environmental, mainly global warming and land use offset, and economic savings to SCP production. Moreover, it emphasizes conventional energy use as a hotspot contributor to all impact categories. However, research on life cycle costing, social life cycle assessment, and environmental nutrition concepts is limited. Current trends project rapid market growth for SCP due to expanding feed, food, and nonfood applications. The rapid influx of transformative innovations such as mixed culture biotechnology and the emerging digital twin concept that present catalytic advantages in achieving market growth and sustainability co-benefits are backing these projections. Several sensor and predictive technologies are available to enable an SCP-digital twin path, presenting an opportunity to enhance green and precision SCP production. Despite these innovations, significant efforts are required to overcome limitations concerning toxicity, legislative restrictions, technical constraints, and consumer neophobia to bolster commerce and market value.

Keywords: Single cell protein, Sustainability assessment, Digital twin, Circular bioeconomy, Second generation substrates, Fermentation

2.1 Background

Current anthropogenic activities have threatened the integrity of the planet. In the food system, for instance, crop and animal production and successive processes that transform raw materials into edible and shelf-stable forms have driven resource demands and increased associated emissions that imperil the earth's impact-bearing capacity (Agyemang & Kwofie, 2021; Aidoo, Raphael et al., 2022). Such is also the case for the protein supply chain, where the increase in the global population has coerced global proclivity toward producing more protein foods to satisfy the current needs of consumers. From empirical data, stakeholders have ascertained how conventional protein supply methods, prominently animal production, result in astronomical environmental impacts, deviating from the global intent to produce and consume sustainably. As a result, several remediation strategies have emerged, including transitioning to more sustainable protein alternatives, including plant-based alternatives, insects, cultured meat, and single cell protein (SCP). In this study, we draw attention to the progressing prominence of microbial SCP in enhancing sustainable protein production and consumption. With an estimated tripling in market value within the next decade (2020-2030), it is more important to understand current dynamics in SCP production to facilitate innovations toward meeting these projections and beyond (Global Market Insights, 2023).

SCP derived its name from being produced from unicellular microorganisms (Abdullahi et al., 2021; Nasseri et al., 2011). The microbes are cultivated from either first-generation substrates of high commercial value (for example, methane, ethanol, methanol, and gas oil) or from second-generation substrates such as nutrient-rich agricultural and agro-industrial residues (Abdullahi et al., 2021; Banks et al., 2022; Owsianiak et al., 2022; Rajendran et al., 2018). Furthering the advantages of first-generation substrates in SCP production, which endeavored primarily to augment protein security in a socioeconomically diverse food system amidst a growing global population, second-generation substrates offer this advantage while benefitting the evolving circular bioeconomy and food system sustainability frameworks. It does so through the renewable bioconversion of waste into high-value microbial protein products. Solid, liquid, and semisolid fermentation technologies have been designed and explored in cultivating several yeast, bacteria, algae, and filamentous fungi strains with appreciable process outputs recorded relative to fermentation conditions, substrate characteristics, and microbial type (Abdullahi et al., 2021; Banks et al., 2022). Among these microbes, yeast is the trajectorial pioneer, having a longstanding historical association with World Wars I and II (Abdullahi et al., 2021; Nasseri et al., 2011). At the emergence of World War I, yeast served as a suitable unconventional alternative to complement protein demand in the war zones. Its nutritional quality subsequently inspired the drastic reduction in protein importation among the Germans during World War I, replacing almost 50% of imported proteins with locally cultivated yeast. What was seen as an economic buffer and a transient protein resort in a period of commotion and national instability has evolved into a revolutionary food system pursuit for enhancing protein-energy security (Abdullahi et al., 2021; Nasseri et al., 2011).

Aside from the nutritional, health, and environmental benefits SCP can provide, extensive research and innovations have attached techno-economic and socioeconomic benefits to their production and consumption. For instance, whereas most conventional proteins have been associated with high resource demand, generation time, and capital investment, SCP production is noted to the contrary. The consumption of readily available and inexpensive residual feedstocks minimizes resource demand and the shorter generation time of microbes (bacteria: 30-120 min, yeast, 40-180 min, algae: 180-360 min) (Nasseri et al., 2011; Sharif et al., 2021) presents comparably attractive product-process efficiency benefits. For capital investment, the argument prevails to associate such benefits with the technology or technique used, with solid-state fermentation (SSF) identified as a relatively cheaper technological option due to less water and energy demand (Areniello et al., 2022). These benefits have enabled a gradually surging preference for alternative proteins such as SCP (Areniello et al., 2022; Ritala et al., 2017; Wada et al., 2022). However, major technical and process inefficiencies, such as difficulty in scaling up the SSF process, high nucleic acid contents, and other metabolic constraints, have limited the commercial potential of SCP production. Thus, there has been a resurging necessity for radical collaboration, continuous research, and technological advancements to improve performance and maximize commercial benefits.

In this study, we review the current outlook of SCPs. The paper is divided into three major broad parts, summarized in Figure 2.1. The first part (Sections 2 and 3) captures the production outlook, highlighting the production approaches, general production steps, and some microbes and substrates for the production process, including emerging advances. The second part (Section 4) discusses the commercial status of SCP, capturing the benefits of bolstering commercial entry, the current market dynamics, and current applications. As sustainability stands as a primary benefit of SCP production, the third part of this review (Section 5) delves into exploring the progress of sustainability research, including Life Cycle Assessment (LCA), Techno-economic Assessment (TEA), Life Cycle Cost Assessment (LCCA), Social Life Cycle Assessment (TEA), and Environmental Nutrition. It also elaborates on the novel digital twin concept as an Industry 4.0 technology for improving SCP production and sustainability. Relevant technologies (sensors) and computer models that could be leveraged are briefly discussed, and specific applications of these technologies are highlighted. For emphasis and clarity, this paper does not capture the actual simulation of a digital twin for SCP production. It instead presents insights into the current potential that could be deployed to enable precision SCP production in an Industry/Technology 4.0 era. The study concludes with a summary of the significant takeaways from the review, drawing into perspective what future research should focus on and recommending pathways for progressing sustainable SCP production (Agyemang & Kwofie, 2021).

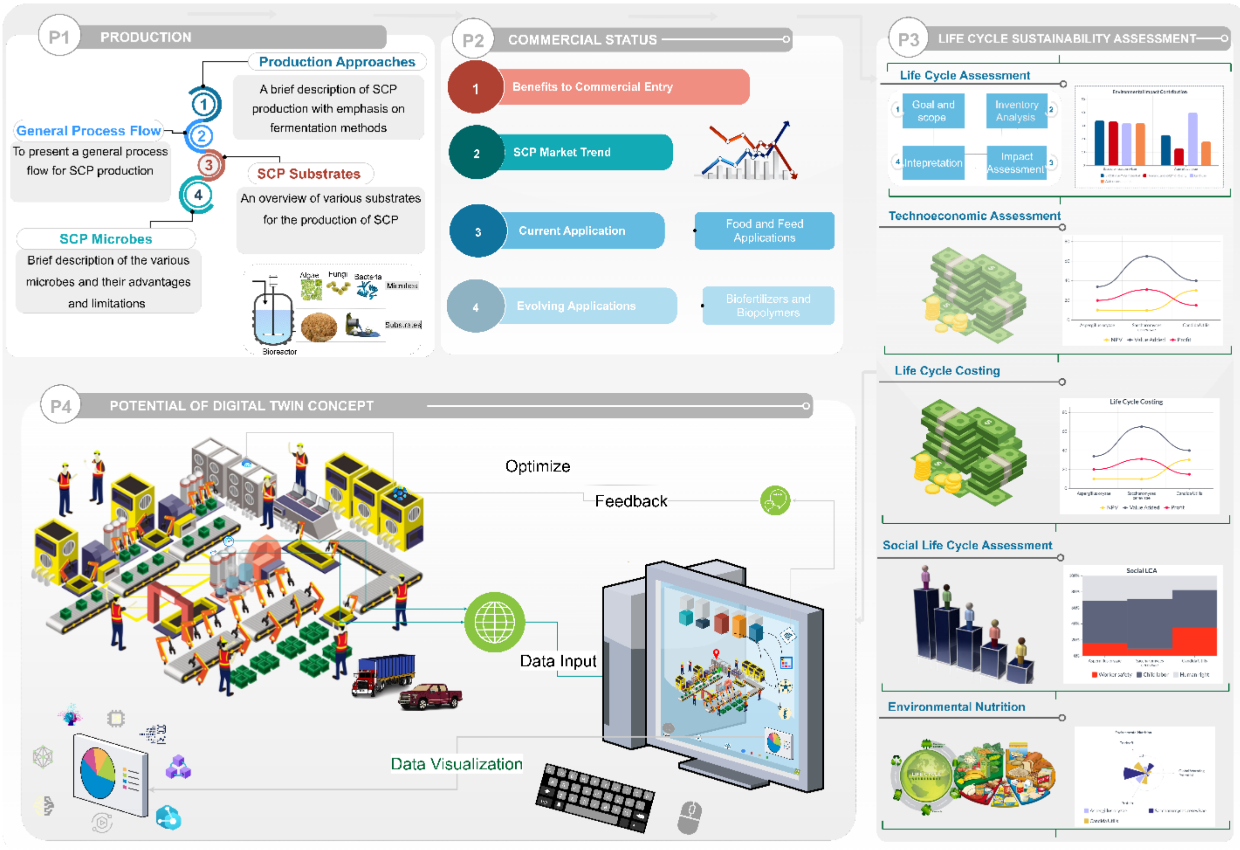

Figure 2.1: Graphical summary of the review scope

2.2 SCP Production

SCP production follows a fermentation pathway that utilizes nutrient-rich feedstock to multiply a microbe into protein-rich biomass under optimal nutrient and substrate concentration conditions and other critical fermentation parameters, such as oxygen, temperature, and pH. This section captures two crucial aspects of SCP production, the fermentation approaches and a general flow of the production steps.

2.2.1 Fermentation Approaches

2.2.1.1 SSF

SSF involves the cultivation of microbes on a solid substrate without a free aqueous phase in varying fermenter designs (Bajpai, 2017). The microbes depend on the intrinsic moisture composition of the moist-solid substrate, requiring little to no additional water. Thus, SSF is restricted to microorganisms that grow efficiently under low water activity, including yeast and some filamentous fungi, requiring pure solid substrates with approximately 60-65% moisture (Sharif et al., 2021). The microbes access nutrients through adsorption or penetration of the solid substrate, allowing them to multiply into protein-rich biomass. A nutrient concentration gradient exists in SSF; thus, nutrient diffusion is required to ensure optimal access to nutrients for microbial doubling. The fermentation process requires an adequate oxygen supply in the liquid phase, achieved through aeration and intermittent media stirring, and optimal temperature, pH, ionic strength, and nutrient conditions for optimal yield (Areniello et al., 2022). SSF is advantageous for its relatively low capital investment and minimal waste generation, offering better economic and environmental compensation (Aggelopoulos et al., 2014). Therefore, it is no surprise that evolving fermentation-based research is rapidly inclining toward the SSF approach, intending to enhance techno-economic and sustainability benefits (Aggelopoulos et al., 2014; Muniz et al., 2020; Webb, 2017). However, SSF faces critical challenges that demand significant innovations to activate its full commercial potential. Prominent among these are the current difficulties in technical scale-up, impeding commercial scale production, and limitations to online monitoring and process control, which demands urgent interventions to spur commercialization and survival of the SSF approach in a rapidly evolving digital economy (Areniello et al., 2022; Bajpai, 2017). Additionally, difficulty in stirring and removing metabolic heat in the SSF approach has been critical to process efficiency; however, recent developments promise better system designs to surmount these limitations (Areniello et al., 2022; Jach et al., 2022).

2.2.1.2 LSF

Unlike SSF, liquid-state fermentation (LSF) requires microbial cultivation in a continuous liquid-phase substrate containing more than 95% moisture. The fermentation process is carried out in a closed bioreactor, usually in continuous mode, with proper control of temperature, pH, nutrients, and oxygen supply. LSF is widely adopted in industrial fermentation processes due to its advantages in easy technical scale-up, uniform distribution of nutrients and oxygen facilitated by its continuous liquid phase, and high protein yield. It is also easy to remove metabolic heat and monitor or control the fermentation process online (Areniello et al., 2022; Sharif et al., 2021). Despite these advantages, the characteristic high capital demand and high waste generation of LSF are gradually reducing its attractiveness in an evolving sustainability-sensitive economy. While research and technology continue to progress toward optimizing these limitations and enhancing system performance, intensified traction toward the SSF approach, primed by its sustainability prospects, seems to suggest the possibility of overtaking LSF in future industrial adoption. However, this possibility will be challenged in an industry/technology 4.0 era unless present complexities in technical scale-up, heat, mass transfer, and digitization evolve simultaneously with growing interest.

2.2.1.3 Semi-Solid-State Fermentation

Semisolid-state fermentation is an intermediary between SSF and LSF. Here, the free-flowing liquid content is increased to facilitate the distribution of nutrients and oxygen (Sharif et al., 2021). This marks its preference as an intermediate approach for microbes that require slightly high-water activity but perform better on a solid substrate. However, as an intermediate approach, its advantages and disadvantages fall between SSF and LSF. For instance, while it offers moderate metabolic heat removal facilitated by its slightly higher liquid phase relative to SSF, it is characterized by high capital investment relative to SSF and lower protein yield relative to LSF (Areniello et al., 2022).

2.2.2 General production steps

A graphical representation of the process flow for SCP production is illustrated in Figure 2.2. Production of SCP starts with substrate preparation: the primary step for transforming substrates into a usable carbon source. The substrate preparation method depends on the type of substrate and fermentation approach. In recent literature, second-generation substrates (SGS) have been prepared using wet, direct, and dry preparation methods (Abdullahi et al., 2021) interlinked with LSF, SSF, and semi-solid state fermentation, respectively (Abdullahi et al., 2021; Pereira et al., 2022; Ritala et al., 2017). The wet preparation method is usually used for fresh fruit, vegetable waste, or substrates of high moisture content. Generally, it involves a series of water or acidic washing, pulverization, filtration, and sterilization to obtain a sterile liquid medium. The dry method is usually used for low-moisture substrates. It employs a drying procedure to reduce moisture further, followed by a diminution and sifting step that produces a fine powder of defined particle sizes. The fine powder is then blended with water, filtered, and sterilized to obtain a sterile moist-solid medium with a prominent aqueous phase (Abdullahi et al., 2021; Areniello et al., 2022).

The direct method is used when the SSF approach is preferred. It involves water washing and hydrolytic procedures, including acid, bio-hydrolysis, or thermal treatment, to convert the substrate into a medium ready for fermentation. Given their low moisture content and bulkiness, lignocellulosic materials usually follow dry and direct preparation methods. However, grinding and digestion of the substrate cannot be compromised in their preparation. Sterilization of the resulting media is usually performed with an autoclave at a temperature of 121°C for approximately 15-60 min. The autoclaving time depends on the substrate and the measure of contamination. However, the sterilization step could be omitted during substrate preparation, taking its significance in the fermentation process and effect on substrate integrity (Gervasi et al., 2018).

Following the substrate preparation step is media enrichment, performed to augment the nutritional capacity of the resulting media. In some cases of the application of second-generation substrates in SCP production, sole dependence on the substrate as the source of nutrients for the fermentation process fostered optimal microbial doubling without enrichment. This highlights the possibility of skipping the media enrichment step. However, it is relevant to understand the nutritional characteristics of the substrate and its capacity to augment the nutritional needs of the microbe before making such a decision.

Next is the inoculation stage, which could involve isolating or culturing microbial cells to attain a substantial load before transferring them to the prepared media. Otherwise, a viable inoculum can be purchased from sellers and used directly, or the back-slopping procedure could be employed accordingly. Next, the inoculated medium is incubated for a defined period. Within this period, the fermentation environment is regulated to enhance the multiplication of the microbes.

Harvesting follows the incubation step. Here centrifugal (yeast and bacteria) and filtration (filamentous fungi) technologies (Nasseri et al., 2011) are employed to separate SCP from the resulting biomass. The SCP is dried to about 10 % moisture content to enhance storage life. Spray, drum, and freeze-drying techniques have been widely used in SCP drying. Based on the use of the SCP, techniques like protein purification (Thiviya et al., 2022), cell disruption (Ugbogu & Ugbogu, 2016), protein extraction (Sharif et al., 2021), and nucleic acid removal (Thiviya et al., 2022; Ugbogu & Ugbogu, 2016)may be required before final stage drying and storage.

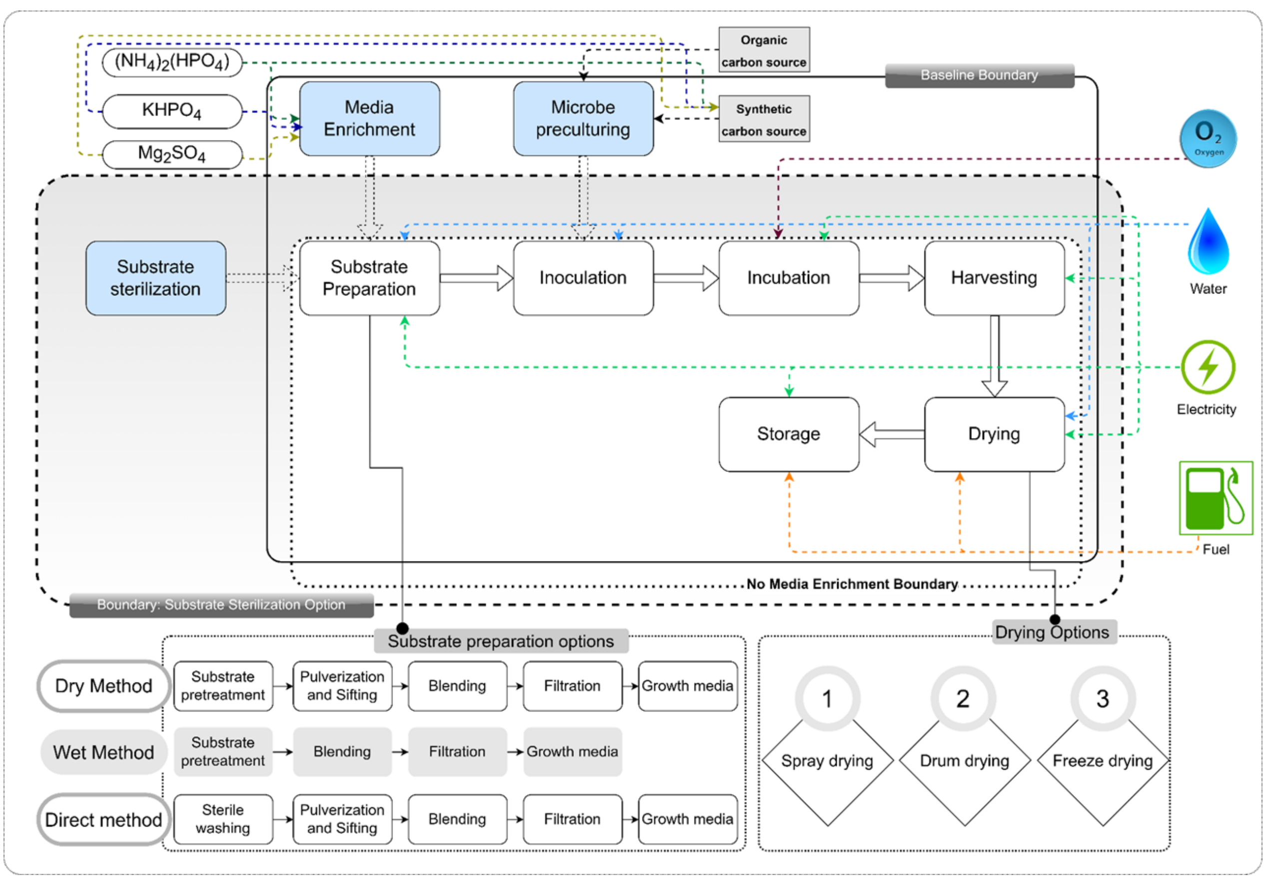

Figure 2.2: Typical Production flow of SCP

2.2.3 Substrates and Microorganisms for SCP Production

The previous section gave an overview of SCP production. This section briefly discusses some substrates and microorganisms explored in SCP production, highlighting their characteristics, advantages, and disadvantages.

2.2.3.1 Overview of Substrates

SCP substrates have generally been classified under first and second-generation categories (Abdullahi et al., 2021; Banks et al., 2022; García Martínez et al., 2022). First-generation substrates include methane (García Martínez et al., 2022), methanol, gas oil (Nasseri et al., 2011; Raziq, 2020), and staple food ingredients like corn, cassava, and rice flour, which are highly competitive for their wide adoption in commercial processes. However, despite advantages like high production rate and protein yield, challenges like high cost, technical constraints, high toxicity and carcinogenicity, commercial competitiveness, and adverse environmental implications continue to limit commercial utility (García Martínez et al., 2022; Nasseri et al., 2011), which has necessitated rethinking their use. This has triggered interest in inexpensive and environmentally friendly SGS, materials that have lost their primary value or are commercially unattractive in their raw forms (Hulsen et al., 2022; Janssen et al., 2022; Pihlajaniemi et al., 2020). SGS are characterized by appreciable nutritional densities and other favorable characteristics that can completely displace first-generation substrates in SCP production while offering exclusive benefits (Banks et al., 2022). A broad list of SGS for SCP production has been identified in the literature and captured under broad categories; on-farm agricultural wastes, including manure and lignocellulosic materials (Bratosin et al., 2021; Spalvins et al., 2018; Thiviya et al., 2022), agro-industrial processing wastes (Bajpai, 2017; Türker et al., 2022), and lost agro-products (Areniello et al., 2022; Gervasi et al., 2018; Ritala et al., 2017).

2.2.3.2 Rising Interest in Starch-rich Pulse Co-products as SGS for SCP production

Pulses are protein-rich crops with numerous advantages as food or feed or ingredients for food and feed products. In present deliberations, their roles as sustainable protein alternatives have been heightened. Therefore, the drastic progress experienced in the pulse industry over the past few years is no surprise. One such is the dramatic expansion in production in response to satisfying raw material needs for plant protein extraction. For instance, within the past two decades, from 1998-2018, pulse production has increased by approximately 63 % (Ben-Belhassen & Rawal, 2023). The leading pulse crops in this expansion are peas, lentils, chickpeas, and faba beans (Ren et al., 2021).

There has been tremendous production growth in the pea industry, with about a 100% increase in global production reported between 1961-2020 (FAO, 2022). Regional distribution of this increase indicates an approximation of 26000% in Estonia, 15500% in Canada, 1500% in West Africa, and 510% in the United States

(US). These values precisely communicate the booming interest in pea and pulse production. However, although this massive expansion is desirable in augmenting raw material supply for alternative protein production, the skewed interest in protein extraction curtails realizing the full sustainability potential of the pulse industry.

Pea contains about 13-40% proteins and 30-50% starch (Daba & Morris, 2021), emphasizing the considerable mass of starch-rich waste generated during pea protein extraction. Currently, pea starch slurry is commonly dried into pea starch flour, which is struggling in commerce due to its undesirable functionalities presented by the high amylose: amylopectin ratio (Daba & Morris, 2021; Ren et al., 2021). Also, fibre and remnant proteins minimize purity, hindering the use of pea starch flour in other non-food applications (Ren et al., 2021). Current technologies have employed modification strategies to enhance functionalities and improve applicability (Gebremedhin & Admassu, 2022). However, these have not charted any satisfactory commercial success. Therefore, enabling the full sustainability potentials of the pea and pulse industries would require finding high-value, market-ready upcycle solutions for utilizing these co-products. As biotechnological innovations gradually populate governmental and industrial strategies toward addressing waste challenges and enhancing protein security, fermented foods have been identified as a momentous circular solution for utilizing these starch-rich pulse co-products to achieve economic, environmental, and food security co-benefits, and SCPs fall within this context (Adebo et al., 2017). Nonetheless, these have not received the necessary engagements, especially for SCP production, presenting a significant gap that could be explored in future alternative protein production.

2.2.3.3 Overview of Microbes

Various fast-growing, nutritious, and generally recognized as safe (GRAS) microbial strains from bacterial, yeast, fungal, and algal sources have been identified and utilized in SCP production (Raziq, 2020). Microbes exhibit varying structural, chemical, and doubling characteristics that determine process dynamics and influence the quality and use of the final microbial biomass. For instance, bacteria and yeast possess high nucleic acid contents and bear the potential to release toxins during fermentation, obstructing interest in human food applications (Abdullahi et al., 2021). It would take process optimization and advanced technologies that can overcome these constraints to spur SCP food application (García Martínez et al., 2022; Ritala et al., 2017).

Regarding microbial doubling or generation time, a decreasing rate order of bacteria, fungi, and algae has been reported, making bacteria and yeast production comparably more yielding than algae (Abdullahi et al., 2021; Sharif et al., 2021; Thiviya et al., 2022). However, contrary to algae and filamentous fungi, harvesting bacterial cells is difficult, given their small size and low density. Therefore, expensive and sophisticated centrifugal technologies are required for optimal harvesting efficiency for bacterial SCP. These dynamics emphasize the need to consider microbial selection as a critical factor in SCP production. Table 2.1 summarizes some recently explored substrates and microbes with details of operational variables, biomass yield, and protein content.

2.2.3.4 Analysis of Compiled SCP Production Studies

To better understand the trends in the compiled studies presented in Table 2.1, further analysis was performed to ascertain variation in the average time, temperature, pH, and protein content of the microbes explored in these studies, as presented in Figure 2.3. The aggregation of results for fermentation conditions from the compiled studies showed that bacteria, on average, require higher pH (more alkaline conditions) than yeast and fungi, with yeast demonstrating the capacity to thrive and multiply under highly acidic pH, rightly aligning with the established dynamics in microbial characteristics. For average fermentation time and temperature, it was conspicuous that bacteria required shorter generation times and relatively low temperatures to reach optimal growth, corroborating existing trends. Subsequent studies should consider such dynamics in process design, optimization, and selection. Considering a linear correlation between product mass and protein content, the average protein content was approximately 48-50% for all microbes. However, Table 2.1 clearly distinguishes these numbers for substrate type and operational conditions. A distinguishing insight from Figure 2.3 regards the dearth of information regarding the use of bacteria in SSF relative to the compiled studies. As emphasized in the literature, SSF favors microbes that can thrive under low water activity or moisture content. Thus, it is understandable that bacteria that thrive mostly under high water activity have not been extensively explored using the SSF approach. This directs a preference for yeast and fungi in the evolving SSF approach for SCP production.

Table 2.1: Studies on the utilization of second-generation substrates

Substrate	Microbes	Microbial type	Operational variables			Experimental Setup	Media Enrichment	Yield	Protein content %	Reference
			Fermentation	Harvesting	Drying					
LIQUID STATE FERMENTATION										
Agro-industrial waste (Whey, Molasses, Potato pulp, Orange Pulp; 3:1:1:10)	*Kluyveromyces marxianus*	Yeast	Temp: 30°C Time: 96 h pH: 7 Airflow rate: 0.5 L/min Stirring speed: *	Centrifugation Speed: 5000rpm Time: 10 min	*	Lab scale: Erlenmeyer flask	NE	0.87gCWM/g	30.23	(Aggelopoulos et al., 2013; Aggelopoulos et al., 2014)
Agro-industrial waste ((Whey, Molasses, Potato pulp, Orange Pulp; 0:1:0:2)	*Saccharomyces cerevisiae*	Yeast	Temp: 30°C Time: 96 h pH: 5.5 Airflow rate: 0.5 L/min Stirring speed: *	Centrifugation Speed: 5000rpm Time: 10 min	*	Lab scale: Erlenmeyer flask	NE	0.80gCWM/g	23.58	(Aggelopoulos et al., 2013; Aggelopoulos et al., 2014)
Agro-industrial waste (Whey, Molasses, Potato pulp, Orange Pulp; 10:1:1:3)	*Kefir*	Yeast	Temp: 30°C Time: 96 h pH: 5.5 Airflow rate: 0.5 L/min Stirring speed: *	Centrifugation Speed: 5000rpm Time: 10 min	*	Lab scale: Erlenmeyer flask	NE	0.48gCWM/g	31.02	(Aggelopoulos et al., 2013; Aggelopoulos et al., 2014)
Corn stover effluent	*Rhodococcus opacus*	Bacteria	Temp: 28°C Time: 48 h Shaking speed: 150rpm pH: 7.0	Centrifugation Speed: 5000rpm Time: 10 min	Freeze drying	Lab scale: shaking flask	NH_4NO_3: 0.005 or NH_4Cl: 0.005	0.27-0.33 gCDM/100 ml	47.0-52.7	(Mahan et al., 2018)
Food waste: a mixture of fish waste (fish head, viscera, skin, and bones), pineapple, banana, apple, and citrus peels	*Saccharomyces cerevisiae*	Yeast	Temp: 30°C Time: 72-120 h pH: 4.5 Airflow rate: 0.5 L/min	*	Freeze drying: -18°C	5 L batch fermenter (Biostat Biotech B, Sartorius Stedim Biotech,	Urea phosphate salt: 2.3 g/L KCl: 0.2 g/L MgSO4·7H2O 3.8 g/L	*	34-42	(Tropea et al., 2022)

			Stirring speed: 300rpm			Goettingen, Germany)	Ca-pantothenate: 0.0833 mg/L Biotin 0.0833 mg/L.			
Lemon waste blend	*Rhodococcus opacus*	Bacteria	Temp: 28°C Time: 48 h Shaking speed: 150rpm pH: 7.0	Centrifugation Speed: 5000rpm Time: 10 min	Freeze drying	Lab scale: shaking flask	NH_4NO_3: 0.005 or NH_4Cl: 0.005	0.22-0.33 gCDM/100 ml	45.8-52.1	(Mahan et al., 2018)
Oat bran hydrolysate	*Candida tropicalis*	Yeast	Temp: 30°C Time: 72 h Shaking speed: 200rpm pH: 4.5-5.0	*	Temp: 105°C Time: Overnight	Lab scale: shaking flask	$(NH_4)_2SO_4$: 0.002 KH_2PO_4: 0.002 (g/L)	7.2 g/100 g	*	(Dimova et al., 2014)
	Candida utilis	Yeast	Temp: 30°C Time: 72 h Shaking speed: 200rpm pH: 4.5-5.0	*	Temp: 105°C Time: Overnight	Lab scale: shaking flask	$(NH_4)_2SO_4$: 0.002 KH_2PO_4: 0.002 (g/L)	7.9 g/100 g	*	(Dimova et al., 2014)
Orange peel	*Candida utilis*	Yeast	Temp: 30°C Time: 96 h Shaking speed: 150rpm pH: 3.7-4.2	Centrifugation Speed: 5000rpm Time: 10 min	Temp: 60°C Time: 24 h	Lab scale: shaking flask	$(NH_4)_2SO_4$: 1.5 KH_2PO_4: 0.75 K_2HPO_4: 0.75 $MgSO_4$: 0.05 (g/L)	1.65 g/100 ml	*	(Carranza-Méndez et al., 2022)
	Candida utilis	Yeast	Temp: 30°C Time: 96 h Shaking speed: 150rpm pH: 3.7-4.2	C *Candida utilis* centrifugation Speed: 5000rpm Time: 10 min	Temp: 60°C Time: 24 h	Lab scale: shaking flask	$(NH_4)_2SO_4$: 0.6 KH_2PO_4: 0.2 $FeSO_4$: 0.002 KCl: 0.8 $MgSO_4$: 0.07 (g/L)	1.03 g/100 ml	*	(Carranza-Méndez et al., 2022)
	Candida utilis	Yeast	Temp: 30°C Time: 96 h Shaking speed: 150rpm pH: 3.7-4.2	Centrifugation Speed: 5000rpm Time: 10 min	Temp: 60°C Time: 24 h	Lab scale: shaking flask	$(NH_4)_2SO_4$: 0.6 Na_2HPO4: 0.640 $FeCl_3$: 0.029 KH_2PO_4: 0.427 $FeSO_4$: 0.002 $CaCl_2$: 1.793 $MgSO_4$: 0.492	0.77 g/100 ml	*	(Carranza-Méndez et al., 2022)

							$CuSO_4$: 0.002 $MnSO_4$: 0.009 $ZnSO_4$: 0.011 (g/L)			
Orange waste blend (pulp, peel, and juice	*Rhodococcus opacus*	Bacteria	Temp: 28°C Time: 48 h Shaking speed: 150rpm pH: 7.0	Centrifugation Speed: 5000rpm Time: 10 min	Freeze drying	Lab scale: shaking flask	NH_4NO_3: 0.005 or NH_4Cl: 0.005	0.23-0.30 gCDM/100 ml	42.2-56.9	(Mahan et al., 2018)
Pineapple skin and rice washing water	*Saccharomyces cerevisiae*	Yeast	Temp: 30°C Time: 56 h Shaking speed: * pH: 3.8-4.5	*	*	Lab scale: Beaker	$(NH_4)_2SO_4$: 1.5 KH_2PO_4: 0.7 NaCl: 0.07 $MgSO_4$: 0.38 $CaCl_2$: 0.07 (g/L)	0.4752 gCDM/100 ml	*	(Mujdalipah & Putri, 2020)
Pineapple peel waste	*Saccharomyces cerevisiae*	Yeast	Temp: 29°C Time: 24-48 h Shaking speed: * pH: 4.5	Centrifugation speed: 3000rpm Time: 10 min	Temp: 60°C Time: 5 h	Lab scale: shaking flask	Fructose or Sucrose: *	0.72-0.84 g CDM/100 ml	65-94	(Nurmalasari & Maharani, 2020)
Potato peel extract and glucose	*Rhizopus oligosporus*	Filamentous fungus	Temp: 35°C Time: 72 h pH: 5.5	Whatmann filter	Temp: 80°C Time: 24 h	Lab scale: shaking flask	KH_2PO_4: * $MgSO_4$: * NaCl: * Yeast extract: *	0.52 gCDM/100 ml	45-55	(Nadeem, 2021)
	Rhizopus oligosporus	Filamentous fungus	Temp: 35°C Time: 72-96 h pH: 5.5 Airflow rate: 1.0vvm	Whatmann filter	Temp: 70-75°C Time: Until constant weight	Stirred tank bioreactor	KH_2PO_4: * $MgSO_4$: * NaCl: * Yeast extract: *	0.45 gCDM/100 ml	~50	(Nadeem, 2021)
	Rhizopus oligosporus	Filamentous fungus	Temp: 35°C Time: 72-96 h pH: 5.5 Airflow rate: 1.0vvm	Whatmann filter	Temp: 70-75°C Time: Until constant weight	Bubble column fermenter	KH_2PO_4: * $MgSO_4$: * NaCl: * Yeast extract: *	0.55 gCDM/100 ml	~50	(Nadeem, 2021)
Rice husk hydrolysate	*Candida tropicalis*	Yeast	Temp: 30°C Time: 72 h	*	Temp: 105°C Time: Overnight	Lab scale: shaking flask	$(NH_4)_2SO_4$: 0.002 KH_2PO_4: 0.002	4.7 g/100 g	*	(Dimova et al., 2014)

			Shaking speed: 200rpm pH: 4.5-5.0				(g/L)			
	Candida utilis	Yeast	Temp: 30°C Time: 72 h Shaking speed: 200rpm pH: 4.5-5.0	*	Temp: 105°C Time: Overnight	Lab scale: shaking flask	$(NH_4)_2SO_4$: 0.002 KH_2PO_4: 0.002 (g/L)	5.8 g/100 g	*	(Dimova et al., 2014)
Wasted Date Molasses (WDM)	*Hanseniaspora guilliermondii*	Yeast	Temp: 30°C Time: 48 h Agitator speed: 150rpm pH: 4.0	*	Temp: 80°C Time: 24 h	Lab scale: shaking flask	Peptone: 4.0 (g/L)	55.30 gCDM/100 g WDM	52.0	(Hashem et al., 2022)
	Hanseniaspora guilliermondii	Yeast	Temp: 30°C Time: 48 h Agitator speed: 150rpm pH: 4.0 Airflow rate: 0.25 vvm	*	Temp: 80°C Time: 24 h	Bioreactor BioFlo/CelliGen 115 (7 L capacity)	Peptone: 4.0 (g/L)	55.82 gCDM/100 g WDM	53.21	(Hashem et al., 2022)
Wasted Date Molasses FT: LSF	*Hanseniaspora uvarum*	Yeast	Temp: 30°C Time: 48 h Agitator speed: 150rpm pH: 4.0	*	Temp: 80°C Time: 24 h	Lab scale: shaking flask	Peptone: 4.0 (g/L)	44.89 gCDM/100 g WDM	50.0	(Hashem et al., 2022)
	Hanseniaspora uvarum	Yeast	Temp: 30°C Time: 48 h Agitator speed: 150rpm pH: 4.0 Airflow rate: 0.25 vvm	*	Temp: 80°C Time: 24 h	Bioreactor BioFlo/CelliGen 115 (7 L capacity)	Peptone: 4.0 (g/L)	47.53 gCDM/100 g WDM	51.53	(Hashem et al., 2022)
Wasted Date Molasses	*Issatchenkia orientalis*	Yeast	Temp: 30°C Time: 48 h	*	Temp: 80°C Time: 24 h	Lab scale: shaking flask	Peptone: 4.0 (g/L)	75.00 gCDM/100 g WDM	54.45	(Hashem et al., 2022)

			Agitator speed: 150rpm pH: 4.0							
	Issatchenkia orientalis	Yeast	Temp: 30°C Time: 48 h Agitator speed: 150rpm pH: 4.0 Airflow rate: 0.25 vvm	*	Temp: 80°C Time: 24 h	Bioreactor BioFlo/CelliGen 115 (7 L capacity)	Peptone: 4.0 (g/L)	75.82 gCDM/100 g WDM	54.34	(Hashem et al., 2022)
Wasted Date Molasses	*Cyberlindnera fabianii*	Yeast	Temp: 30°C Time: 48 h Agitator speed: 150rpm pH: 4.0	*	Temp: 80°C Time: 24 h	Lab scale: shaking flask	Peptone: 4.0 (g/L)	46.90 gCDM/100 g WDM	45.0	(Hashem et al., 2022)
	Cyberlindnera fabianii	Yeast	Temp: 30°C Time: 48 h Agitator speed: 150rpm pH: 4.0 Airflow rate: 0.25 vvm	*	Temp: 80°C Time: 24 h	Bioreactor BioFlo/CelliGen 115 (7 L capacity)	Peptone: 4.0 (g/L)	47.53 gCDM/100 g WDM	48.72	(Hashem et al., 2022)
Wheat bran hydrolysate	*Candida tropicalis*	Yeast	Temp: 30°C Time: 72 h Shaking speed: 200rpm pH: 4.5-5.0	*	Temp: 105°C Time: Overnight	Lab scale: shaking flask	$(NH_4)_2SO_4$: 0.002 KH_2PO_4: 0.002 (g/L)	7.9 g/100 g	*	(Dimova et al., 2014)
	Candida utilis	Yeast	Temp: 30°C Time: 72 h Shaking speed: 200rpm pH: 4.5-5.0	*	Temp: 105°C Time: Overnight	Lab scale: shaking flask	$(NH_4)_2SO_4$: 0.002 KH_2PO_4: 0.002 (g/L)	8.6 g/100 g	*	(Dimova et al., 2014)
Yam starch	*Yeast*	Yeast	Temp: 28.5°C Time: 60 h Shaking speed: 200rpm pH: 4.5	Centrifugation Speed: 3500rpm Time: 10 min	*	Lab scale: shaking flask	*	241.54±0.15 g wet weight/100 g dry starch	*	(Chen et al., 2016)

SOLID STATE FERMENTATION										
Agro-industrial waste (Whey, Molasses, Potato pulp, Orange Pulp)	*Pleorotus ostreatus*	Filamentous fungus	Temp: 25°C Time: 120-168 h pH: 4.0-7 Airflow rate: 0.5 L/min Stirring speed: 300rpm	*	*	Lab scale: Petri dish	NE	3.95-5.94 g/100 g	27.96-38.35	(Aggelopoulos et al., 2018)
Agro-industrial waste (Whey, Molasses, Potato pulp, Orange Pulp; BSG, MSR; 3:1:1:10:0:8)	*Kluyveromyces marxianus*	Yeast	Temp: 30°C Time: 96 h pH: 7	Centrifugation Speed: 5000rpm Time: 10 min	*	Lab scale: Petri dish	NE	0.21gCWM/g	*	(Aggelopoulos et al., 2013; Aggelopoulos et al., 2014)
Agro-industrial waste (Whey, Molasses, Potato pulp, Orange Pulp; BSG, MSR; 10:1:1:3:2.5:6)	Kefir	Yeast	Temp: 30°C Time: 96 h pH: 5.5	Centrifugation Speed: 5000rpm Time: 10 min	*	Lab scale: Petri dish	NE	0.10gCWM/g	*	(Aggelopoulos et al., 2013; Aggelopoulos et al., 2014)
Agro-industrial waste (Whey, Molasses, Potato pulp, Orange Pulp; BSG, MSR; 10:1:1:3:2.5:6)	*Saccharomyces cerevisiae*	Yeast	Temp: 30°C Time: 96 h pH: 5.5	Centrifugation Speed: 5000rpm Time: 10 min	*	Lab scale: Petri dish	NE	0.26gCWM/g	*	(Aggelopoulos et al., 2013; Aggelopoulos et al., 2014)
Cashew bagasse	*Saccharomyces cerevisiae*	Yeast	Temp: 30°C Time: 9 h pH: 4.2 Setup: Lab scale (Tray type)	*	Temp: 55°C Time: Until constant weight	Lab scale: Tray	NE	11.1 gCDM/100 g	15.8	(Muniz et al., 2020)
Guava peels	*Saccharomyces cerevisiae*	Yeast	Temp: 30°C Time: 9 h pH: 3.6	*	Temp: 55°C Time: Until constant weight	Lab scale: Tray	NE	11.3 gCDM/100 g	28.1	(Muniz et al., 2020)

Rice straw pulp	*Trichoderma reesei*	Filamentous fungus	Temp: 30°C Time: 288 h pH: 5.0	*	*	Lab scale: flask	NE	*	19.71	(Novita et al., 2019)
Wheat bran	*Candida utilis*	Yeast	Temp: 30°C Time: 96 h pH: 6.5	*	*	Lab scale: Erlenmeyer flask	KH_2PO_4: 0.25 $MgSO_4$: 0.05 Soluble starch: 0.5 Peptone: 0.25 (NH_4NO_3: 0.25 (g/L)	*	48.01	(Irfan et al., 2011)
Wheat bran	*Rhizopus oligosporus and Candida utilis*	Mixed culture	Temp: 28°C Time: 48 h pH: 3.5	*	*	Lab scale: Freehold plastic bag	$(NH_4)_2SO_4$: 40 g/kg Chloramphenicol: 0.4 g/kg	*	41.02	(Yunus et al., 2015)
Yam peel mash	*Aspergillus niger*	Filamentous fungus	Temp: 28°C Time: 168 h pH: 3.5	*	*	Lab scale: Erlenmeyer flask	$(NH_4)_2SO_4$: 2.0 g/L	*	16.78	(Akintomide & Antai, 2012)
	Saccharomyces cerevisiae	Yeast	Temp: 28°C Time: 168 h pH: 3.5	*	*	Lab scale: Erlenmeyer flask	$(NH_4)_2SO_4$: 2.0 g/L	*	21.30	(Akintomide & Antai, 2012)

Legend: CDM – Cell dry mass, NE: No enrichment, *: Not specified, Temp: Temperatur

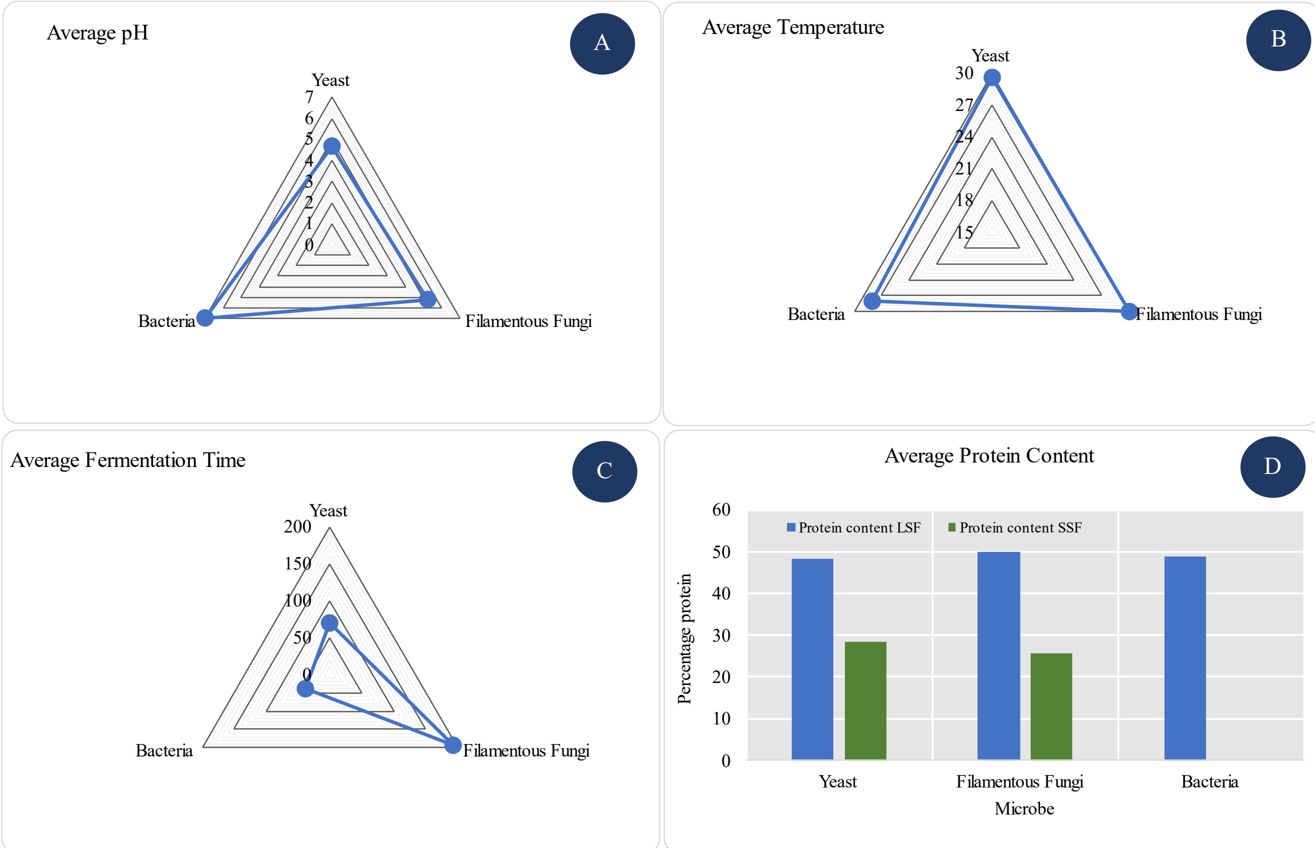

Figure 2.3: Dynamics in SCP production for different microbial types: Average fermentation conditions and protein content

2.2.3.5 Mixed Culture Biotechnology

Microbes' cultivation rate and performance in SCP production depend on their ability to thrive in a dynamic environment of varying pH, temperature, substrate composition, and toxicity, among other unseen kinetics. Most microbes die out and fail to provide the required output when process conditions become unfavorable. The evolution of mixed culture biotechnology is rising as a solution. In mixed culture technology, different microbes synergize in a fermentation environment to overcome antagonism or recalcitrance and yield desired process outputs (Bajpai, 2017). Several reports have highlighted the benefits of mixed culture, or coculturing, in SCP production. Using Kluyveromyces marxianus and Candida kusei on whey could alter the removal efficiency of chemical oxygen demand (COD) to an optimal level while minimizing the susceptibility of media to contamination (Bratosin et al., 2021). The outcome was an improved production rate with enhanced SCP quality. A synergistic association was also identified when chemoorganoheterotrophic bacteria were cocultured with purple nonsulfur bacteria (PNSB). The former facilitated the fermentative breakdown of sugar into volatile fatty acids and alcohols, serving as organic carbon sources for the latter (Wada et al., 2022). Similarly, coculturing prominent fungal strains such as Aspergillus niger and Saccharomyces cerevisiae rendered a desirable synergy. Herein, the former facilitated an enzymatic breakdown of cellulose in fruit peels into fermentable sugars, and the latter utilized the product as a carbon source for growth (Thiviya et al., 2022). The mixed culture approach is noted for its cost-saving benefits in SCP production, considering compensations for cost and energy-intensive processes such as sterilization offered by mixed-microbial culture (Sakarika et al., 2022). It is important to emphasize that the intent of mixed culture biotechnology is not necessarily to displace pure culturing in SCP production. Instead, it is designed to provide a reliable and sustainable alternative to overcome avoidable challenges, such as antagonism in producing SCP from some microbial strains, especially when a symbiotic association is beneficial. Mixed culture technology is still in its infancy and has not been widely explored for most SGSs in SCP production. This could be an interesting area to focus future SCP research for improved performance.

2.3 Commercial Status of SCP

There is a generic accession to the significant roles that incorporating SCP into the protein supply and consumption chain would play. The rich protein composition of SCP biomass (up to 80%) can contribute significantly to addressing the ever-increasing protein demand amidst global population growth. Others, specifically sustainability enthusiasts, recommend SCP for commerce mainly because of the environmental and economic benefits they confer through the circularization of cheaper waste resources and value recovery from renewable substrates like sunlight and carbon capture. In this section, we summarize the commercial status of SCPs, starting with what benefits are triggering commercial potentials, then a brief outlook of the current and future market, and a final discussion on the current and emerging applications.

2.3.1 Benefits to Commercial Entry

Researchers and other food system stakeholders have embraced confidence in SCP's capacity to complement nutritional needs and reinforce regenerative economic models when given a commercial space (Durkin et al., 2022). Regarding nutritional capacity, appreciable protein content (about 30-80%), including limiting amino acids like methionine, lysine, threonine, and cysteine, have been characterized for SCPs, emphasizing their potential to serve the needs of the rising protein consumer market. These have given SCPs recognition from international organizations like the Food and Agriculture Organization (FAO) and the National Aeronautics and Space Administration (NASA) (Altmann & Rosenau, 2022; García Martínez et al., 2022). For instance, FAO deems Aspergillus oryzae a well-balanced protein source (García Martínez et al., 2022; Ritala et al., 2017). NASA has already employed Spirulina as a complete protein food for astronauts during space missions, illustrating an emerging exploration of SCP in space foods. Aside from proteins, SCPs are packed with significant amounts of carbohydrates, lipids, dietary fiber, minerals, and vitamins (Can Karaca et al., 2022; Pereira et al., 2022; Thiviya et al., 2022). They also contain considerable amounts of essential fatty acids like eicosapentaenoic (EPA) and docosahexaenoic acid (DHA), linolenic acid, and palmitic acid that have been quantified in Spirulina and Chlorella SCPs (Gogna et al., 2022; Ragaza et al., 2020), and substantial quantities of fat-soluble vitamins like A, D, E, K (Alagawany et al., 2021; Gogna et al., 2022; Pereira et al., 2022) and vitamin B-complex (Altmann & Rosenau, 2022; Can Karaca et al., 2022; Jach et al., 2022; Türker et al., 2022). Moreover, SCPs are also loaded with trace minerals like phosphorus, calcium, sodium, magnesium, and manganese (Gogna et al., 2022; Jach et al., 2022) and contain appreciable quantities of bioactive compounds like carotenoids and chlorophyll A (Barka & Blecker, 2016; Carter & Codabaccus, 2022; Gogna et al., 2022) providing potential therapeutic or nutraceutical capacities.

The availability of inexpensive second-generation feedstock for SCP production makes it economically attractive (Elyasi et al., 2021; Matassa et al., 2020), which explains the increasing exploration of fruit and vegetable waste, lignocellulosic materials, and industrial wastewaters as substrates in production at different scalar levels (Pereira et al., 2022; Rajendran et al., 2018; Raziq, 2020). Contrary to conventional animal and plant protein production, the synergy of shorter generation time and high doubling rate of microbes makes SCP production highly efficient and relatively profitable. Reports establish that for the same fold of land, the caloric and protein yield of SCPs could be ten-fold and two-fold, respectively, higher than that of protein-rich pulses, meats, and grains like millet and wheat (Bajpai, 2017; Leger et al., 2021). This presents SCP as a scalable technology solution to a sustainable protein supply and accentuates the economic benefits accompanying SCP commerce. The following subsection briefly captures current commercial engagements in the SCP arena.

2.3.2 Market Dynamics

2.3.2.1 Current and Projected Market Value

We delineated some significant benefits of accelerating SCP commercialization in the previous subsection. Here, we elaborate on the current SCP market size while highlighting projections of its market value in the near decade. A significantly growing market and consumer base have been identified for SCP driven by the increasing demand for sustainable protein alternatives, resurging interest in biotechnological technologies, and the expanding scope of its food and feed applications (Global Market Insights, 2023; Market Research Intellect, 2022). The industry was valued at USD 8 bn in 2021, according to a Global Market Insight survey involving 21 countries across 5 continents. The industry's value is anticipated to surpass USD 18.5-18.8 bn by 2030 and USD 20.64 bn by 2032 at an estimated compound annual growth rate (CAGR) of 9-9.7 % (Global Market Insights, 2023; Transparency Market Research, 2023). A country-wise analysis shows dramatic growth in Malaysian and Vietnam SCP markets, with 2020 market values of USD 9.7 mn and USD 26.7 mn, respectively. These values are projected to go beyond USD 24.5 mn and USD 69.4 mn, respectively, by 2030, and rapidly rising markets in China, the US, and parts of Western Europe (Transparency Market Research, 2023). Regional insights also present a tremendous boom in SCP on the European market because of the increased pressure for feed protein supply. The current market is valued at USD 1.11 bn, representing about 30 % of the 2023 global market share. The European market could surpass USD 4.5 bn by 2030 if expectations are backed with innovations, technological development, and advanced research (Global Market Insights, 2023). North America is also performing well, dominated by the US, with a regional market share of about 92 %, also expected to grow at a CAGR of over 5 % within 2022-2030 (Global Market Insights, 2022; Persistence Market Research, 2023).

Regarding the microbial category, algae and fungi are leading global commerce, adding up to 60 % of total microbial protein extraction. Algal SCPs are valued at USD 1.41 bn, representing approximately 38 % of the global SCP market value (Persistence Market Research, 2023). Production volume is expected to surpass 410 kt by 2030 due to booming private and public sector interest (Global Market Insights, 2023). A segmentation of SCP application shows a 2.7 %, 3 %, 4 %, and 6 % rise in food, dietary supplement, cosmetic, and animal feed applications, respectively (Maximize Market Research, 2023). These trends are expected to continue rising due to burgeoning industrial and research interest, technological innovations, and consumer demand (Global Market Insights, 2022). However, achieving this perceived growth would require radical collaboration between governments, industry, and researchers. Such collaboration could be linearized as willingness on the part of governments to fund industries or start-ups that are interested in SCP production and efforts by these industries or start-ups to engage experts and researchers in finding sustainability-sensitive technological solutions.

2.3.2.2 Global Distribution of SCP Businesses

To demonstrate the spatial dispersion of the SCP market consequent to the increasing commercial recognition, a global-wide literature search was undertaken to consolidate a list of start-ups and industries engaged in the SCP business. The global distribution of existing businesses is presented in Figure 2.4, while a list of SCP businesses can be found in the supporting information (SM.2.A). Industries and start-ups interested in SCP production are primarily dense in the US and China, with a fair distribution across Europe, especially in Germany, France, the UK, and the Netherlands. Countries in Africa, Southern America, and most parts of Europe seem to lag in adoption per the data accumulated. However, some reports signal a slowly emerging interest in these areas, particularly in South Africa, Nigeria, Brazil, and Argentina (Maximize Market Research, 2023). Being no exception in waste generation, rapid adoption into their food value chain would be a reliable step to creating circular agri-business models and advancing sustainability in their food and agricultural systems.

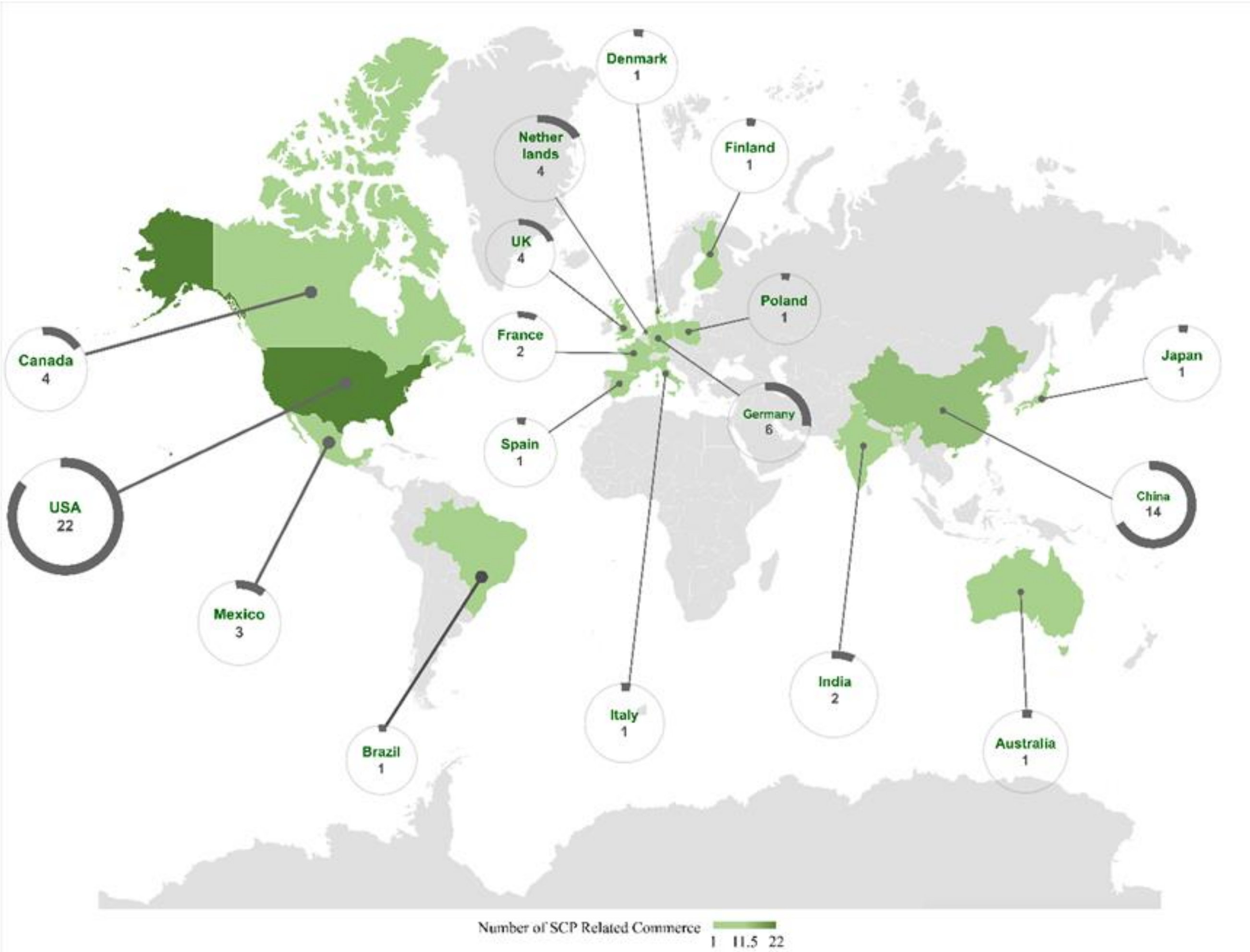

Figure 2.4: Global distribution of SCP companies

2.3.3 Current and Evolving Applications

2.3.3.1 Feed and Food Applications

There is a rapidly growing exploration of different varieties of SCP in feed production. To some, utilizing SCP in animal production is a suitable nutritional alternative and cost relief to farmers, given its protein adequacy and affordability. Additionally, its low requirement for land space situates its ability to decouple animal production from huge pastureland requirements, further triggering interest in feed production. SCPs have been used to produce feed or feed supplements for pets, swine, cattle, poultry, and aquaculture (Altmann & Rosenau, 2022; Global Market Insights, 2022; Persistence Market Research, 2023). Its expanding application in aquafeed production is particularly fascinating, catalyzed by the exponentially growing aquaculture industry and associated increase in demand for high quality-low cost protein feed (Owsianiak et al., 2022; Ragaza et al., 2020; Transparency Market Research, 2023; Yang et al., 2021). Recent exploration has realized successful and beneficial substitution of aquafeed ingredients such as fish meal and soybean meal with SCPs such as brewer's yeast (Guo et al., 2019a; Jin et al., 2018), yeast hydrolysate (Jin et al., 2018), and bacterial strains such as *Corynebacterium ammoniagenes* (Hamidoghli et al., 2018) for shrimp, salmon, trout, and carp production. Nonetheless, these studies correlate SCP's best substitutional benefits to using optimal proportions in feed rations (Guo et al., 2019a; Guo et al., 2019b; Hamidoghli et al., 2018).

Utilization in food formulations is gradually dispersing across the food and beverage industry (Banks et al., 2022; Bratosin et al., 2021), with adoption in meat analogs, bakery, dietary supplements, dairy alternatives, cereals, snacks, and beverages dominating the current food application trend (Global Market Insights, 2022; Persistence Market Research, 2023). For instance, given its remarkable functional, nutritive, and radical scavenging capacities, Razzaq et al. (2020) inferred its suitability in food emulsions, minced meat, baked foods, and frozen desserts for improving product characteristics and health benefits. In this regard, SCP from food waste (banana peel, citrus peel, potato peel, and carrot pomace) has been successfully utilized in breadmaking for improving dough characteristics and enhancing the nutritional benefits of the resulting bread, wherein 4% addition was identified as the optimal concentration for achieving desirable functionalities (Khan et al., 2022). The North American and ASEAN (Association of South East Asian Nations) markets are constantly innovating ways to ameliorate algae (*Spirulina and Chlorella*) and fungi (*Fusarium*) production as a "superfood" to enhance regional commitment to convenient and healthy living (Transparency Market Research, 2023). Additionally, Health Canada has permitted and is keenly regulating the use of whole algal SCP as an alternative protein source in foods, signaling a future boom in food application (Global Market Insights, 2022). Nonetheless, the application of SCP in food and feed applications is limited by reported toxicity and metabolic constraints, elaborated upon in the following subsections.

a. Nucleic acid and Toxicological Limitations

The toxicological status of products is necessary for approving their use in food and feed. Despite the emerging interest in food and feed application, the presence of undesirable amounts of nucleic acid in most SCP has limited acceptance as a food and food ingredient, given the health risks associated with a high intake of nucleic acid (Bajic et al., 2022; Carter & Codabaccus, 2022). Nucleic acid is synthesized into uric acid in humans, an undesirable chemical that instigates vulnerability to health detriments such as carcinogenesis, urinary diseases, and renal diseases such as renal calculus and gout – rich man's disease (Ugbogu & Ugbogu, 2016). Unfortunately, humans lack uricase, a uric acid-degrading enzyme; therefore, they are at risk of uric acid bioaccumulation upon consumption of SCP (Nangul & Bhatia, 2013; Ritala et al., 2017). At undesirable levels, uric acid instigates vulnerability to health detriments such as carcinogenesis, urinary diseases, and renal diseases such as renal calculus and gout – rich man's disease, making SCP particularly unattractive for human consumption and sometimes for feed application (Ugbogu & Ugbogu, 2016). Nonetheless, techniques have evolved to reduce the nucleic acid content of SCP to the fairest and safest minimum, implicating the potential to spur its adoption as a food and feed ingredient (García Martínez et al., 2022; Ritala et al., 2017). Additionally, the toxicological orientation of some microbes has presented limitations in leveraging SCPs as human food. Gram-negative bacteria and filamentous fungi can produce endotoxins and mycotoxins, respectively, which attaches safety risks to SCPs from these microbes and decreases their suitability and acceptability for human consumption (Ugbogu & Ugbogu, 2016). In subsequent developments, scientists proposed process optimization and deployment of aseptic techniques to overcome such toxicological impediments (García Martínez et al., 2022).

b. Metabolic constraints

Concerns about the metabolic biochemistry of food and feed materials have elicited remarkable research interest. In this space, contemporary nutrition science and gastroenterological studies have aimed to understand the correlation between the nutritional properties of materials or products and digestive performance. The outcome of these studies has supported the prominent claim that the "classification of material or product as nutritious hinging on the in vitro quantification of their nutritional compositions is erroneous." They do not argue the relevance of assessing the nutritional content. However, they prioritize the digestive chemistry of a food or feed material/product as the "main deal" in dietary applications. This context, coupled with surging interest in leveraging SCP in food and feed applications to augment protein supply, has inspired several studies that sought to understand the digestibility of SCP. Existing results emphasize that SCP from algal and some bacterial cells have lower digestibility, induced by poorly digestible cell walls, making it difficult for humans to access and metabolize the available proteins and nutrients (Nasseri et al., 2011; Ritala et al., 2017; Ugbogu & Ugbogu, 2016). Consequently, there has been a drastic contention against using bacteria-based SCP as human food. If this persists, the current prospection of these SCPs in augmenting future protein demand and food shock will contradict expectations despite the evolving development and innovations in their processes and production. Fortunately, several methods have been developed for SCP cell wall degradation, with varying efficiencies and desirable outcomes, highlighting substantial progress in surmounting the present constraint.

2.3.3.2 Non-Food Applications

Aside from the overarching interest in SCP for food and feed, the unique composition of biobased coproducts like polyhydroxyalkanoates (PHA) for some bacterial species bolstered interest in biopolymeric materials production (Kunasundari et al., 2013). The major advantage of this pathway is the possibility of requiring little to no strong chemicals in biopolymer synthesis while co-benefiting access to microbial protein to satisfy protein needs. For instance, in an animal-based model for SCP production, it was possible to recover indigestible polyhydroxybutyrate (PHB) - a useful biopolymer in the bio-revolution of bioplastic films and packages, from the fecal matter of rats fed with Cupriavidus necator SCP, requiring no strong chemical in the extraction process (Chee et al., 2019; Kunasundari et al., 2013). In this regard, SCP stands as one of the evolving technology solutions for addressing the current climate emergency, amongst other environmental burdens accompanying the production of synthetic plastics, while providing sustainable plastic alternatives with desired mechanical, thermal, and biodegradable qualities (Areniello et al., 2022). However, the high cost of producing SCP-based biopolymers is stalling the scalar adoption of this procedure, requiring advanced research to improve current techniques (Kunasundari et al., 2013).

The agricultural industry has also found the application of SCP as an organic fertilizer for recovering the soil quality of farmlands (Kantachote et al., 2016). Microbes have been grown on agricultural biomass to recover essential nutrients like nitrogen, phosphorus, and potassium, which are later applied to farm soils for bio-enrichment and consequent improvement of plant growth (Areniello et al., 2022). In practice, less demand for operational conditions like drying and the possibility of using multiple substrates make the process cost-efficient. However, biofertilizers' current market value seems to curtail the potential for expanding production and verging into commercial sales (Areniello et al., 2022). Research is therefore needed to understand current limitations and uncover opportunities for commercialization.

Overall, SCP is expanding in application, and the influx of novel and innovative technologies instigated by extensive research and development, promises an opportunity to expand further and exhaust the vast potential in the SCP industry.

2.4 Sustainability Outlook of SCP Production

The previous sections have established SCP as a rapidly evolving biotechnological solution to biomass conversion, underpinning its production status to meet growing protein demand and commercial prospects. In this section, we delve into understanding the sustainability outlook of SCP production, highlighting the application of sustainability metrics and findings for further improvement. A thorough literature search on Google Scholar, Web of Science, and Scopus using query words that combine selected sustainability metrics and microbial protein or SCP showed that LCA and TEA had been extensively used in SCP sustainability assessment. However, other sustainability metrics, like LCCA, SLCA, and Environmental Nutrition, have not been investigated. This section is divided into three parts. The first subsection summarizes available LCA, including their methodological structures, significant findings, and meaningful recommendations. It also discusses the sustainability prospect of a novel power-to-food (PtF) technology in SCP production. The second subsection discusses relevant TEA studies, and the third subsection elaborates on current gaps and recommendations for future studies.

2.4.1 LCA

2.4.1.1 Summary of LCA Studies

Table 2.2 summarizes relevant LCA studies gathered from the literature. It captures the scope, methodologies, findings, gaps, and or recommendations from these studies.

Table 2.2: Summary of SCP-LCA Studies

Scope of Study	Methodological Structure			Major Findings	Gaps/Recommendations	Reference
	Functional Unit	System Boundary	Inventory Data/Modelling			
Empirical attributional LCA assessment of SCP production with expanded system boundary and impact categories Microbe: Hydrogen-oxidizing bacteria (HOB) Fermentation Approach: LSF	1 kg of MP product prior to packing with a 5% moisture content at the factory gate	Cradle-to-gate with scenarios for electricity consumption created for sensitivity analysis.	Pilot scale production data (Plant area was 1580 m^2, and by-products were cut-off in the analysis) Database: Ecoinvent 3 Impact method: ReCiPe 2016 v1.1 Midpoint (H), AWARE (water use), and CED v1.11 (energy use) Sofware: SimaPro 9.1.0.11 Secondary analysis: Sensitivity and Uncertainty Analysis	• Consuming SCP (65% protein assumed) instead of the dairy herd or bovine meat would offset an average of **16 m^2** and **36 m^2** of LU per 100 g of protein, respectively. • Electricity consumption contributed the most to all impact categories. • Hydropower could offset up to **87.5%** GWP and about **25 times less** LU relative to electricity mix with a high percentage of nuclear power	• Consequential LCA of SCP production to ascertain expected changes when SCP is commercialized. • Expand impact categories to include others like biodiversity, which closely correlates with conventional protein production	(Jarvio et al., 2021)
Consequential LCA of different biorefinery pathways using a mixture of organic fractions of municipal waste and supermarket waste as substrates to identify the most sustainable valorization pathway. Substrate: Organic Fraction of Municipal Waste and Supermarket Waste Microbe: Methane-oxidizing microbe	Management of 1 tonne of biopulp with an average TS of 18.3 %	Gate-to-gate	Lab-scale experiments Database: Ecoinvent (v3.3) Impact method: Impact 2002 + Software: SimaPro 8.5 Secondary analysis: Sensitivity	• SCP-based pathways could save up to **155 kg CO_2 eq per tonne** of biopulp compared to conventional protein sources like fish, soybean, and palm kernel meals. • Renewable energy options increased environmental savings of design pathways. • The environmental benefit from each scenario depends not only on the biorefining pathway but also on the selected downstream process	• Employ multi-criteria decision support tools for SCP sustainability assessment considering energy performance, economics, environmental, consumer, and regional legislations.	(Khoshnevisan et al., 2020) (Elyasi et al., 2021)
Attributional LCA to assess the environmental impact of SCP from oat-side stream.	1 kg of dried SCP product	Cradle-to-gate	Experimental data, literature, and technical reports	• Impact contribution was based on the nature of substrates, with wet-side streams offering better impacts than the dried-side stream.	• Genetic modification of yeast for improving biomass yield, generation time, substrate use efficiency, and nutritional value.	(Kobayashi et al., 2023)

Substrate: Oat-side stream			Database: Ecoinvent 3.8, Agribalyse 3, Agri-footprint 5.0). Impact method: ReCiPe 2016 Midpoint (H) Software: SimaPro 9.3.0.2 Secondary analysis: Sensitivity	• The dried-side stream had **about 8 and 18 %** increases in FC and GWP values relative to the wet-side stream • Regional sensitivity demonstrated fossil energy-dense regions to contribute highly to most impact categories. • About **61 %** LU offset was achieved for SCP relative to soy protein concentrates	• Integration of renewable energy systems to enhance the environmental savings of SCP production systems	
Comparative study of an optimized closed-loop mycoprotein framework with animal-based proteins Microbe: Fusarium venenatum	kg of microbial protein	Cradle-to-gate	The average global profile of Quorn™ fermentation process data and global average feedstock and energy profiling Impact Method: ReCiPe Midpoint (H) Software: OpenLCA	• Closed-loop SCP system offsets up to **96%, 99%,** and **85%** impact values of CC, LU, and WC, respectively, relative to beef. • Substituting future beef consumption with an equivalent quantity of SCP would reduce the impact on CC and LU from **45** and **24%** to **2** and **0.2%,** respectively	• Further optimization of microbial protein systems to provide more environmentally sustainable and scalable SCP technology solutions to meet future protein demands	(Durkin et al., 2022)
A static LCA approach for future projections of the environmental impacts of substituting ruminant meat with sugar-based SCP using spatially explicit land-use model MAgPIE Substrate: Sugar	Per capita replacement of ruminant meat with SCP in forward-looking land-use scenario	Cradle-to-gate (A middle-of-the-road scenario for future population, income, and food demand)	Literature data Impact Model: Model of Agricultural Production and its Impact on the Environment (MAgPIE)	• Global forest loss based on the current agricultural system is estimated at 175 Mha by 2050. • **20, 50, and 80%** per capita substitution can offset **56, 82, and 93%** of deforestation; **56, 83, and 87%** of net carbon dioxide emissions; and **11, 26, and 39%** of methane emission by 2050, respectively	• Include consequences of reducing the production of commercially viable ruminant production by-products like hide skin, fats, organs, bones, and blood in LCA. • Precision fermentation as a future technology for promoting alternative protein production	(Humpenoder et al., 2022)
A life cycle assessment to compare four food waste management scenarios. Substrate: Food waste Microbe: Purple non-sulphur bacteria (PNSB) Fermentation approach: LSF	Food waste produced by a city of 50,000 people per day at 0.31 kg FW/day/per person	Gate-to-gate (Waste collection to manufacturing gate)	Literature data Database: Ecoinvent 3.1, LCA Food DK Impact method: TRACI Software: SimaPro Secondary analysis: Sensitivity and Uncertainty Analysis	• The environmental benefits associated with SCP are based on the product it is replacing. • Replacing soybean with PNSB production presents better environmental compensations than fishmeal replacement.	• Future PNSB technology could be improved by optimizing growth rate, organic loading, and degree of substitution	(LaTurner et al., 2020)
Quantify the relative and absolute environmental performance of a pilot-scale SCP production from starch-rich process water and use it as feed relative to conventional feed sources. Substrate: Starch-rich potato process water Microbe: Aerobic heterotrophs Fermentation approach: LSF	Provision of nutritional value to edible white leg shrimp (Litopenaeus vannamei) required to produce 1 tonne per year of shrimps in an Intensive aquaculture production system at a feed conversion ratio between 1.2 and	Gate-to-gate (From the supply of substrate to the management of aquaculture biowaste)	Pilot scale and flowsheet simulation data Impact method: Multiplying elementary flows by characterization factors and summing resulting indicator scores (Compared with ReCiPe 2016 Midpoint (H) Software: SimaPro 9.2.0.2 Secondary analysis: Sensitivity and uncertainty	• The environmental impact of SCP-based feed depends on the substitution level and the type of meal being replaced. • SCP feed outperformed soybean meal in terms of GW and LU • Greener energy modeling is not sufficient to make SCP sustainable in absolute terms	• Using the bioreactor off-gas as the carbon source for hydrogen oxidizing bacteria, purple phototrophic bacteria, or green microalgae SCP feed would be a more sustainable technological alternative to improving SCP resource use efficiency.	(Owsianiak et al., 2022)

	1.8 and yield of at least 61 t/ha of pond					
Attributional life cycle assessment is used to compare the environmental impact of replacing soy ingredients with SCP in salmon feed. Microbes: Methanotrophic bacteria and Yeast Substrates: Fossil methane (Bacteria), Wheat by-product (Yeast)	Per 660 g of protein (1kg of soy protein, 0.94 kg of bacteria meal, and 1.07 kg of yeast protein concentrate)	Cradle-to-gate	Literature data, Norwegian imports data Impact method: ReCiPe (v.1.11) Impact calculation: According to Pauly and Christiansen's (1995) equation	• Yeast SCP had the lowest impacts in all categories and overall. • Bacteria SCP had similar CC and FWC impact results like soy protein but performed moderately in other categories. • Overall, replacing soy protein with bacterial or yeast SCP can significantly reduce environmental impacts	• Using diverted methane instead of natural gas for bacteria production can substantially offset environmental impacts. • Industrial-scale production developments are required to improve the benefits of SCP in aquaculture	(Couture et al., 2019)
To assess the environmental sustainability of the lignocellulosic SCP and compare it with food-derived SCP and conventional proteins Microbe: Fusarium venenatum Substrate: Rice straw	1 kg mycoprotein paste at biorefinery gate, with a solids content of 25% based on a production capacity of 40 000 tonnes per year	Cradle-to-factory gate	Field and literature data Impact Method: ReCiPe 2016 Midpoint (H) Software: SimaPro V9 Secondary analysis: Contributional analysis, Sensitivity analysis	• External electricity use and production represents **72.9 % and 58.4 %** of GWP and TA • Straw production contributes about 75 % and **67.58 %, and 95 %** of WC, ME, and LU • Cutting-off emission from rice production reduces SCP emissions substantially	• Integrating renewable energy resources into lignocellulosic SCP would be a reliable decarbonization solution. • SCP could be a transformative solution to future protein security due to manufacturing in a controlled environment and short generation time.	(Upcraft et al., 2021)
To examine the environmental implications of replacing soybeans with novel ingredients in chicken feed formulations Microbes: Yeast, Bacteria	One bird grown to a live weight of 2.2 kg	Gate-to-gate	Literature data Database: Agri-footprint, Ecoinvent Impact method: ReCiPe Software: SimaPro Secondary analysis: Comparative, Sensitivity, and Uncertainty analyses	• Substituting soybean meal with yeast SCP possess environment and nutrition co-benefits. • Replacing soybean meal with yeast SCP could offset **55 % and 32 %** of GWP and LU • Environmental impact was sensitive to SCP yield and level of impact allocation	• Work is required to upscale feed production from novel SCP. • Breaking technical and legislative barriers is next in achieving success in commercial adoption	(Tallentire et al., 2018)
Comparative analysis of meat substitutes' environmental performance to estimate the most promising options	1 kg of a ready-to-eat meal	Cradle-to-gate	Field and literature data Impact methods: ReCiPe and IMPACT 2002+ Secondary analysis: Sensitivity analysis (Calorific energy content of product used)	• SCP had a similar impact on human health to chicken meat but performed better than lab-grown meat. • Energy demand contributed **about 45 % and 25 %** of the impact of SCP processing and frying. • Chicken meat outweighed SCP in terms of calorie-based FU	• Comparison of meat substitutes in the same production conditions with sole dependence on field data needed. • The functional unit definition should be core in LCA since it could dramatically alter impact results	(Smetana et al., 2015)

Legend: CC: Climate Change, GWP: Global Warming Potential, LU: Land Use, WC: water consumption, FWC: Fresh water consumption FU: Functional Unit, ME: Marine Eutrophication, TA: Terrestrial Acidification, PNBS: Purple Non-Sulphur Bacteria

2.4.1.2 LCA: Application of the Innovative Power-to-X technology in SCP production

Power-to-X (PtX, where X could be any biobased product) technologies are reportedly self-sustaining alternatives for producing products from renewable energy sources via water electrolysis and other complimentary processes (Secreters European Union's Horizon Programme, 2022). Using this concept, a recent study compared a power-to-food (PtF) pathway for producing SCP relative to soybean and other SCP pathways. The study indicated a 60% offset of global warming by the PtF-SCP approach (0.81-1.00 kgCO2eq/kgprotein) relative to soybean production (0.89-3.74 kgCO2eq/kgprotein). This signifies the potential PtF-SCP bears in addressing the current climate emergency and, from the endpoint view, decoupling SCP production from human health damages (Sillman et al., 2020). Regarding land use, water use, and eutrophication, the PtF-SCP offered approximately 84.0-99.5% impact savings compared to soybean production, further signaling its sustainability inclination. Relative to other SCP pathways, such as methane-based SCP and Quorn mycoprotein (fungal SCP, Fusarium venenatum), PtF-SCP maintained its low GWP advantage. However, both reference pathways outweighed the PtF-SCP in eutrophication impact (Sillman et al., 2020; Smetana et al., 2015). The authors related global warming offsets of the PtF-SCP system with renewable electricity sources such as wind and solar, contrary to the use of conventional fossil electricity sources in the other pathways (Sillman et al., 2020). Additionally, a sensitivity analysis of the impact of different renewable energy sources on the performance of the PtF-SCP approach laid an exciting trend, highlighting wind electricity as a better alternative to solar for improved energy utilization.

Overall, land and global warming savings are the major environmental benefits driving the adoption of SCP production. Energy modeling is also honed as a critical activity for improving SCP sustainability, with recommendations underpinning renewable energy and waste heat recovery systems as more sustainable alternatives to electricity grids that rely on fossil energy. It is, however, notable how SCP fails to satisfy the absolute limitations of some planetary boundaries, reinstating the vitality of hotspot analysis and impact category expansion to understand the trade-offs and improvements required for augmenting future SCP technological scenarios.

2.4.2 Techno-Economic Assessment (TEA)

A brief description of TEA would place the perspectives discussed in this study into context. TEA is mainly used to analyze technical hotspots and viabilities of a product, process, or service (Giacomella, 2021). It has experienced a continuous escalating interest within academia and industry due to its support in *a priori* and *a posteriori* technical decision-making (Kumar & Tewary, 2021; Kurambhatti et al., 2021), revealing the cost variability in a system, project, and investment. It relies on technology and performance to contemplate the economic conditions of varying technical adoptions, which often triggers proclivity toward high-performing technologies or technical solutions with optimal economic impacts (Giacomella, 2021). These advantages establish the cruciality of TEA in enhancing the technical feasibility and performance of SCP production. Although comprehensive standalone studies on the TEA of SCP production are limited, quite a few studies have considered the integration of SCP production as an economically advantageous upcycle pathway in complex industrial settings.

Whole stillage is a major nutrient-dense byproduct of bioethanol production (Bulkan et al., 2020). In a typical application, whole stillage is processed into distiller's dried grains with solubles (DDGS) through energy-intensive centrifugation and evaporation. Empirical estimations highlight such steps to contribute an average of 35% and 43% to the electrical and thermal energy demands of a typical dry mill ethanol production plant, respectively, vitally affecting capital investment demands and intensifying environmental consequences (Bulkan et al., 2020). A techno-economic study targeting the identification of economic and energy-saving pathways asserted the substitution of conventional whole stillage processing with a downstream process that produces additional ethanol and protein-rich fungal biomass (practically SCP production) as a turnkey solution (Rajendran et al., 2016). Although this choice demanded capital investment to increase by USD 1.2 million to reach USD 70.2 million, the resulting increase in net present value (NPV) by USD 31 million and an attractive profit margin boom underlined its preference over the conventional production approach. Energywise, the novel approach resulted in approximately 2.5% energy savings, enhancing the technical efficiency of the process. An expansion of the system boundary of this study by (Bulkan et al., 2020) further buttressed the economic advantage of an integrated SCP pathway in ethanol production, also projecting an approximate 6% additional increase in NPV when fungal biomass is sold in the human food market.

Simulation of SCP production from grass silage via steam explosion, enzymatic hydrolysis, and alkaline pretreatment techniques was assessed for its techno-economic viability (Pihlajaniemi et al., 2020). For a processing capacity of 60,000 tons of silage (dry matter), a capital investment of approximately € 38.8 – 55.8 million was needed, equivalent to USD 42.02 – 60.42 million at present (January 2023), at an exchange rate of approximately 1 € = 1.082874 USD (Forbes, 2023). The steam explosion process was cost-intensive, placing the alkaline (ammonia) pretreatment technique as a feasible alternative for localized, small-scale SCP production. The authors highlight variable costs such as enzyme, silage protein, and protein quality as limiting factors to commercial success. Therefore, they recommend optimizing the processes in favor of these variables to enhance technoeconomic feasibility and progress commercial entry.

As accentuated by Giacomella (2021), the absence of a defined standard for TEA practice diversified the cost inclusion and exclusion criteria, indicator selection, and scope of the assessment for the studies considered in this review, subjecting generalized economic assumptions from present assessments to uncertainties. On that premise, it is strongly recommended that efforts toward developing a robust TEA standard be intensified to enhance the reproducibility, reliability, and generalizability of TEA studies and findings. Additionally, the unavailability of economic data for novel technologies has stalled techno-economic scrutiny of novel SCP pathways such as photovoltaic-driven SCP (in which microbes utilize chemical energy generated from bioconversion of solar energy) (Leger et al., 2021), PtF (Sillman et al., 2020), and microalgae (Janssen et al., 2022). Subsequently, generating such data would enhance the TEA of evolving pathways and promote a smooth transition to sustainable protein in an economically beneficial manner.

2.4.3 Current Gaps in SCP Sustainability Assessment

Recently, LCCA has gained traction in economic analysis due to its alignment with the LCA standards (ISO 2006:14040,14044) and consideration of a broader perspective of relevant system elements in cost analysis (Giacomella, 2021). It is distinguished by its intricate consideration of economics, cash flows, and other externalities like greenhouse gas emissions (Giacomella, 2021; Ioannidou et al., 2022). This presents current cost meanings of systems with little to no technological influence and situates cost modeling and decisions within the eco-economic decoupling frame (Allotey et al., 2023). SLCA is another critical component of the reformation in life cycle sustainability thinking, which aims to expound the relevance of social burdens or benefits associated with a product, service, or process in typical sustainability decisions (Caruso et al., 2022; Tsalis et al., 2022). It considers several social indicators, complexly characterized, usually qualitatively, to represent the accrued social impacts of a defined system on associated workers, the local community, consumers, and other value chain actors (Allotey et al., 2023; Caruso et al., 2022; Tsalis et al., 2022). While LCCA and SLCA have evolved to drive sustainability within economic and social contexts, rapid penetration of the environmental nutrition concept, especially into food and nutrition assessments, has also been witnessed (Aidoo et al., 2023). Holding this evolution is the desire to negotiate for sustainability pursuits with environmental and nutrition co-benefits (Agyemang, P. et al., 2022; Aidoo et al., 2023). In this regard, decision-making targets solutions that can facilitate the achievement of optimal environmental and nutritional benefits (Aidoo et al., 2023). Life Cycle Sustainability Assessment presents a holistic and robust perspective to sustainability assessment, combining all the sustainability metrics and applying trade-off or multi-criteria decision analysis for identifying optimal solutions (Allotey et al., 2023; Wada et al., 2022). Despite the apparent significance of these concepts in robust sustainability assessment, a dearth of studies has explicitly applied them in SCP production systems according to our present knowledge.

Thus, we foresee their integration as an opportunity to improve the SCP process and product design and provide a reliable baseline for making sustainable decisions at micro, meso, and macro levels.

2.4.4 The Digital Twin Concept for Improving Production and Sustainability

Digital Twin is an Industry 4.0 technology that uses a virtual reality concept incorporating computer simulation into actual system operations (Dyck et al., 2022). Complex system operations are brought into real-time virtual view by coupling sensors with other graphical, mathematical, or predictive computer models. Dyck et al. (2022) mentioned three dynamic elements of a Digital Twin: a physical product in a physical space, a virtual product in a virtual space, and a mediative element that ties the physical product to its virtual representation using data and information. Whereas the adoption of digital twins into the food industry is still in its early stages, the benefits have been colossal, with massive improvements in productivity and the greenness of the systems that have applied the concept (Hassoun et al., 2022a). In the recent application of Digital Twin in agriculture, the virtual representation of farms has been modeled to enhance data transmission, processing, and optimization of physical processes to maximize efficiency and reduce energy use, improving the overall sustainability of farms (Nasirahmadi & Hensel, 2022). Also, integrating digital twins in food and other industrial production lines has eased traceability, hotspot identification and accelerated input of corrective actions to improve system sustainability performance (He & Bai, 2020). By embracing a digital twin system in the gradually evolving SCP production trend, we can enable a unique trajectory of smart protein production in an Industry 4.0 era while placing SCP production within the sustainability tenets. A summary of some available models or technologies that could sponsor a digital twin trajectory in SCP production is presented in Table 2.3 and the following subsections.

2.4.4.1 Available Technologies and Models for Establishing an SCP-Digital Twin

Table 2.3 captures some available sensor technologies that could be utilized in building an SCP-digital twin. It briefly describes their mode of operation, current application, and relevant findings from the use.

Table 2.3: Sensor Technologies for Monitoring Fermentation Process

Technology	Mode of Operation	Current Application	Major Findings	Reference
Microbial Potentiometric Sensors (MPS)	Considering fermentation as a complex redox reaction, the potentiometric sensor measures the potential difference between the sample and a reference electrode probe which produces a signal to represent the stage of fermentation	To monitor the completion time of kefir-facilitated milk fermentation	• The MPS technology could monitor kefir fermentation in real-time with high reproducibility. • The regression analysis approach was able to discern a correlation between the fermentation completion time	(Hristovski et al., 2022)

			and the mass of kefir inoculum	
Thermodynamic Sensors (TDS)	Based on the energy measurement, which is supplied to the circuit to temperature setting and equilibration of temperature element with the ambient. The signals read are translated as the state of microbial activity	Preliminary studies have tested the performance of TDS in monitoring some phases like the end of the fermentation process in dairy fermentation, beer brewing, yogurt fermentation, and baking processes	• Need for advanced studies to verify its validity and performance at large industrial scales	(Adamek et al., 2022)
Electrochemical Glucose Biosensors	Enzymatic oxidation of glucose to gluconic acid followed by the re-oxidation of flavin groups to form H_2O_2 generation. H_2O_2 undergoes anodic oxidation on the surface of a working electrode which produces signals that are translated into a glucose concentration	Glucose quantification for batch-fed fermentation of yeast	• Fast and accurate measurement of glucose concentrations in fermentation • Glucose detection ranges up to 150 mM	(Pontius et al., 2020)
Fluorescence-based optical sensors	Luminophor (a polymeric matrix) absorbs photons to reach a higher energy state with excited electrons. The interference in energy absorption influenced by the medium's physical (temperature and pressure) and chemical properties (nutrient composition) alters the energy gain of electrons in the luminophore, translated as the level of luminescence. Such variation in electron energy before and after oxygen interference is a measure of luminescence.	Oxygen concentration in winemaking	• Bears advantages in small and large-scale applications	(Trivellin et al., 2018)
Combined Internet of Things and CO_2 Sensor System	The system consists of a CO_2 tunnel that collects CO_2 produced during fermentation. A sensor in the tunnel detects CO_2 concentration which is quantified and measured against a threshold. A fan is connected to the system, which activates and deactivates if CO_2 is above or below the threshold. A wireless system transmits data from the sensors for instant visualization and feedback.	Real-time monitoring of an alcoholic fermentation process proved an efficient solution for optimizing and controlling wine fermentation at a laboratory scale	• The instant visualization and feedback of the system minimize the need for human intervention. • The system can determine the state of fermentation and identify whether it is in the natural course, beginning, tumultuous, or completing using CO_2 signals	(Canete-Carmona et al., 2020)

2.4.5 Data-Driven Models

Data-driven models have gained core relevance in industrial processes, including fermentation. Its role in practical real-time analysis of sensor data and prediction of process variables, fault detection, and design optimization is outstanding. Recent developments have focused on overcoming process constraints like complexity, non-linearity, and time variations (Zhu et al., 2020). A detailed review of selected modern soft sensing models with application in fermentation is briefly highlighted under this subsection.

2.4.5.1 Support Vector Machine (SVM) Model

This standard data-driven soft sensing model has been used for predicting the output of complex non-linear processes or systems even with small sample data. In practice, it is considered ideal for fermentation processes with small sample data to self-learn and accurately predict characteristics for accurate generalizations (Zhu et al., 2020). For large datasets, using SVM models can be very cost prohibitive. The coupling of SVM with other predictive and optimization methods like generalized predictive control (GPC), Particle Swarm Optimization (PSO), Least Square (LS), and Multiple Output Variable Least Square (MLS) models rendered better prediction of process parameters such as biomass and substrate concentration (Robles-Rodriguez et al., 2016; Wang & Ji, 2015).

2.4.5.2 Fuzzy-Logic (FL) Models

FL has also been deployed in process optimization, parameter or pattern identification, and system control. Having the potential to imitate the reasoning prowess of humans, FL has gained utility prominence in making intelligent data-driven guesses of the dynamics in fermentation systems. A typical example is the fuzzy neural networks (FNN) model for predicting variables like biomass, substrate, and product concentration of a penicillin fermentation process (Yonghong et al., 2012). For input variables like dissolved oxygen, carbon dioxide, and sugar concentration, a uniform incidence degree algorithm has been employed for their identification with a high degree of performance (Zhu et al., 2020).

2.4.5.3 Deep Learning (DL) Models

DL-based soft-sensing models are also data-driven machine-learning models that have evolved in modern science and engineering design, optimization, and control. Unlike SVM, deep learning models portray the benefits of effectively handling non-linear structures, big process data cases, and better parameter approximations at a relatively affordable cost (Shang et al., 2014). DL-based models such as deep neural networks (Ke et al., 2017), Hierarchical Extreme Learning Machines (HELM) (Yao & Ge, 2018), among others, have been satisfactorily used in the parameter estimation of the penicillin fermentation process and other multivariable bioprocesses with extensive process data (Gopakumar et al., 2018). Recently, Xu et al. (2022) also harnessed the integrative capacity of mechanistic modeling and deep learning models like Recurrent Neural Network (RNN) for predicting and optimizing the media enrichment step in a bioenergy production process involving polyhydroxyalkanoates (PHA) forming bacteria. The optimization procedure was to identify the best combination of additives to achieve optimal PHA with desirable functionalities. These models could be duly deployed in an SCP-digital twin system for real-time monitoring or estimating such parameters for process decisions.

2.5 Future Perspectives and Conclusions

The review highlights SCP as an evolving protein alternative with the potential to enhance nutrition security without compromising environmental integrity. Expanding its commercial visibility emulates a trajectory for advancing circular bioeconomy goals in a much-linearized protein business model while producing enough protein to complement deficits in the current supply. A graphical summary of some potentials and limitations to SCP production is shown in Figure 2.5. We base on the revamped interest in process and technological improvement to envision a rapid influx of innovations to facilitate the SCP bio-revolution in response to the need to provide sustainable protein alternatives. For instance, current projections estimate an increase in SCP market value to USD 18.5 billion by 2030 (Global Market Insights, 2023), which provides enough motivation to recommend reinforced research and development in this emerging industry. Governmental and non-governmental agencies are incrementally enthused about this trajectory, signaling comprehensive funding sources to support the current and projected SCP business scale. Certainty prevails that treading the SCP pathway also presents essential sustainability benefits, especially in reducing global warming and land use. However, significant gaps in applying LCCA, SLCA, and Environmental Nutrition are noted, which could subsequently be explored together with LCA and TEA in a multi-objective manner to provide justifiable baselines for sustainability decisions.

The expansion of SCP production within a global scope of diversified consumers comes with several limitations, with consumer disorders like food neophobia - the distrust and reluctance to try new foods, strict and limiting regulations, and some eliminable safety concerns like high nucleic acid contents standing out amongst the cluster of limitations. Food neophobia has been associated with the dwindling bacterial SCP market share and has contributed colossally to the existing resistance to SCP commerce. To some, boosting the penetration of bacterial SCP on the consumer market would demand intense sensitization and education to reconscientize the sheer number of consumers who uphold asceticism to escape the alarming pathogenicity of bacteria (Mazac et al., 2022). In proposition, subsequent SCP food design and development should prioritize food snob or fear factor as a significant parameter to estimate consumer resistance. This can guide decisions about optimizations and further market actions. Some novel SCPs, like microalgal SCP, have also suffered delays in commercial launch due to complex and costly regulatory demands. For instance, it is very time-consuming and cost-intensive to fulfill the regulatory demands for introducing entirely new products on the food market in the face of efforts to maximize compliance with safety and quality requirements. Consequently, SCP commerce has become unattractive for small and medium enterprises (Janssen et al., 2022). Thus, strategies to address these limitations while progressing innovations toward maximizing system efficiency must be engaged to activate the full commercial potential of sustainable SCP production.

In a world where digitization has become an indelible norm, virtualizing SCP production has benefits in optimizing performance and maximizing competitive advantage. The digital twin concept seems to hold high prospects in advancing SCP production, which could be activated by exploring the actual performance of the discussed technologies and models. Success in enabling the SCP-digital economy would strengthen its resilience in a digitally evolving food system and ease process design, optimization, monitoring, and control of current and future designs.

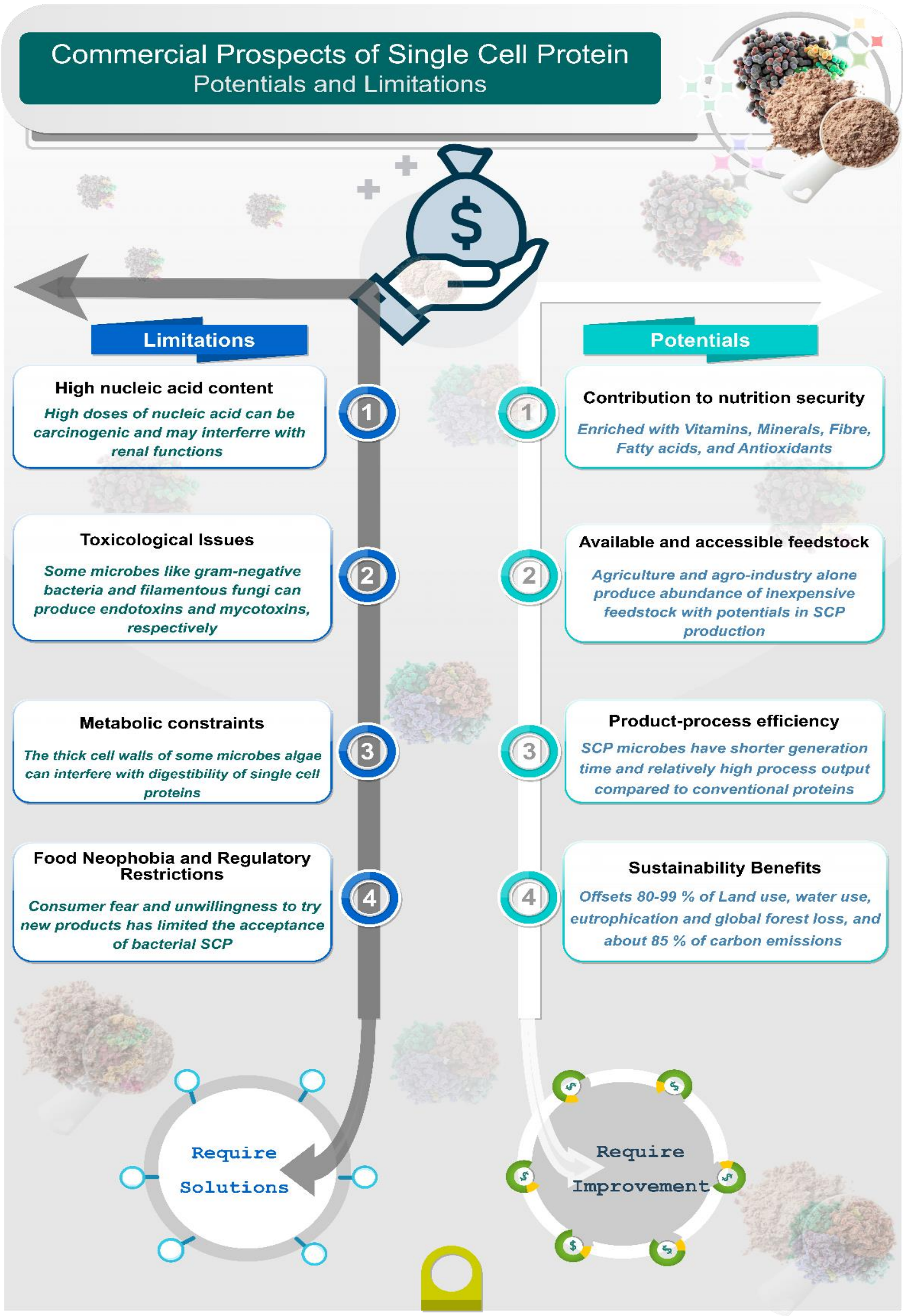

Figure 2.5: Potentials and Limitations to the Commercial Entry of SCP

CONNECTING TEXT II

The manuscript for chapter two has been published in Trends in Food Science and Technology.

The literature review (Chapter 2) provided a comprehensive overview of the production, commercial prospects, and sustainability outlook of Single Cell Protein. It highlights SCP as a noteworthy circular bioeconomy approach considering co-benefits in value recovery, enhancing sustainability benefits, and the potential to promote protein availability. Solid-state fermentation is emphasized as a more sustainable approach for SCP production, with relatively less energy and water demand. The review also highlights the significant global warming and land use offsets associated with SCP production relative to some conventional animal and plant-based proteins and how adopting renewable energy sources and second-generation substrates could augment sustainability performance. For second-generation substrates, pulse co-products like crude pea starch are underscored as a potential substrate, considering their rich composition of nutrients, mainly starch, and how that could economize raw material needs for SCP production. However, such substrates have not been satisfactorily explored. Overall, the chapter establishes the relevance of pursuing SCP production as a biocircular alternative in the agro-industry and draws explicit attention to expanding exploration in the pulse industry.

However, before venturing into the mainstream circularization of pea starch for SCP production, Chapter 3 addresses a critical gap in circular bioeconomy practice: developing a robust and adaptable circular bioeconomy accounting framework to guide the exploration process.

3 CHAPTER THREE: C-BEAT DEVELOPMENT

Circular Bioeconomy Accounting Tool (C-BEAT): A Comprehensive Framework for Improving Agro-Industrial Circular Bioeconomy Practice

Abstract

As sustainability evolves, the need for circular business models has become more critical. As a result, food system stakeholders have become more interested in finding valorization pathways for waste outputs or coproducts along the value chain to reduce the global waste burden and enhance resource use efficiency. However, a major shortfall in this evolving circular bioeconomy transition is the limitation in assessing uncertainties attached to circularity pathways, especially in comprehensively understanding their sustainability performance before real-time deployment. In addition, experts have noted the limited participation of multiple stakeholders in circular decisions, especially on the consumer side, breeding significant uncertainties regarding public interest and commercial success. To bridge these gaps and improve our progress to a sustainable food system transformation, this study leverages the relevance of stakeholder engagement, sustainability assessment, and multicriteria decision analysis in developing a comprehensive and sector-adaptable circular bioeconomy framework. This framework intends to provide an adaptable guideline that prioritizes stakeholder participation in identifying circularity pathways within a given system and enables a robust life cycle sustainability assessment of identified pathways. Life cycle Assessment and Costing are deployed for environmental and economic analysis, and a novel employment index, representing the contribution of a pathway to locational unemployment rate reduction, is harnessed as an easy-to-quantify social metric. For accessible communication and enhanced relatability of circular performances, a relative circular bioeconomy index (CBI) is mathematically modeled as a unified sustainability accounting function of the three sustainability indicators. Also, a combined multicriteria decision analysis approach, BWM-CoCoSo, is introduced as a robust approach for multi-objective trade-off analysis for enhanced circular decision-making amongst practitioners or businesses.

Keywords: Circular Bioeconomy, Framework, Circular Bioeconomy Index, Multicriteria decision analysis, Employment Index

3.1 Introduction

3.1.1 Background

Various theoretical proponents have defined the primary drivers of economic growth. Some argue a strong correlation between exponential economic growth and technology, labor, and capital development, popularly classified as the neoclassical growth theory (Abibo et al., 2022). In plain words, the neoclassical school of thought asserts that without technological innovation, skilled and available workforce, and high capital influx, an economy's desire to increase goods and services would constantly backfire. However, the prominent new growth theory challenges this perspective and predicates economic growth on a reformed proclivity towards knowledge improvement (Apostol et al., 2022). It treats knowledge as an asset and emphasizes advanced knowledge to hold every economic ambition that seeks to satisfy growing consumer wants and maximize profit incentives. This theory claims that it is only through improved knowledge that we can have new technologies, products, and innovations to ameliorate economic growth. Several other economic growth theories present other interesting placements of labor, capital, knowledge, and technology in competitive development (Afonso, 2020; Soskice, 2022). The interest in presenting these theories is not to deliberate the conflicting intellectual perspectives. Instead, it is to accentuate the relentless efforts toward defining realistic patterns for exponential economic welfare and growth to either improve or overcome the limitations of the dominant linear economy.

A linear economy follows a make-use-dispose resource model, where resources are used once, and wastes generated are disregarded in subsequent growth strategies. It focuses on continuous production and consumption with unsatisfactory consideration of restoration or regeneration (Alhosni et al., 2022). This suggests the continuous generation of waste or by/coproduct, as the desire for economic growth and increasing consumer demands trigger the continuous production and consumption of resources. Unfortunately, the underutilization of these valuable residues poses significant economic, environmental, and social threats, placing local and global sustainability boundaries in great jeopardy (Liu et al., 2022; López et al., 2022). For instance, food waste across the food value chain is attached with an annual economic cost of $1 trillion, an environmental cost of $700 billion, and a social cost of $ 900 billion, amounting to an avoidable annual global expenditure of $2.6 trillion (FAO, 2013; Riesenegger & Hübner, 2022). In addition, these wasted foods consummately account for approximately 8-10% of global greenhouse gas emissions, establishing its detriment to the global carbon target and climate adaptation plans (UNEP, 2021). These findings expose significant fractures in the dominant linear economy's ability to fulfill the global agenda of sustainable growth and economic prosperity. They hint at the unreliability of existing linear growth theories and business models and emphasize the need to formulate new theories and strategic growth models for a green and regenerative revolution (Stefanakis & Nikolaou, 2022). Several transformative solutions have evolved regarding this premise, among which we find the rapid evolution of the circular bioeconomy concept. In expert opinion, circular bioeconomy presents a regenerative growth pattern to relieve the earth of the aggravating implications of the traditional linear agri-food system - characterized by massive waste generation, high carbon footprint, and planetary damages (Massimiliano & Luigi, 2022; Robinson, 2022). Backing this assertion is the waste-to-resource strategy of the circular bioeconomy, which envisions system-wide sustainability through the efficient valorization of waste biomass through competitive upcycling techniques (Alhosni et al., 2022; Brummelhuis & Marinelli, 2022). This has accelerated the evolving global adoption and instigated remarkable advocacy amongst food system stakeholders, including governments, manufacturers, scientists, researchers, and policymakers, rallying for its centralization in political and non-political agri-food system transformation strategies (Schulze, 2016).

3.1.2 Traditional Gap Analysis

While circular bioeconomy progressively gains momentum in policy, research, and industrial engagements, it has become expedient to augment the framework to address overlaps in adoption and guide sustainable practice (Alhosni et al., 2022; Mikroni et al., 2022; Minguela et al., 2022; Nowakowski et al., 2022; Saha & Mathew, 2022; Sepetis, 2022). To achieve this, Kershaw et al. (2021) delineate critical impeding factors to a sustainable circular bioeconomy trajectory that needs to be addressed. Among the outstanding gaps, marginalization of sustainability dimensions, limited stakeholder and expert participation diversity, and narrow problem and solution framings have been reiteratively emphasized. D'Amato et al. (2020) and Stegmann et al. (2020) affirm the existing marginal interest in intersecting the fundamental sustainability orientation, that is, economic prosperity, ecological protection, and social equity, in circular bioeconomy trajectories, inferring the need to harmonize these dimensions as a means to attaining a robust circular bioeconomy. The study by Palahí et al. (2020) also recognizes the disconnect in stakeholder participation, recommending the engagement of multiple actors to enable co-creation and responsible innovation. Recently, the integration of decision tools has been recommended and perused in circular bioeconomy practice, portraying relevance in addressing the multipurpose objectives of circular bioeconomy decisions (Romero-Perdomo & González-Curbelo, 2023). Hitherto, these gaps have been tackled from standalone perspectives, missing the benefits of consolidating their solutions into a comprehensive framework to guide an equitable, resilient, and socially robust circular bioeconomy practice. This forms the basis of this study’s central hypothesis, which supposes the integration of the triple-bottom-line sustainability assessment, stakeholder participation, and multicriteria decision analysis as a robust strategy for accelerating an inclusive and sustainable circular bioeconomy practice (Köhler et al., 2022; Massimiliano & Luigi, 2022).

Therefore, this study explores the development of a system-adaptable circular bioeconomy decision support framework, herein termed "circular bioeconomy accounting tool" (C-BEAT), to guide inclusive and sustainable practice in agricultural and agro-industrial systems. This proposed accounting tool considers multiple stakeholder participation, including managers, investors, researchers, and consumers, for the co-identification and co-validation of circularity pathways; three sustainability accounting tools: life cycle assessment (LCA), life cycle costing (LCC), and social assessment (SA) for a comprehensive sustainability assessment; and two decision tools that allow an aggregative and multicriteria decision making. The C-BEAT is framed to offer multiple benefits. Thus, in addition to its regard for aligning circularity practice with global sustainability commitments, it also offers an opportunity to nudge and maximize the participation of multiple decision actors to ensure competitive circularity outcomes.

3.2 Conceptual Framework

A graphical summary of the study flow is presented in Figure 3.1. The study commences with the development of a theoretical framework. This framework follows a 5-phase procedure that prioritizes identifying, defining, validating, designing, and simulating possible scenarios, assessing the sustainability performance of the validated scenarios, and engaging decisions making tools in assessing the possible trade-offs. In phase one, stakeholder inclusion and participation are captured through a series of engagements to solicit internal and external feedback on the circularity potential of the focus system. For instance, internal stakeholders like managers, shareholders, and workers would be engaged to identify the potential circularity options. Then, through a market survey, consumers or customers would be engaged to validate which internally identified circularity potentials have better chances of thriving in the available market space. Finally, further internal engagement and subjective assessment would validate the identified pathway(s) worth pursuing. Such engagements would yield a list of deployable, market-ready pathways that would be further treated in the subsequent phases. Phase two involves a detailed description of the process flow, process assumptions, technological and methodological options, purpose, characteristics, and target users of the products from the identified pathways. This would give an explicit understanding of the identified pathways to facilitate the design of possible scenarios.

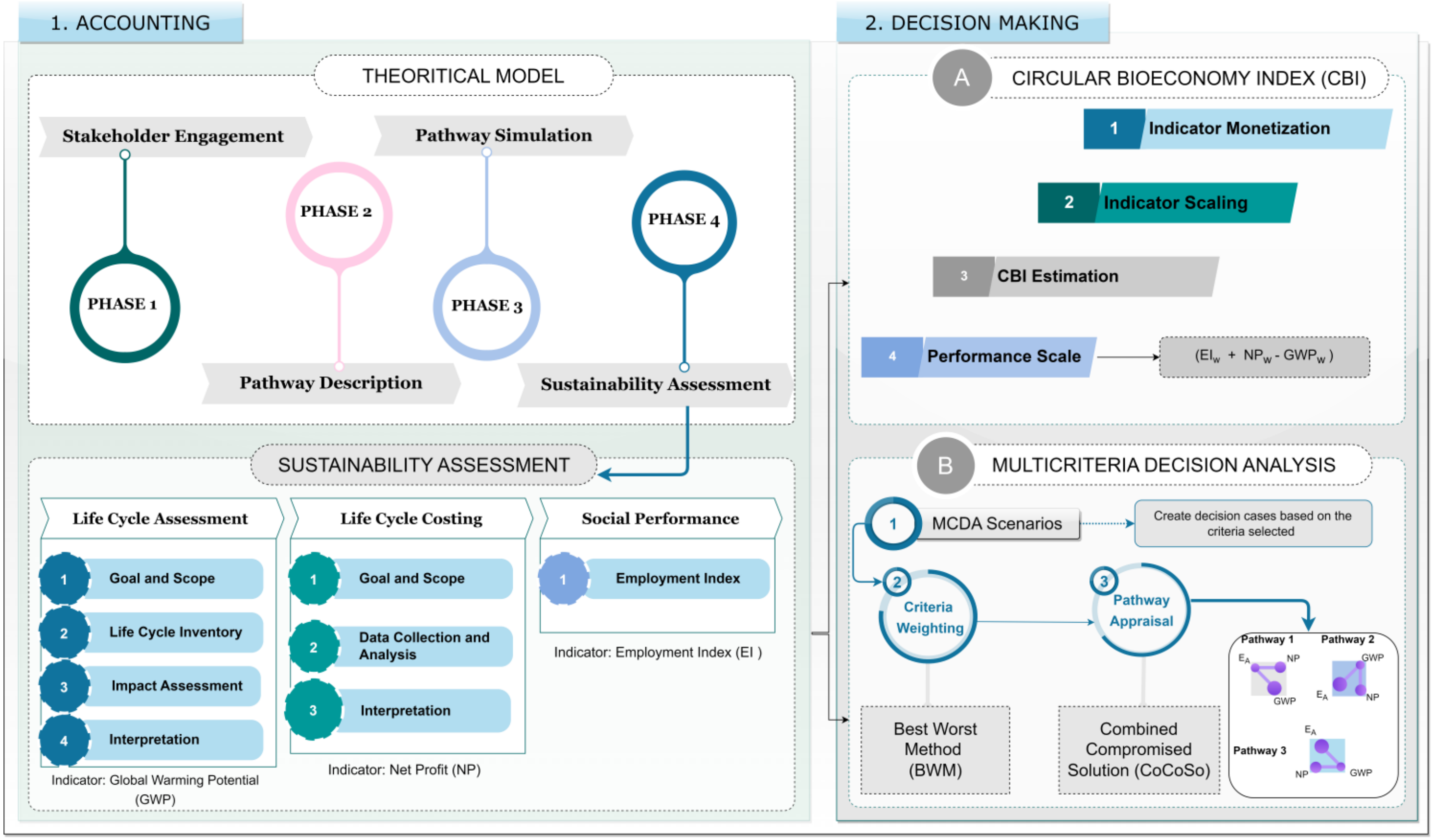

Figure 3.1: Conceptual Framework of Study

Pathway simulation, captured as phase 3, involves designing and simulating all possible scenarios for the pathways identified, given the diversity in processing steps, assumptions, and methodological or technological alternatives described. These scenarios would then be assessed for their sustainability performance, captured as phase four in the theoretical framework. The sustainability performance involves life cycle assessment, life cycle costing, and societal impact assessment of the designed pathways. Results from these assessments would prime phase 5 of the framework, circular bioeconomy decision-making, which takes two quantitative perspectives. First is the circular bioeconomy index (CBI) – a single score to represent the performance of models based on values from three sustainability indicators (environmental, economic, and social performances). Next is the use of multicriteria decision analysis for user or stakeholder-specific ranking. This enables biocircular decisions that represent the ideal preferences of the stakeholders. The first option would enhance circular bioeconomy communication, while the second would ameliorate internal decisions amongst firms, industries, or stakeholders based on expert weightings assigned to the indicators.

3.3 Theoretical model: Circular Bioeconomy Accounting Tool (C-BEAT)

This section describes the proposed circular bioeconomy accounting tool. The tool is framed into four interrelated phases, subsequently elaborated on with details of all steps and activities considered for each phase. Figure 3.2 is a graphical summary of the theoretical model.

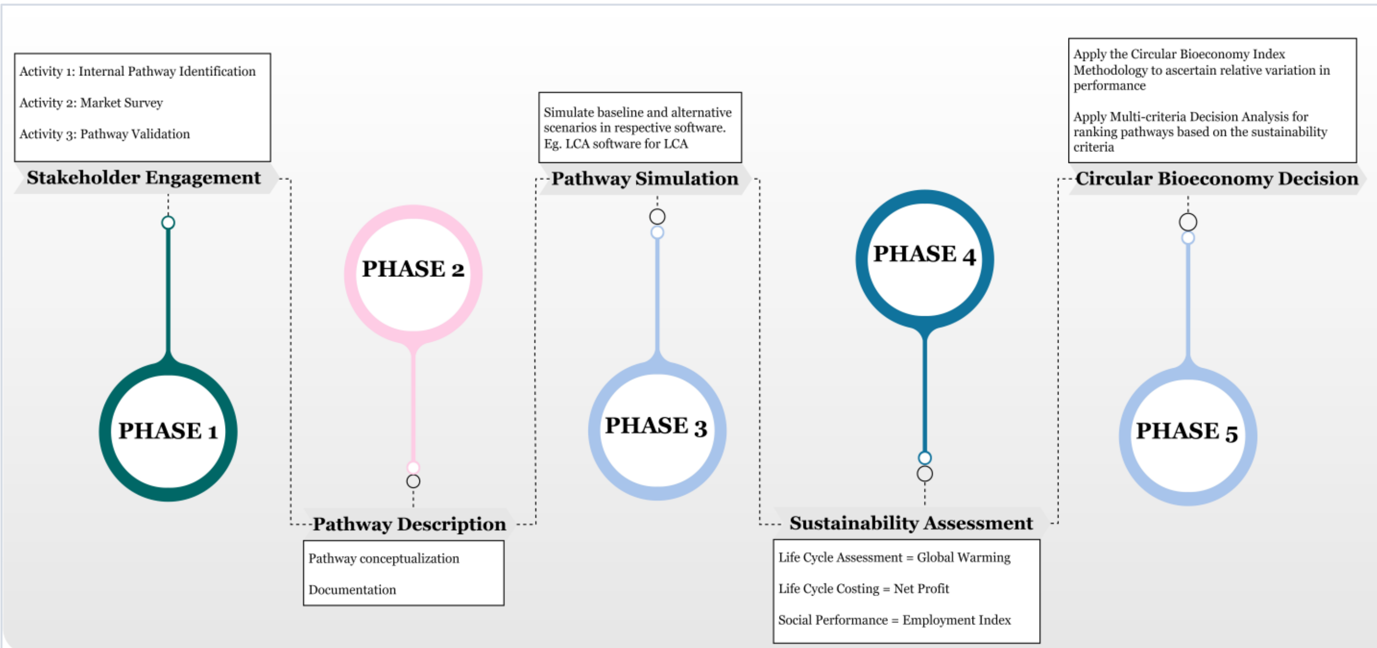

Figure 3.2: Theoretical Model

3.3.1 Phase 1: Stakeholder Engagement

Various project management experts have reiteratively emphasized stakeholder exclusion in projects or programs as one of the most significant risks of failure (Husgafvel et al., 2022). It has been ascertained that a project that does not receive the maximum approval and involvement of important stakeholders will most likely be ignored or suffer redundancy even after implementation. The vitality of prioritizing stakeholders in circularity practice and designs encouraged the integration of a stakeholder engagement component in this theoretical framework. Here, we assume the participation of these stakeholders as a means of cementing accessions on desired pathways and creating an exclusive ground for establishing shared commonalities. Inarguably, hierarchical and representational variances may exist among different organizations or companies. However, in this step, it is expected that internal business drivers like managers, workers, investors, consultants, and similar portfolios, and external stakeholders like consumers (customers), researchers, retailers/wholesalers are fairly represented and adequately engaged in the selection of desired valorization pathways, in this case, for the by/coproducts of the focus system. The step is divided into four major activities, which detail the actions for a successful stakeholder engagement.

3.3.1.1 Activity 1: Core team selection

This is the first activity in the stakeholder engagement process. It involves choosing the right expertise to lead the entire circular bioeconomy practice in the focus system. This is where attention should be given to the issue under study and who would enhance the compilation of strategic solutions, herein, internal circular pathways. A fair representation of the stakeholders that play crucial roles in investment (Yadav et al., 2020), marketing (Govindan et al., 2019), distribution (Singh et al., 2022), managing, consulting, research, and development (Govindan & Gholizadeh, 2021), sustainability, and actual production (Govindan et al., 2019) is required in the team selection. Such diversity is needed to minimize the risk of unilateral ideas and maximize the opportunity to formulate pathways with optimal feasibility and success rate.

3.3.1.2 Activity 2: Internal Pathway Identification

The initial idea-generation step of the framework happens in this activity. Numerous valorization options may exist, however, not all these options would be in the interest of a focus system based on their goals or willingness to divert into a new business niche. It is, therefore, important that ideas of possible circularity pathways are first drawn through an internal forum. This forum is proposed to allow the internal stakeholders (core team) to comprehensively discuss and identify available circularity options adjacent to the system's goals, vision, mission, or explorative prospects. At this stage, it is expected that a brief description of the identified pathways is done to enhance understanding amongst the team and also to facilitate subsequent actions.

3.3.1.3 Activity 3: Market (Consumer) Survey

Profit-making is a longstanding reality in every business entity (Lahti et al., 2018; Tur-Porcar et al., 2018). However, unless supply meets the demands of an existing market, business models may suffer severe economic discrepancies, contradicting the indelible profit-making goal of the business-as-usual. Thus, this stage provides actionable insights for shortlisting internally identified circularity pathways based on consumer orientation, market readiness, and profitability. It is practically the public engagement arm of the model that solicits public or market, or consumer perspectives on the potentials of the internally identified pathways. Such would ensure data-driven decisions that place selected pathways in a high class of commercial viability. It places the selected pathways within a consolidated structure for maximizing profit and closely satisfying multistakeholder desires.

At this stage, radical collaboration with survey experts, researchers, or business strategists is needed to complement the relevant skills and competencies required for strategizing and effectively executing the survey.

3.3.1.4 Activity 4: Pathway validation

This activity is eliminable. However, it portrays strong relevance in further shortlisting the selected pathways from consumer surveys against supportive exclusion-inclusion tools. Screening models like the SWOT analysis and decision models like the Direct Ranking Multicriteria Decision Model, among other data analysis tools, could be deployed as easy-step validation tools. The outcome would be a multistakeholder validated pathway(s) that benefits the interest of consumers and manufacturers alike.

3.3.2 Phase 2: Pathway Description

This is another vital procedure in the theoretical framework. Here, design perspectives are deployed to develop and precisely describe the pathways. It considers a comprehensive conceptualization of the validated pathways, where raw material handling, processing steps, process assumptions, potential scenarios, and the characteristics of the final product are deduced. The outcome of this step is a thorough documentation of the pathways' life cycle, including feedstock handling, processing steps, final product handling, and waste alternation steps, with a detailed explication of the material, technological and methodological options. The life cycle may include final product handling depending on the defined circular bioeconomy accounting boundaries. In this case, a description of the two Ps of the final product: performance and presentation, should be considered. Performance indicates what functionalities or qualities are expected of the final product, while presentation deals with the aesthetic component of the final product, primarily packaging and labeling. These would further facilitate the selection of reference flows, mass, and energy balance calculation and enhance accuracy in the overall accounting process.

3.3.3 Phase 3: Simulation of Pathways

3.3.4 Simulation has been regarded as a reasonable approach that allows the extrapolation of process or system performance through dynamic model development. It is guided by defined criteria or assumptions concerning the dynamics of a real system (Ingalls, 2011). This phase is where baseline and alternative scenarios for the validated pathways are designed and simulated in computational software, carefully considering the assumptions or features described, the type of assessment, and respective assessment standards or guidelines. In this framework, the three tenets of sustainability, summarized in Figure 3.3, are considered in the sustainability assessment. The guidelines or methods for assessing these tenets are elaborated in Phase 4.

Figure 3.3: Sustainability Tennets

3.3.5 Phase 4: Sustainability Assessment

3.3.5.1 Life Cycle Assessment (LCA)

LCA considers material input, emissions, and output flows of a product or system during its life cycle to ascertain the environmental impact or emission burden potential of such product/system (Berton et al., 2021). It provides a baseline for understanding the environmental burdens associated with a system or product, driving strategies for improving performance. ISO 2006: 14040/44 outlines and describes the standard procedure for conducting a life cycle assessment, categorizing the steps into four interlinked phases – goal and scope, life cycle inventory, life cycle impact assessment, and interpretation (Duan et al., 2022; ISO, 2006; Nabavi-Pelesaraei et al., 2022; Shafique & Luo, 2022). For further details on these steps, refer to the stated ISO standards. For reproducibility and easy generalizations, this model recommends the ISO 14040,14044:2006 standards. The selection of the impact assessment method should be based on the geographic location, availability of data, and other preferences of practitioners and clearly stated in the impact assessment stage of the LCA. Also, it is recommended that the mass functional unit be used to estimate the environmental impact over the operational life span to easily attribute other social and economic burdens to the reference flow. Other functional units, like the caloric functional unit, may be helpful for the calculation of specific or function-mediated impacts. However, it does not provide a relatable base for associating social burdens or benefits since most stakeholders can easily relate to mass or weight than intrinsic qualities like calories or protein content.

3.3.5.2 Life Cycle Cost Assessment

Cost performance is an inextricable component of process design and all business models. As such, cost analysis of systems, processes, or products has become essential in sustainable process decisions, intending to enhance a win-win benefit in achieving economic gains while reducing environmental impacts. In this model, an ex-ante environmental life cycle costing (E-LCC) is deployed as the cost analysis instrument because of its broad utilization in the cost assessment of agricultural products like food waste (De Menna et al., 2018), its alignment with LCA, and its multi-actor cost modeling perspective (Giacomella, 2021; Hauschild et al., 2018). Notably, its alignment with LCA places it well within the sustainability context, allowing a more reliable and robust life cycle sustainability accounting. Also, unlike conventional LCC, considering multiple actors or elements in the product enhances the comprehensiveness of the cost modeling, ensuring a more sustainable economic decision (De Menna et al., 2018; Giacomella, 2021). The current theoretical model recommends the methodology described by (Rödger et al., 2018) for environmental life cycle costing. However, here, externalities like waste disposal costs are not considered due to the resolve of the model to drive and integrate the waste-to-resource approach in the scenario modeling. Thus, it is assumed that most waste management sections would impose more benefits than harm. Without a defined standard for LCC analysis, Rödger et al. (2018) underscore a three-step procedure enlisted as goal and scope definition, data collection, and interpretation and sensitivity analysis, similar to descriptions and definitions in the standardized LCA methodology. In this framework, total capital investment (TCI), operation, and maintenance expenditure (OPMEX), cost of environmental emissions, and end-of-life cost from the manufacturer's viewpoint are considered the key economic parameters for the cost analysis inspired by recommendations from existing literature (Xiao et al., 2022). The framework recommends that relevant expenses should be aggregated separately under these parameters to ease hotspot analysis. The calculated net profit for each pathway would be the basis for comparison. However, the decision models are flexible to take any preferred variable.

3.3.5.3 Social Impact Assessment

Often, it is challenging to quantify or qualify the social impact of a project or an intervention due to regional and inter-industrial variances in standards or performance indicators. Moreover, there is a limitation to accurately predicting the extent to which some subjective social indicators like human rights, employee satisfaction, or welfare, among others, are breached in qualitative and quantitative terms, which has been a significant obstruction to Social LCA development. Nonetheless, it is possible to accurately quantify other simple but relevant socioeconomic indicators, like employment rate, employment-to-population ratio, labor force participation, and fair wages, when dealing with the contribution of businesses to societal prosperity. In this initial model, the employment index, calculated using the employment rate principle, is deployed in the social impact assessment of the identified circularity pathways. This step works within the system boundaries of the alternatives. Thus, the number of people employed for operations within the system boundary is considered.

- Employment Index (EI)

The employment index follows the principle of employment rate to estimate the potential contribution of the selected circularity pathways in enhancing labor force engagement within a defined geography. It intends to exhibit the impact of the pathway in enhancing economic health through unemployment rate reduction. Conjugating the employment index with the other sustainability indicators would allow a robust business decision for deploying the most beneficial circularity pathway. The steps for calculating the employment index are outlined as follows.

i. Estimate the pathway life span (T). The pathway life span is the service life of the pathway or how long the implemented pathway is supposed to be in operation.
ii. Gather data on the population (Pi) and the potential labor pool (labor force) (L_i, where "i" represents the year in consideration) in the considered geography for the nearest ten-year interval at the time of the analysis implementation.
iii. Based on the popularly hypothesized positive linear correlation between population and labor force as seen in most economic instances, construct a scatter plot of the population against labor force value for the ten-year interval.
iv. Determine the linear regression model (a linear equation ($Pj' = mLj' + c$) of the line of best fit with available statistical software and estimate the projected labor force value ($L_{j'}$, where j′ is the projected year) for each year in the pathway life span (T) using projected population for each year (P′).
v. Determine the number of employees (Ej) for each year of the operation span and calculate the employment index (ϵ) as a function of the ratio of the number of employees to the projected labor force value (Eq 3.1)
vi. Calculate the actual employment index for each year (Eq. 3.2). An average of the employment indices along the operation span would represent the Actual employment index (ϵ_A) of the pathway (Eq. 3.3)

Employment Index Variables

- Pathway Life Span = T
- Population = Pi, where i represents the year with known population
- Labor force for i_{year} = Li
- Projected Population for j_{year} = Pj´
- Projected Labor force for j_{year} = Lj´
- Estimated number of employees for j_{year} = Ej
- Employment Index = ϵ

Employment Index Equations

- Linear regression model for projected labor force

$$Pj' = mLj' + c \text{ ------------------------------------ } Eq\ 3.1$$

Where, **'m'** is the gradient of the graph, and

'c' is the population value when Labor force is zero

- Employment index for j_{thyear}

$$\epsilon = \frac{Ej_{th}}{Lj_{th}} \text{ ------------------------------------ } Eq\ 3.2$$

- Actual employment index for Project life span

3.3.6 Phase 5: Circular Bioeconomy Decision

The circular bioeconomy decision takes two accounting perspectives, (i) a unified or single score and (ii) the multicriteria decision analysis. These perspectives are further elaborated in the following subsections.

3.3.6.1 Unified score – Circular Bioeconomy Index (CBI)

The unified score approach translates the circularity performance of the pathways into a single score or index for easy relativity. First, it deploys monetization factors to estimate the monetary equivalence of all indicator values, which are scaled and weighted to achieve CBI indicator scores. Furthermore, based on multi-objective modeling that desires economic and social progression and environmental impact minimization, an aggregated CBI value is estimated from the weighted indicator scores. In this case, the environmental impact score is negative (bad score), while those for social and economic impacts remain positive (good score). The assumption is that a higher positive CBI value represents a pathway that provides relatively better social and economic benefits and appreciable environmental impact offset. It is, however, important to mention that the CBI value is not an absolute value but the aggregation of scores from different sustainability metrics with varying indicator conversion requirements. This presents a level of uncertainty in the calculation procedure. However, for communication reasons, CBI can be used to demonstrate the performance of selected pathways in relative terms. The following subsections intricately describe step-by-step procedures for CBI estimation.

a. Step 1 - Indicator monetization

The indicator monetization stage uses cost factors to translate the indicator values into their monetary equivalence. For environmental impacts, several monetization methods are available for cost conversion. The employed methodology should be explicitly stated at this stage for reference and reproducibility. Since the economic indicator, net profit, is already monetary, further conversion is not required. From the literature, per labor cost of unemployment ($\dot{\alpha}$) is estimated at £6,243 (approximately $7742) per year in benefits and lost tax revenue (Pettinger, 2019). Thus, the average number of employees over the pathway lifespan would be multiplied by the estimated cost of unemployment to achieve the unemployment cost reclamation value per year (U_{cr}), presented as equation 3.4

$$\text{Unemployment cost reclamation per year } (U_{cr}) = \frac{\sum_{j=1}^{n} Ej}{T} \times \dot{\alpha} \text{ ---------------------------------- } Eq\ 3.4$$

b. Step 2 - Indicator scaling

Scaling the indicator values across each indicator (criteria) allows intra-indicator comparison, but more so, provides a quantitative value representing the variance between the indicator results of the various pathways. Moreover, it emphasizes the subjective cost offsets of transitioning from one pathway to another. Herein, the indicator values were scaled down to their nearest hundredth relative to the highest indicator score. The estimation involved a ratio of the target indicator value (Xi) to the highest indicator value (X_{max}) multiplied by 100, presented in equation 3.5.

$$Scaled\ Value\ (Xs) = \frac{Target\ indicator\ value\ (Xi)}{X_{max}} \times 100 \text{ --- } Eq\ 3.5$$

c. Step 3 - CBI Estimation

In the final score aggregation, Unemployment cost reclamation (Ucr) and net profit (NP) scores are considered good scores, and the Global Warming Potential (GWP) score is considered a bad score. However, before aggregation, the scores are subject to weighted score ranges. Table 3.1 shows the score ranges with their defined assumptive weights. The assumptive weight is a subjective number consisting of 0, 0.5, and 1, which denotes the sustainability preference of the scaled indicator values. The mathematical equation for the CBI calculation is shown in equation 3.6. The following assumptions were made for good score weighting.

a. A weight of zero (0) represents an unsustainable region, showing that the target indicator score is 0-39% of the highest score (about 61-100 % lower than the highest score) for the given indicator.
b. A weight of 0.5 places the pathway in a partially sustainable region relative to the given indicator, also denoting that the indicator score is about contributing 40-69% of the highest indicator score.
c. A weight of 1 indicates that the target score is about 70-100% of the highest indicator score, also placing the pathway in a sustainable region.

Contrary to the good scores, where 0 and 1 marked sustainable and unsustainable regions, respectively, the contrary was modeled for bad scores. The assumptions are described below.

a. A weight of zero (0) places the pathway in a sustainable region, showing that the target indicator score is 0-20% of the highest bad score (about 80-100% offset relative to the highest score) for the given indicator.
b. A weight of 0.5 places the pathway in a partially sustainable region, also denoting that the pathway offsets about 51-79% of the highest indicator score.
c. A weight of 1 indicates that the target score is about 70-100% lower than the highest indicator score, also placing the pathway in a sustainable region.

CBI = $Wt.Ucrw + Wt.NPw - Wt.GWPw$ --*Eq 3.6*

Where Ucrw is the weighted Unemployment cost reclamation per year

NPw is the weighted net profit per year, and

GWPw is the weighted global warming potential per year

Table 3.1: Sustainability Scale for Scaled Circular Indicator Scores.

Circularity Indicators	Sustainable scores/Weight (Wt)	Partially Sustainable scores/Weight	Not Sustainable scores/Weight
GWPs (Environmental Impact)	0-20 Wt = 0	21-49 Wt = 0.5	50-100 Wt = 1
Unemployment Cost Reclamation (Social Indicator)	70-100 Wt = 1	69-40 Wt = 0.5	39-0 Wt = 0

Net Profit (Economic Indicator)	70-100 Wt = 1	69-40 Wt = 0.5	39-0 Wt = 0

d. Step 4 – Performance Description: CBI Scale

The CBI value can be found on a 7-point hedonic scale (-1, -0.5, 0, 0.5, 1, 1.5, 2) which dictates the degree of preference of one pathway to another. For example, negative one (-1) represents the worst scenario where all indicators are in the unsustainable region. In contrast, two (2) represent the best scenario where all indicator values are in the sustainable region. The intermediate scales (-0.5, 0, 0.5, 1, and 1.5) can arise from several circumstances, including good and bad indicator scores in partially sustainable or unsustainable regions. Some combinations are captured in Figure 3.4. Therefore, the indicators' performances must be rightly captured when communicating circular bioeconomy performance using the CBI scale.

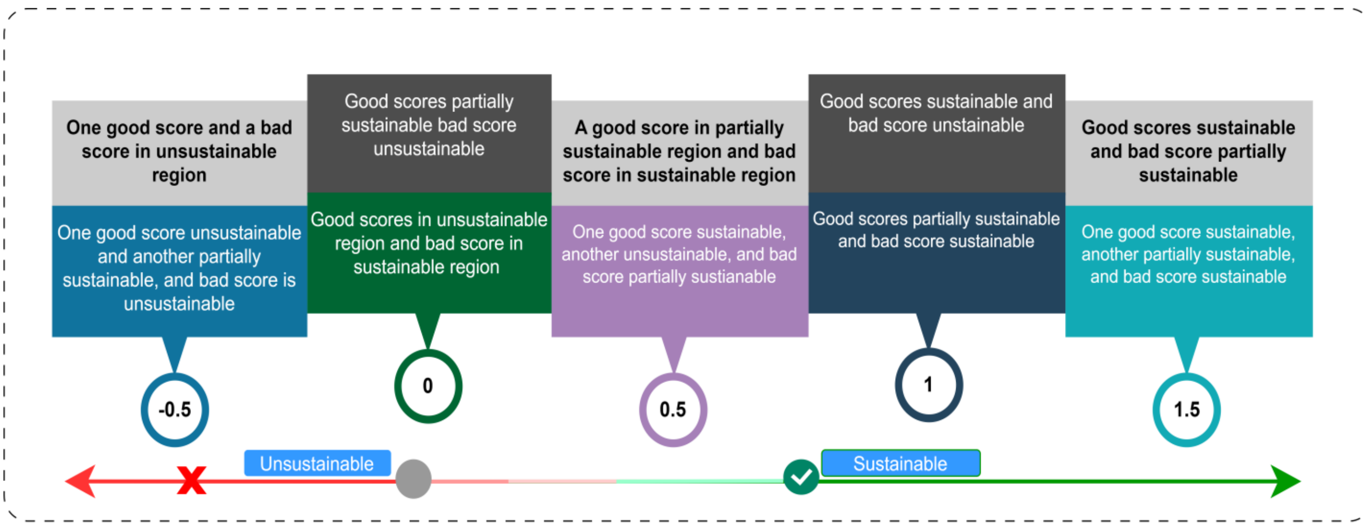

Figure 3.4: CBI Scale with possible combinations of the degree of sustainability preference

3.3.6.2 Multi-criteria decision analysis (MCDA)

Industrial and business decisions are usually characterized by considering several criteria or alternatives in making an exclusive judgment, description, prediction, or prescription that represents the holistic interest or satisfies vital performance indicators (Yalcin et al., 2022). In these instances, MCDA has evolved as an outstanding data-driven decision analysis tool for developing a numerical case for decision-making (Pérez-Dominguez et al., 2021). There are several MCDA approaches usually applied based on different factors. These factors include the ease of use, the number of criteria, the relationship between the criteria, the appropriateness and validity of the method, the sensitivity of results to the method (Pérez-Dominguez et al., 2021; Yalcin et al., 2022), the focal stakeholders, and decision making cognition for the selected criteria (Saaty & Ergu, 2016). Usually, most standalone MCDAs measure the distance of the optimal solution from the ideal or negative ideal solution (Yazdani et al., 2019) using either a linear relation or a summative or multiplicative aggregation algorithm. However, significant uncertainty has been identified using standalone MCDA approaches, premising the evolving developments in integrated MCDA. In this vein, Saaty and Ergu (2016) recommended using combined MCDA approaches for a single decision case whenever possible, emphasizing its advantages in substantially reducing uncertainties in decision-making and improving the precision of the identified optimal regions or decisions. In this theoretical model, we leveraged the combined capacity of highly rated, adaptable, and comprehensive MCDA approaches in evaluating the pathway alternatives based on the predefined criteria, improving social and economic benefits while reducing the environmental footprint. The Best-Worst Method (BWM) and Combined Compromise Solution (CoCoSo) approach were found to be comprehensive approaches upon review of the benefits they offer in the assessment of medium benefit, opportunity, cost and risk (BCOR) decision cases (Ecer & Pamucar, 2020; Liu et al., 2021; Rezaei, 2015; Saaty & Ergu, 2016; Ulutaş et al., 2020; Yalcin et al., 2022). BWM is a preferential criterion weighting method to other single vector methods such as SMART family and Swing, and full matrix methods like AHP (Analytical Hierarchy Process) due to its consistency in evaluating criteria using paired comparison. It leverages subjective data in analyzing judgements and behaviors of a human panel while offering a better avenue to check the consistency and reliability of the provided pairwise comparison (Ecer & Pamucar, 2020). The CoCoSo MCDA approach also presents a novel ranking methodology for identifying optimal solutions from a set of alternatives with predefined criteria. It intersects the strengths of the weighted product method (WPM) and weight sum method (WSM) in deriving a cumulative algorithm for rating decision alternatives (Ecer & Pamucar, 2020; Liu et al., 2021; Yazdani et al., 2019). Combining these two methods, that is, BWM and CoCoSo, promises an opportunity to draw optimal circular bioeconomy solutions closer to the ideal solution (Yazdani et al., 2019). In this framework, BWM and CoCoSo will follow the methodologies described by Rezaei (2015, 2016) and Javaid et al. (2022), respectively. The respective steps for the combined approach with corresponding mathematical models are summarized in Figures 3.5 and 3.6.

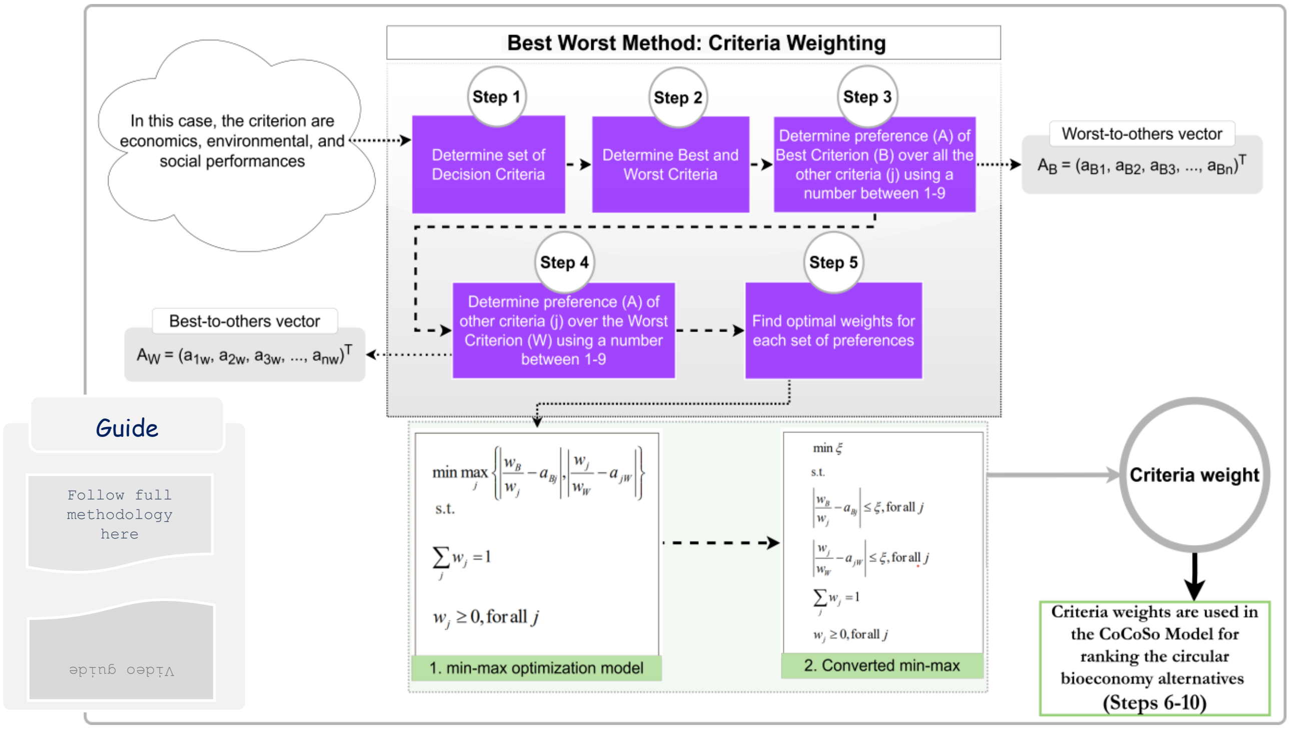

Figure 3.5: Graphical Summary of Linearized Best Worst Method for Criteria Weighting

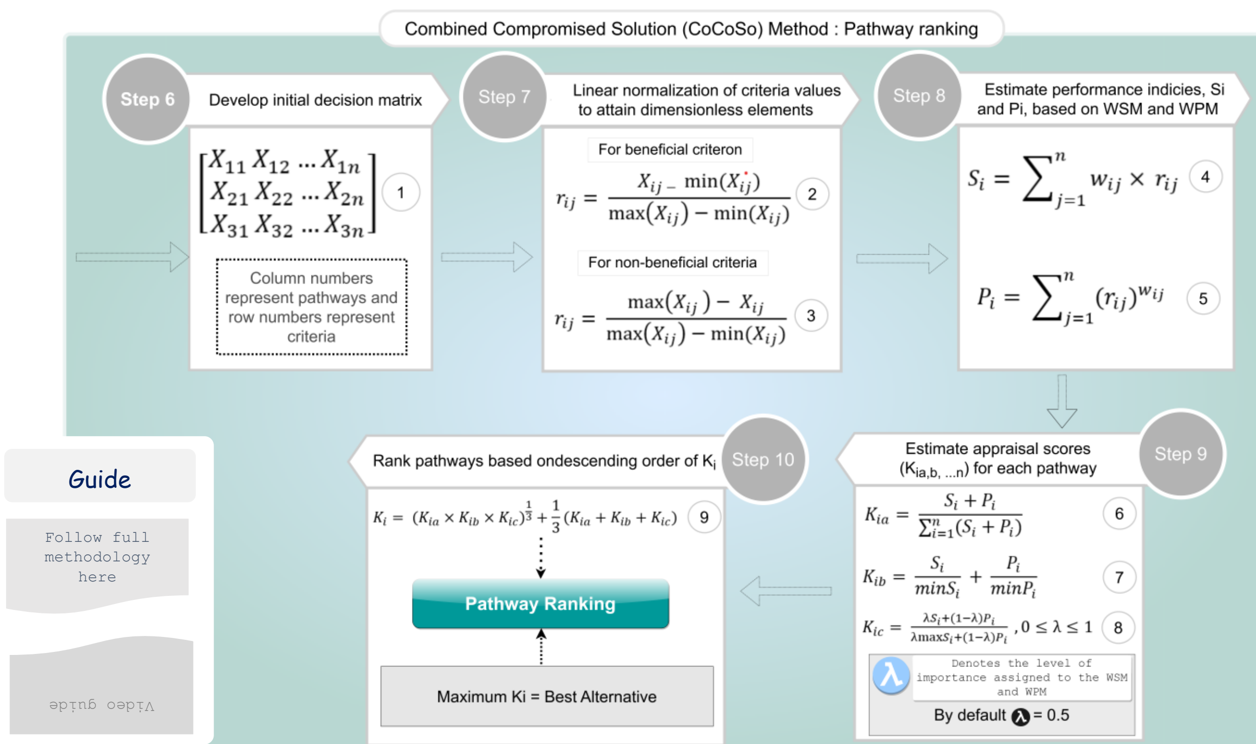

Figure 3.6: Steps for pathway ranking using the Combined Compromise Solution MCDA method

3.4 Conclusions

The study highlights the relevance of a comprehensive theoretical framework in advancing circular bioeconomy practice. The theoretical framework integrates multiple stakeholders, sustainability assessment, and robust decision-making to guide practice in favor of the interest of the diversified stakeholders of the agri-food system while promoting economic value, social equity, and reducing ecological implications. The single score circular performance value, the CBI, provides an excellent platform to communicate circular performance to laypersons and experts in more relatable numerical terms, allowing easy comprehension and nudging post-engagements in sustainable circularity pathways. However, in internal or business decision-making, the multicriteria decision analysis provides a systematic option for critically examining possible trade-offs based on varying decision cognition and assumptions described in selected MCDA approaches to draw more actionable insights. The study provides a preliminary yet comprehensive circular bioeconomy accounting tool to enhance practice in agricultural and agro-industrial systems and maximize the potential of possible outcomes in micro and macro-level policies. Policymakers, industries, and researchers could deploy it in subsequent strategies and engagements.

CONNECTING TEXT III

Chapter three discussed the limitations of the dominant linear economy, presenting arguments on the various schools of thought for economic growth and establishing circular bioeconomy as a redemptive approach for sustainable economic growth. It presented a detailed description of a circular accounting model, capturing stakeholder inclusion, life cycle sustainability assessment, and multicriteria decision analysis as critical components of the model for enhancing biocircular decisions. In chapter four, we follow recommendations of including life cycle assessment in circular bioeconomy practice, considering the number of studies emphasizing SCP's commercial prospects and future market value. A solid-state fermentation SCP baseline system is designed and assessed for environmental impacts using the ISO 2006 14040/44 guideline. Results from the baseline assessment informed several sensitivity analyses to identify the impact of different electricity and transportation scenarios on the baseline system for further system improvement. It was also necessary to compare the impacts of the baseline system with conventional protein systems like animal proteins and protein feed to ascertain the variations in sustainability performance and project the relevance of adopting the pea starch-based SCP over such systems in protein supply.

The manuscript for chapter four is currently under review in the Science of the Total Environment

4 CHAPTER FOUR: ATTRIBUTIONAL LIFE CYCLE ASSESSMENT OF PEA STARCH SCP

Designing Sustainable Circular Bioeconomy Solutions for the Pulse Industry: The Case of Pea-Starch Based Single Cell Protein

Abstract

Valorization of crude pea starch has become a key focus in the pea industry's sustainability pursuit. This study aimed to explore the circularity potential of crude pea starch in the production of Single Cell Protein (SCP). Following the ISO 2006:14040/44 standard, a life cycle assessment (LCA) was performed to ascertain the environmental performance and operational dynamics of baseline and scenario pea-starch SCP designs to identify optimal design considerations. Results demonstrated a higher relative contribution to marine ecotoxicity and human non-carcinogenic toxicity, with a relatively lesser contribution to global warming and land use. The harvesting process was identified as the hotspot, contributing about 85% and 98% to global warming and marine ecotoxicity, respectively. Generally, train and air freight were more sustainable than lorry freight, respective of mileage and mass. While hydropower demonstrated preference as a sustainable electricity source, it was also noteworthy that a lower hydro: wind power ratio showed better impact offsets. Regarding system alteration, eliminating the media enrichment process could offset about 26% of the land footprint, with a similar trend for most impact categories. Process benchmarking showed slightly lower land use impacts for pea starch SCP relative to fishmeal and soybean meal, with up to 99% potential offsets. Consequential LCA showed a general sustainability preference for substituting the protein-feed products with pea starch SCP, with a stronger emphasis on fishmeal substitution. Overall, these findings highlight the potential of pea starch SCP as a sustainable upcycling approach with substitutionary potentials for conventional feeds, recommending further exploration and commercialization in the long run. The pea industry could explore its sustainability potential by enhancing current practices while creating additional revenue and job streams to bolster its economic and social contribution.

Keywords: Pea starch, Single Cell Protein, Life Cycle Assessment, Circular Bioeconomy, Sustainability Performance, Consequential LCA

List of Abbreviations

Name	Abbreviation
Food and Agriculture Organization	FAO
Life Cycle Assessment	LCA
No Media Enrichment	NME
Organic and No Media Enrichment	ONME
Organic with Media Enrichment	OME
Single Cell Protein	SCP
Fine particulate matter formation	FPMF
Fossil resource scarcity	FRS
Freshwater ecotoxicity	FEC
Freshwater eutrophication	FET
Global warming	GW
Human carcinogenic toxicity	HCT
Human non-carcinogenic toxicity	HNCT
Ionizing radiation	IR
Land use	LU
Marine ecotoxicity	MEC
Marine eutrophication	MET
Mineral resource scarcity	MRS
Ozone formation, Human health	OZHH
Ozone formation, Terrestrial ecosystems	OZTE
Stratospheric ozone depletion	SOD
Terrestrial acidification	TA
Terrestrial ecotoxicity	TEC
Water consumption	WC

4.1 Background

The evolving need for global food security amidst rapid population growth has driven the dramatic shift toward agricultural innovations that enhance productivity, availability, accessibility, and efficient utilization of agricultural commodities (Alae-Carew et al., 2022; Daba & Morris, 2021). The emerging global sustainable food system transformation agenda is in response to this, characterized by the capacity to enable food and nutrition security while decoupling food system activities from severe environmental and social impacts. Hitherto, the global food system transformation has successfully driven stakeholder efforts toward novel designs with desirable sustainability benefits. However, there is an increased focus on intensifying sustainable crop and livestock production and advanced methods and technologies for catalyzing sustainable agro-processing (Hassoun et al., 2022a; Hassoun et al., 2022b). While such advancements are inarguably desirable, the supposed transformation agenda has not adequately tapped into the prospects of addressing other critical burdens with even severe economic and environmental disbenefits, presenting limitations to the sustainable food system trajectory. A typical example is the issue of food waste which significantly impacts people, the economy, and the planet. With a reported annual global food waste of about 931 million tonnes which can supposedly feed all hungry people in the world (Marchant, 2021), presenting an annual economic loss of about $1 trillion, and $3 trillion when social and environmental costs are considered (UNEP, D., 2021), it is clear that food waste is a major driver of food insecurity and economic instability. Moreover, food waste alone contributes about 8-10% to global greenhouse gas emissions, strongly correlating with the rising climate crisis and resulting impacts. As such, catalyzing revolutionary actions toward addressing food waste challenges demonstrates an opportunity to drive global capacity in reaching the triple benefits of food system sustainability. It presents an exceptional pathway to improving productivity, economic growth, and social equity within the global net-zero carbon target (Agyemang & Kwofie, 2021; Marchant, 2021; UNEP, D., 2021). Efforts in this vein have premiered the evolution of conceptual solutions like the circular bioeconomy, framed to foster sustainable biosystems through efficient resource utilization and alternation of agricultural or bioindustrial wastes for value creation (Aidoo, Raphael et al., 2022). In addition, experts assert circular bioeconomy as a sustainable revolution in waste recovery and management. Enabling biowaste valorization for food, feed, material, and energy strengthens global commitment to reducing fossil resource depletion and minimizing the rampant underutilization of valuable bioresources, allowing the offset of associated planetary implications while creating economically favorable business models (Feleke et al., 2021). These premises resound the necessity to expand the adoption of circular bioeconomy to solve the ever-increasing waste burdens of the booming agro-industry. Already, momentum is piling toward building a strong industrial aptitude towards circular business models, which have spiked innovations in biorefinery and other bioindustrial processes. Nonetheless, such traction has not satisfactorily evolved in the pea industry, triggering the authors' curiosity to explore such opportunity.

The pea industry has a vast potential for circular bioeconomy adoption. Pea consists of about 13-40% protein and approximately 50-60% starch, depending on variety and other geographic and ephadic factors (Ratnayakea et al., 2002). With the current business model favoring protein extraction (Allotey et al., 2023), large volumes (approximately 60-80%) of underutilized crude pea starch are generated (Daba & Morris, 2021; Emkani et al., 2021). In the business-as-usual, crude pea starch is purified or modified for various food, feed, and non-food applications. However, such approaches have not yielded substantial commercial success due to the competitive advantages of high-quality and functional starch alternatives like corn, wheat, and rice (Daba & Morris, 2021; Ren et al., 2021). This has instigated actions toward identifying other upcycling alternatives to improve their commercial prospects and sustainable practices in the pea industry. In this study, we hypothesized the benefits of exploring pathways that could simultaneously further the purpose of the expanding pea industry: enhancing resource availability to improve protein security while reducing environmental footprint. Through careful consideration of the literature, cellular protein agriculture was identified as an ideal choice, driven by its advantages in coupling sustainability with the compensation of protein deficits (Global Market Insights, 2022, 2023; Nyyssola et al., 2022). In the field of cellular protein agriculture, single cell protein (SCP) has accrued substantial traction, with several recent studies underpinning its prospects to revolutionize sustainable protein supply using relatively cheap substrates and microbes (Global Market Insights, 2023; Upcraft et al., 2021; Wada et al., 2022). SCP is simply the cultivation of protein-rich microorganisms on carbon-rich substrates through complex fermentation kinetics (Bratosin et al., 2021; Pereira et al., 2022; Upcraft et al., 2021). In the general evolution of biotechnology, solid-state fermentation (SSF) is projected as a greener technological option considering its relatively high productivity, low cost, and resource use efficiency (Areniello et al., 2022; Bajic et al., 2022; Martau et al., 2021).

In light of the stated hypothesis and supporting premises, the current study intends to explore the sustainability benefits of producing SCP from crude pea starch leveraging SSF as a green technological option in the proposed design. The sustainability assessment is divided into three parts. The first part focuses on the proposed crude pea starch-based SCP production system's baseline design and Life Cycle Assessment (LCA). The second part delves into understanding the system's sensitivity to process exclusion, energy modulation, and technological variations. It also identifies hotspot processes of the designed system and elaborates on the possible factors influencing their contribution to inform needed optimization. The third and last part compares the SCP system with benchmark scenarios, including prominent animal and plant protein sources.

4.2 Methodology

The LCA followed the recommendations and steps in the ISO 2006: 14040/44 standards. This section describes the four stepwise LCA procedures: goal and scope, life cycle inventory, impact assessment, and interpretation.

4.2.1 Goal and Scope

The study aimed to ascertain the environmental impacts associated with the production of SCP from crude pea starch slurry using solid-state fermentation. This is to provide baseline insights on the environmental footprint of a proposed system and identify operational hotspots for subsequent optimization and sustainable adoption. In addition, the findings were intended to convince industrial players of the sustainability potentials of integrating SCP as a sustainable value recovery strategy to improve the pea industry's contribution to local and global sustainability visions. A functional unit described as "quantity of crude pea starch slurry produced from wet fractionation of 1 tonne of pea protein, with a 20% protein yield and 3% production mass loss" was used.

4.2.1.1 System Boundary

The baseline system was designed to capture elements from the processing gate to the distribution gate (gate-to-gate approach). Manitoba, Canada, was selected as the preferred location for the SCP system in perspective. Thus, all system elements, including resource inputs and emissions, were modeled to represent the ideal situation in Manitoba. However, the absence of some region-specific flows for Manitoba in the ecoinvent database necessitated the adoption of other regionalized, globalized, and Rest of the World (RoW) data for some input flows like synthetic nutrients, transportation, amongst others indicated in the supplementary material (SM.4.2.A and B). Figure 4.1 represents the baseline product system, including all inputs and major outputs. In the baseline, SCP production was assumed as a downstream process at the pea protein processing site, requiring no feedstock transportation before processing. Thus, aside from the transportation of feedstock, inventory included all material, energy, and emissions attached to substrate preparation, enrichment, inoculation, incubation at 30 °C for 20h in an 8 m^3 tank, harvesting with a centrifuge, drum drying of the product to a 10 % moisture content, and storage of the final product. The raw slurry was assumed to contain about 50 % moisture and produced under aseptic conditions. Thus, substrate preparation primarily involved water addition to raising the moisture content to 70 % and mixing to achieve a uniform slurry before the solid-state fermentation process. The enrichment stage captured the addition of synthetic nitrogen (diammonium phosphate - $(NH_4)_2(HPO_4)$) and minerals (potassium phosphate – $KHPO_4$ and magnesium sulfate – Mg_2SO_4. For inoculation, facility-based inoculum production (Saccharomyces cerevisiae) was preferred. Thus, inventory for inoculation included material inputs for Yeast Peptone Dextrose (YPD - yeast extract, peptone, dextrose, and water), electricity inputs for a 4-day pre-culture incubation at 30 °C, and autoclaving at 120 °C for 15 minutes. All material and energy inputs associated with the main fermentation process were captured under incubation. Energy inputs considered electricity for aeration and heat energy for raising and maintaining fermentation temperature at 30 °C for 20h. All flows related to agitation were outside the system boundary, assuming no agitation for the fermentation set-up. Also, water for cooling the resulting biomass was included in the incubation inventory. The harvesting stage captured energy inputs for the centrifugation process, estimating an average energy input of 4.15 kWh per 1000 kg of fermented biomass (Najjar & Abu-Shamleh, 2020). Drying captured electricity for Storage considered electricity inputs for a 4°C storage temperature and a one-day storage time. For distribution, the baseline processing and uptake sites were designed to operate in a localized zone with an estimated mileage of 120 km. Thus, the distribution captured the conveyance of the final product (490 kg) to the assumed uptake site over an estimated 120 km mileage using a freezing reefer truck. The baseline product system did not include resource use and flows associated with waste management, asset establishment, and application of the SCP. The complete inventory, calculations, and assumptions for the product system and defined boundaries are elaborated in the supplementary material (SM4.A and B).

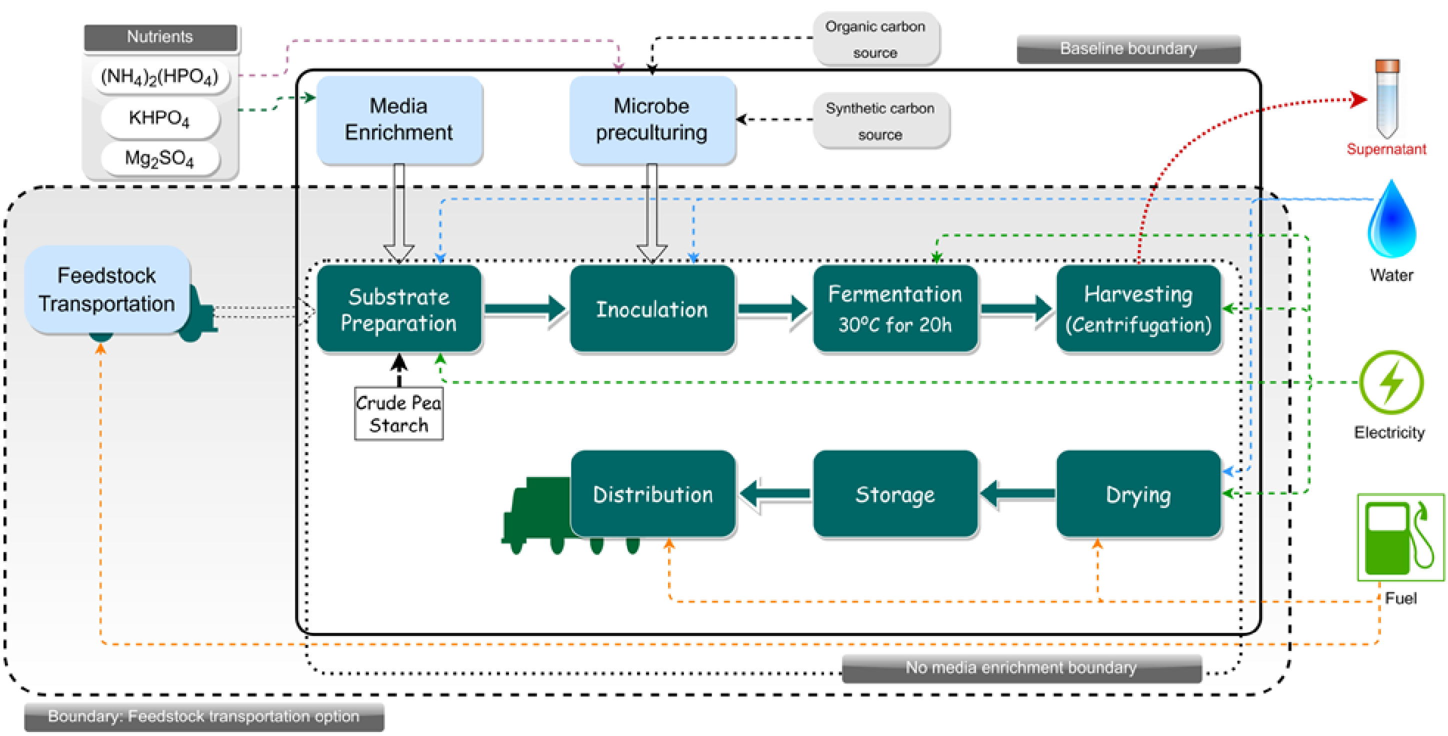

Figure 4.1: Process flow for solid-state fermentation of crude starch into Single Cell Protein

4.2.2 Life Cycle Inventory

The life cycle inventory data were consolidated primarily from available literature and the ecoinvent cut-off database (v 3.7.1). Quantitative estimation of resource inputs was based on linear estimations from similar LCA studies and the adoption of relevant equations with minimum-to-no modification. All equations for the calculation are detailed in the supplementary data sheet (The complete inventory, calculations, and assumptions for the product system and defined boundaries are elaborated in the supplementary material (SM.1.A and B). The inventory data were simulated in the OpenLCA software (v 1.11.0) and assessed using the ReCipe 2016 Midpoint (H) impact assessment method. The World 2010 (H) normalization and weighting set was used to normalize characterized impact results for comparative assessment.

4.2.3 Interpretational Analysis

4.2.3.1 Contributional Analysis

Contributional analysis, also known as hotspot analysis, is done to identify operational hotspots of the system in focus, which informs decisions on the necessary system changes or optimization for better system performance (Aidoo et al., 2023; Upcraft et al., 2021). This study performed further analysis to identify the dynamics in impact contribution among the unit processes considered. Such an analysis was necessary for identifying essential considerations for improving subsequent designs.

4.2.3.2 Sensitivity Analysis

Three sensitivity analyses were performed based on varying process assumptions, briefly described in the following subsections. The details are provided in the supplementary material (SM.4.2.C).

a. Unit process scenario sensitivity

Three other process scenarios were designed to test the system's reaction to unit processes and flow variations. The three scenarios include (1) elimination of media enrichment (SCP NME), (2) elimination of media enrichment while substituting synthetic carbon sources with organic carbon sources (sugar beet molasses) for inoculum production (SCP ONME), and (3) media enrichment and substitution of synthetic carbon source with organic carbon sources (sugar beet molasses) for inoculum production (SCP OME).

b. Transportation sensitivity

The transportation sensitivity was intended to test two transportation models in the system design to determine which transportation mode is environmentally beneficial. This is in response to the rising call to design and adopt sustainable fleet options and decouple the transportation sector from the persisting climate crises (Transport Canada, 2022). Thus, it was deemed reasonable to understand the trade-offs between; (a) downstreaming SCP production at the site of pea protein production (Transportation Scenario 1, TS1) and (b) establishing the upcycle system close to the primary point of use (uptake site), in this case, an aquaculture company (Transportation

Scenario 2, TS2). To further test the sensitivity of these scenarios, the SCP uptake sites were estimated to be farther from the pea processing site. Herein, an average mileage of 2600 km was assumed as the ideal distance for the sensitivity analysis, representing the distance between a new pea processing plant in Manitoba, Canada (Portage Prairie), and high aquaculture-producing provinces in Canada such as British Columbia, New Brunswick, and Nova Scotia.

c. Sensitivity for Electricity Source Modulation

With its dominance in the global climate emergency, sustainability discussions have reiterated the necessity to adopt sustainable energy sources. In this vein, the current study delved into testing the sensitivity of the SCP system to electricity variances, wherein dynamics in impact characterization in response to electricity variations were tested. The sensitivity analysis was performed by modifying the composition of geothermal, solar, wind, and hydropower sources in selected regional electricity supply. The regions considered include Manitoba (Baseline), British Columbia (ES1), Global (ES2), Europe without Switzerland (ES3), and India (ES4).

4.2.3.3 Process benchmarking

There is a growing discourse around substituting animal-based proteins and conventional feed products with microbial proteins as a complementary step toward transitioning to a sustainable economy (Humpenoder et al., 2022; Smetana et al., 2015). This impressed a reason to compare benchmarked animal proteins (beef and chicken) and aquaculture feed products (soybean meal and fishmeal) with pea starch SCP on a mass functional unit of 1kg. In this case, inventories for chicken, beef, fishmeal (63-65 % protein), and soybean meal (about 65 % protein) production systems were generated from literature and the ecoinvent database and assessed for their environmental impacts using the ReCiPe 2016 Midpoint (H) assessment method. The baseline benchmark scenario was generated from a backward mass and energy balance using a functional unit of 1kg of SCP with a 10 % moisture content and assumed 68 % protein content. The inventory data are compiled in the supplementary material (SM.4.2.D).

4.2.3.4 Consequential Scenarios

The ILCD handbook gives detailed insight into the type of LCA required in certain decision cases, emphasizing consequential LCA as a needful step, primarily when macro-level decisions are envisioned (JCR-IES, 2011). Therefore, two consequential scenarios were created to ascertain the consequences of substituting prominent aquaculture feed (fishmeal – FM; and soybean meal - SB) with pea-starch SCP. The substitution scenarios were formulated with equivalent and non-equivalent performance assumptions between the reference products and pea-starch SCP. Therefore, a mass of SCP was either assumed to bear an equivalent performance with the same mass of the reference feed (labeled as SB/FM100) or otherwise with substitutionary capacities of 25% (SB/FM25), 50% (SB/FM50), and 75% (SB/FM75). For instance, for the SB/FM25 substitutionary scenario, 1 kg of SCP was assumed to be equivalent to 0.25 kg of soybean meal/fishmeal. Thus, a four-fold mass of SCP would be required to satisfy the performance of a given mass of SB/FM in a proposed formulation. The complete inventory of the baseline and scenario assumptions are detailed in the supplementary material (SM.4.2.E and F). Also, it is important to mention that baseline and substitution scenarios were simulated using the ecoinvent consequential database (v3.7.1) as recommended for standard LCA practice (Hauschild et al., 2018; JCR-IES, 2011).

4.2.4 Results and discussion

This section describes the trends identified in the impact results and outlines related implications and relevant insights.

4.3 Results and Discussion

4.3.1 Baseline Results

Table 4.1 presents the impact results of the baseline SCP design. The normalization of impact results converts the characterized impact values into dimensionless and comparable values that show the relative contribution of the production system to the impact categories. The trend of the normalized results in Table 4.1 highlights a higher relative contribution of the baseline system to the toxicity categories, including marine ecotoxicity (MEC), freshwater ecotoxicity (FEC), human-carcinogenic toxicity (HCT), and human non-carcinogenic toxicity (HNCT), disposing of its relative potential to jeopardize planetary boundaries with high sensitivity to toxic emissions. Marine and freshwater ecotoxicities result from bioaccumulating toxic heavy metals like arsenic, cadmium, chromium, lead, mercury, and nickel, amongst others (Bello et al., 2019), which at severe limits distort the favorability of these environments, affecting the overall marine and freshwater ecosystems. Beyond displacement of the biodiversity in these systems, which may present global food security challenges due to dramatic losses in the aqua population, such toxic accumulation could go a long way to instigate toxicological public health risks. A possible risk pattern would be consuming toxic products that escape safety scrutiny to settle on consumers' plates, leading to bioconcentration and associated detriments (Kumar et al., 2022). HCT and HNCT result from anthropogenic pathways that maximize human exposure and inhalation of toxic metals, instigating their susceptibility to carcinogenic and non-carcinogenic detriments (Kumar et al., 2022; Nyambura et al., 2020). Existing public health studies correlate long and short-term exposure to heavy metals with potential cancer development, organ dysfunction, and death, mainly in severe cases (Bello et al., 2019; Jarup, 2003; Krishna et al., 2022). Some studies specifically associate lead and arsenic with leukemia, immune diseases, reproductive disorders, and renal, cardiovascular, and neurological complications, among a long list of diseases, which have coerced substantial global efforts toward confronting metal toxicity as a public health issue to minimize exacerbating impacts (Kumar et al., 2022).

While the above discussion echoes danger, its original intent is to renew our perspective of what it means for a system to be classified as toxic and to contextualize the vitality of enabling interventions toward optimizing toxic systems. On this basis, it is highly recommended that the baseline SCP system is optimized to reduce its contribution to the toxicity categories. The high toxicity contribution of the baseline system may be strongly connected with fossil resource consumption in almost all unit processes and other organic waste generation. These provide typical hotspots for subsequent system optimization and improvement, with possible considerations including adopting renewable energy and fuel sources and proper biowaste management techniques like recircularization. Unlike the toxicity categories, the baseline system seemed to have minimal impacts on other categories like land use (LU), global warming (GW), and fossil resource scarcity (FRS), which corroborates existing findings on the potential of SCP production to offset global warming, land use, and fossil resource scarcity burdens (Couture et al., 2019; Tallentire et al., 2018; Upcraft et al., 2021). This trend notably demonstrates the stance of pea starch-SCP as a remediative circular bioeconomy action for progressing evolving environmental remediation plans that target the reduction of global greenhouse gas emissions and protection of forest land zones. Moreover, it emphasizes the potential reduction in forest land clearance and fossil resource depletion that could be achieved from supporting commercial and research traction of the pea-starch SCP strategy. The process benchmark section of this study expounds on such benefits.

Table 4.1: Impact contribution of baseline SCP product system

Impact category	**Reference unit**	**Result**	**Normalized**
Marine ecotoxicity	kg 1,4-DCB	726.02	703.51
Freshwater ecotoxicity	kg 1,4-DCB	558.82	455.44
Human non-carcinogenic toxicity	kg 1,4-DCB	9649.97	64.75
Human carcinogenic toxicity	kg 1,4-DCB	82.34	29.72
Terrestrial ecotoxicity	kg 1,4-DCB	921.47	0.89
Freshwater eutrophication	kg P eq	0.13	0.20
Global warming	kg CO_2 eq	1272.59	0.16
Fossil resource scarcity	kg oil eq	74.02	0.08
Ozone formation, terrestrial ecosystems	kg NO_x eq	1.06	0.06
Water consumption	m^3	14.22	0.05
Ozone formation, human health	kg NO_x eq	1.05	0.05
Terrestrial acidification	kg SO_2 eq	1.06	0.03
Stratospheric ozone depletion	kg CFC11 eq	0.00	0.02
Fine particulate matter formation	kg PM2.5 eq	0.44	0.02
Ionizing radiation	kBq Co-60 eq	8.13	0.02
Marine eutrophication	kg N eq	0.03	0.01
Land use	m^2a crop eq	16.21	2.63E-03
Mineral resource scarcity	kg Cu eq	2.52	2.10E-05

4.3.2 Interpretational Analysis

4.3.2.1 Contributional Analysis – Baseline system

Figure 4.2 shows the relative contribution of various process flows to some selected impact categories. The harvesting process using energy-demanding centrifugal technology consistently demonstrated higher relative contribution to most impact categories, especially GW and toxicity categories. Taking the established connection between energy consumption and the emission of greenhouse gases and toxic heavy metals, pursuing innovations to optimize the energy use of harvesting technologies is critical for improving the impact offset of the proposed system.

In this study, synthetic minerals and nitrogen sources were considered for media enrichment in the baseline, which is the most probable reason for the high contribution of media enrichment to fossil resource scarcity (FRS) and land use (LU). This rightly portrays the aggravating burdens attached to the use of synthetic chemicals, as emphasized in several extant studies (Ayilara et al., 2023; Okagu et al., 2023). Most of these synthetic chemicals are extracted from fossil sources, contributing to the high exploitation of finite fossil resources and subsequent scarcity. We can assume the continuous clearing of forest reserves to extract these fossil resources and the building of refineries for chemical extraction as the major driving forces for land use. This instigates the argument of whether exploring other non-fossil nutrient sources is relevant. The following subsection provides relevant insights into how substituting synthetic carbon sources with organic carbon sources changes the impact dynamics.

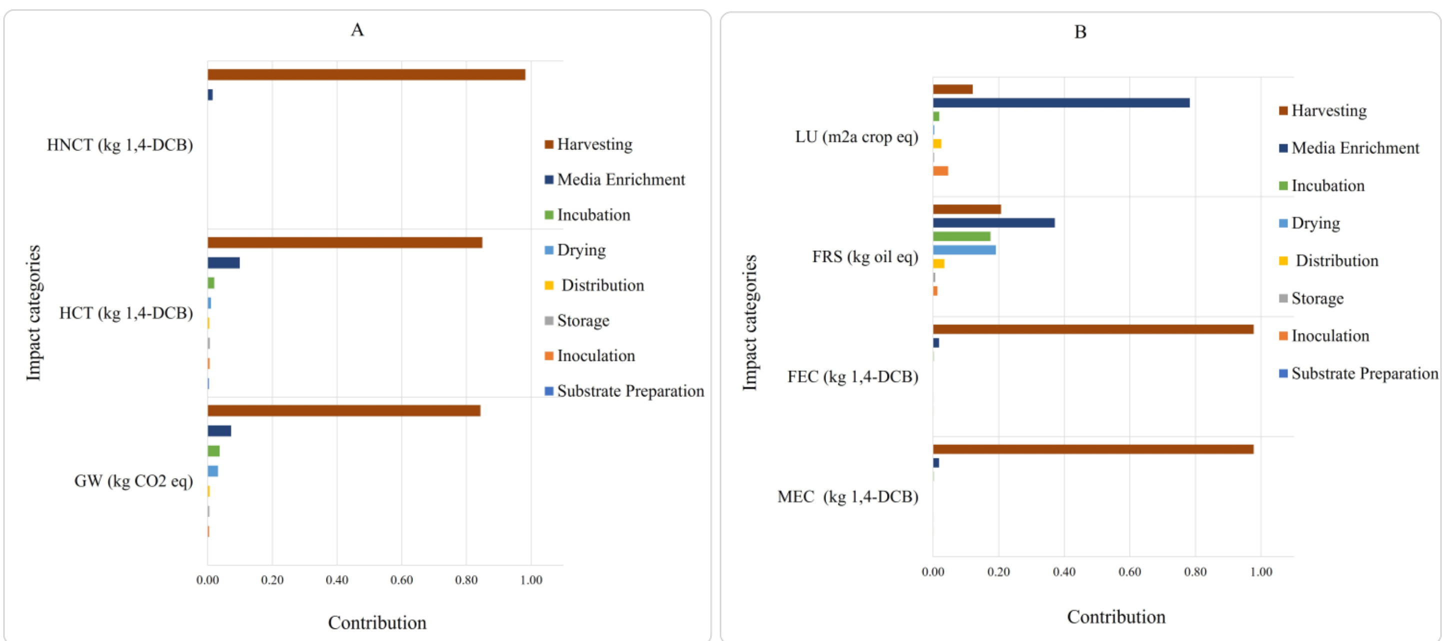

Figure 4.2: Process contribution to baseline impact

4.3.2.2 Process sensitivity

The impact results of the three process scenarios relative to the baseline system were presented in two parts. The first part (Figure 4.3) shows results for the impact categories with lower normalized scores, and the second part (Figure 4.4) shows trends for the top four impact categories on the normalized scale and two popularly discussed categories - global warming and land use. It is worth mentioning that the values were converted on a logarithmic scale to ensure a clear depiction of the trend identified. Generally, the scenarios without media enrichment, SCP NME, and SCP ONME performed relatively better in most impact categories. For instance, SCP NME and SCP ONME enabled about 25% and 28% offsets for land use, respectively, making them relatively better than the baseline scenario. These trends emphasize the high burden potential of the media enrichment process, signaling the relevance of innovating strategies that would facilitate sole dependence on second-generation substrates for all nutrients needed for the fermentation process or the reliance on renewable or organic nutrient sources. This assertion is further validated by the result for SCP OME, wherein despite the use of organic carbon sources for inoculum production, impact results were consistently high for most impact categories due to the media enrichment process. Whereas the previous assertion stands inarguable, the contradictory trend noted for terrestrial ecotoxicity (TEC) and ozone formation categories, where SCP NME and SCP ONME rendered about 200% and 61% additional impacts, respectively, compared to the baseline, asserts that a system cannot be ideally decoupled from environmental burdens. It supports the attainable region theory, which proposes that systems can be engineered to offer optimal and attainable environmental solutions. Thus, the recommendations for finding better media enrichment alternatives may not be solely feasible in areas or regions where ozone formation and TEC boundaries are already in their high-risk or danger zones. This further magnifies the relevance of mapping the impacts of a system against local, regional, or global boundaries, based on the scope and relevance of the system, to ensure the appropriation of redressive actions toward achieving optimal environmental benefits.

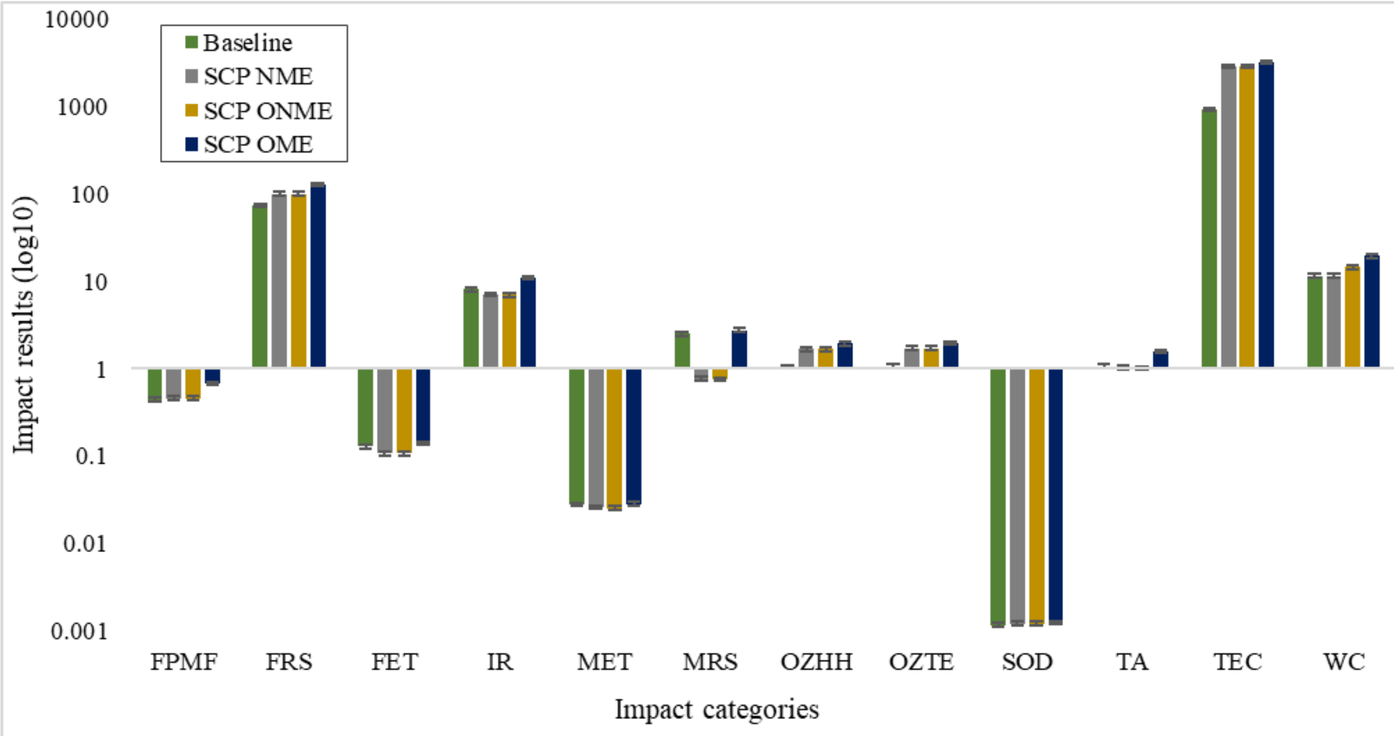

Figure 4.3: Contribution of baseline and process scenarios to impact categories with lower relative impacts

Key: SCP = Single Cell Protein, NME = No Media Enrichment, ONME = Organic carbon source (sugar beet molasses) for microbial pre-culturing and no media enrichment, OME = Organic carbon source (sugar beet molasses) for microbial pre-culturing and media enrichment

Like most impact categories, we find a similar trend for human non-carcinogenic toxicity and marine ecotoxicity in Figure 4.4. The "no media enrichment" scenarios, SCP NME and SCP ONME, had lower HNCT and MEC values than the baseline scenario, with a combined average of 67 kg 1,4-DBP eq HNCT value (about 0.7%) and 8.41 kg 1,4-DBP eq MEC value (about 1.16%) reduction relative to the baseline scenario. The trend further resounds the toxic metal emission potential of the media enrichment process, supporting the assumption previously made about the impacts of synthetic nutrients on ecosystem toxicity and carcinogenic and non-carcinogenic infections. Therefore, it also encourages using organic or renewable sources of nutrients for the enrichment and pre-culturing processes. The results of global warming and human carcinogenic toxicity, shown in Figure 4.5, were like terrestrial ecotoxicity and ozone formation impact categories, highlighting the baseline scenario to perform slightly better than the no media enrichment scenarios. However, the no media enrichment outperformed the scenario where media enrichment and organic carbon sources were synergized in the system design. Though this seems to argue the widespread assumption that using organic feedstocks and limiting synthetic nutrients improves sustainability performance, it highlights another insightful commendation for sustainable designs: optimal sustainability benefits could be achieved through well-optimized synergistic designs.

It asserts that undesirable combinations of process inputs can compromise sustainability performance and even distract the performance of beneficial inputs.

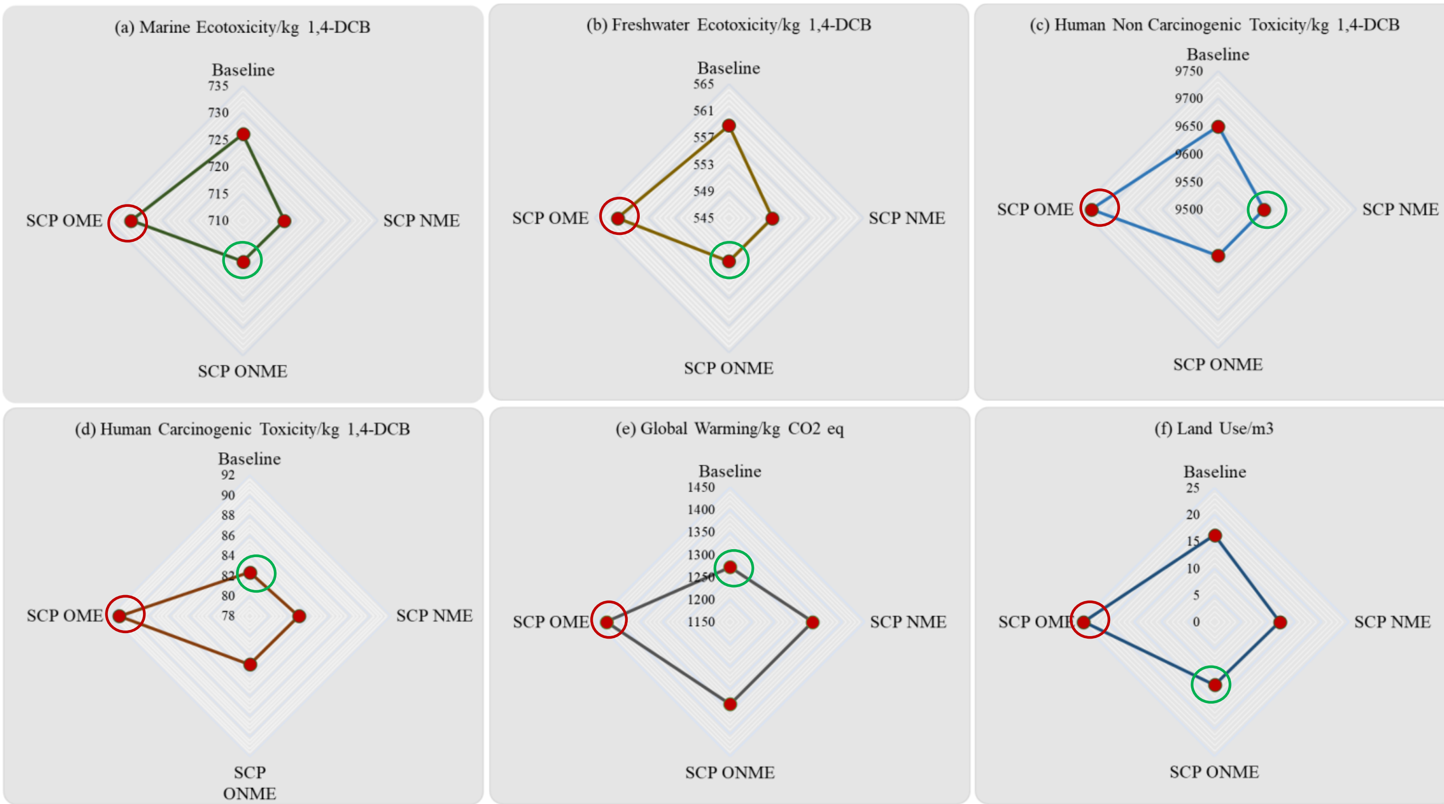

Figure 4.4: Contribution of baseline and process scenarios to impact categories with higher relative impacts

Key: MEC=Marine ecotoxicity, FE =Freshwater ecotoxicity, HNCT=Human non-carcinogenic toxicity, HCT=Human carcinogenic toxicity, GW=Global warming, LU=Land use [The red ring represents the scenario with the highest relative impact contribution; the green ring represents the scenario with the lowest relative impact contribution]

4.3.2.1 Transportation Sensitivity

a. Scenario and Mileage Sensitivity

The trends in Figure 4.5 highlight system performance considering earlier described transportation scenarios and mileage. Starting with the scenarios, we find TS2 performing better than TS1 across all impact categories, implying a preference for TS2 in achieving sustainable SCP production. For instance, TS2 could offset about 15 % of human-non-carcinogenic toxicity, 66 % of global warming, 79 % of land use, 88 % of fossil resource scarcity, and 90 % of terrestrial ecotoxicity relative to TS1. The trend suggests sustainability preference in undertaking a direct downstream of SCP at the point of co-product generation instead of transporting feedstock to other locations for the production process (Yang & Suh, 2015).

In terms of mileage, it was obvious that distance is directly proportional to impact results, where increasing distance resulted in high impacts across all categories, respective of the mass of the material, but irrespective of the mode of transportation. For instance, for all the impact categories, TS1 and TS2, which had a relatively higher distance, had consistently higher values than the baseline. For example, GW values for TS1A and TS2A were about 227% and 12%, respectively, higher than the baseline value. Similarly, GW values for TS1B, TS2B, TS1C, and TS2C were about 139%, 6.5%, 443%, and 56.53%, respectively, higher than the baseline value presented in Appendix A, Table A.2. The same trend was seen for the other impact categories, where an increase in distance similarly resulted in high impact values.

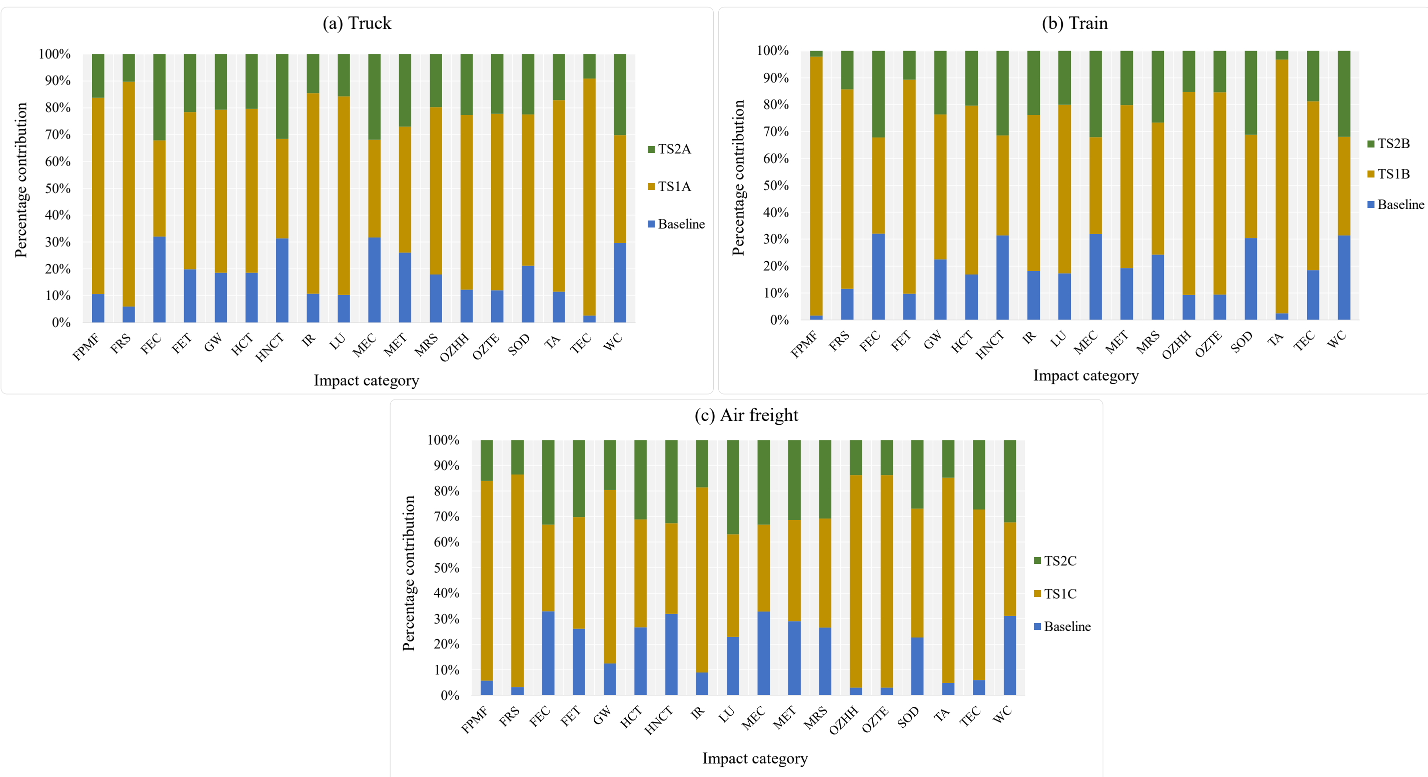

Figure 4.5: Percentage impact contributions of transportation scenarios to impact categories

[Key: TS denotes transportation scenario; 1- denotes the transportation of feedstock from the point of co-product generation to the point-of-use (where the final product will be utilized) for the production of SCP (an average distance of 2650 km); 2 - denotes the production of SCP at the site of co-product generation and transportation of final product to the point-of-use; A - denotes transportation by truck; B - denotes transportation by train; C - denotes transportation by air freight]

b. Sensitivity of transportation scenarios to food mass

It was considered relevant to capture how variation in feedstock (substrate) or product (SCP) mass changed the impact dynamics of the transportation scenarios. Figure 4.5 a-c depicts an explicatory trend where TS2, which had a relatively low mass (about 490 kg), consistently scored lower impact values than TS1, with a mass of about 4900 kg. These trends assert two practical perspectives for a product system's transportation or distribution modeling. First, when higher food mass is to be transported over a long distance to produce a product of relatively lower mass, it would be sustainably prudent to integrate the upcycle system into the co/by-product generating system, taking such an option is available. The findings of Areniello et al. (2022) and Kobayashi et al. (2023) buttress this perspective, also portraying better environmental benefits when bioethanol, biogas, and SCP production were integrated into the waste-generating system or plant.

c. Mode of transportation

The selected geographic boundary operates varying freight: truck, train, and air. Thus, the study also considered ascertaining how each freight system affects variation in impact values. Figure 4.6 showcases a relevant trend on how variation in transportation mode affects the environmental impact contribution of the baseline SCP product system. The relationship between the mode of transportation and impact category varied with each scenario, demonstrating some benefits and disbenefits across the impact categories, influenced primarily by mass, mileage, the target category, and probably other factors beyond the scope of this analysis. For instance, whereas an increasing global warming trend of the train (B), lorry (A), and air freight (C), thus, B<A<C, was noted for both TS1 and TS2, suggesting train as a better transportation mode for reducing greenhouse gas emissions regardless of mileage and mass, a contrary trend was identified for land use. Herein, an increasing land footprint trend of C<B<A and B<A<C were identified for TS1 and TS2, respectively. Thus, while air freight portrays land use offset advantages for high mileage and mass transports, such advantages seem compromised when relatively low mileage and mass are considered. This suggests that the transport mode selection should carefully parameterize food mileage and mass. The position of lorry freight for the land use trend is rational because road infrastructure for lorry transportation demands a high land area relative to air and train freight. Thus, lorry or truck transport is more likely to demand clearing a large forest land area for road infrastructure for the same mileage as air or train freight. These variabilities in trend highlight the relevance of transportation modeling in sustainable system design. Furthermore, it emphasizes the need to consider geographic or regional sustainability diversity, product mass, and mileage in environmental modeling.

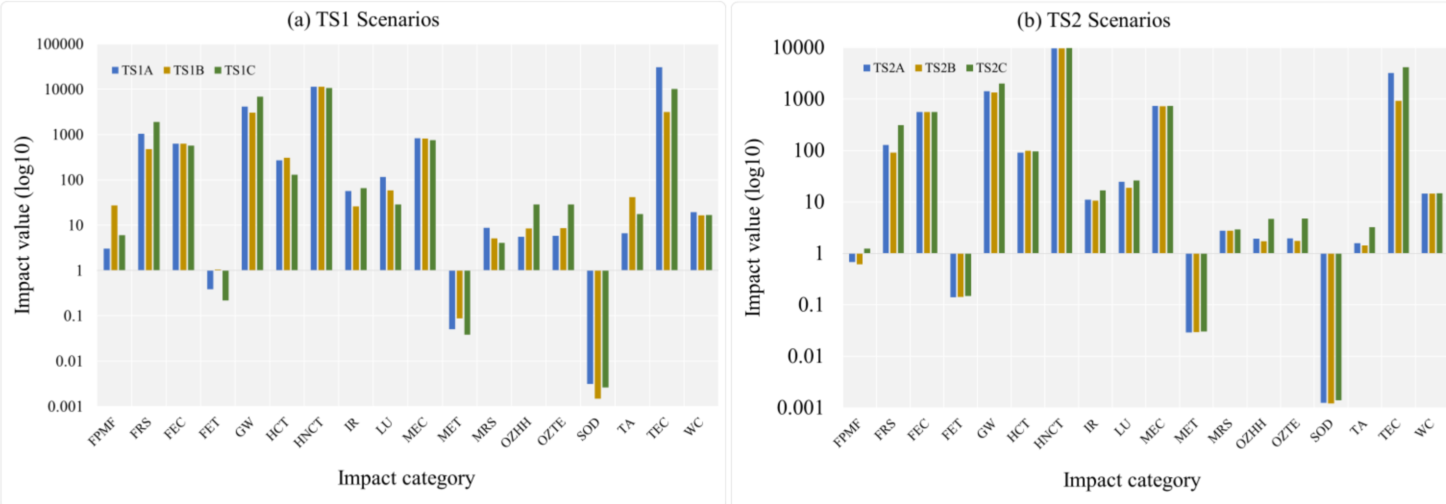

Figure 4.6: Impact dynamics for changes in transportation modes

4.3.2.2 Electricity Sensitivity

The electricity sensitivity was modeled to determine how regional variance in the electricity sources would affect the sustainability performance of the baseline SCP product system. In addition, it was meant to provide insight into the relative impact variation in building the SCP plants in these regions.

The contributions of different energy sources to the varying energy supply are presented in Figure 4.7. Herein, an explicit variation in electricity generation was identified amongst the selected providers. The variation seems to correlate with resource availability and explorative capacities in these regions, not overemphasizing the subtle influence of geopolitics and other inherent geographical limitations. For instance, Canada is recognized to have the fourth-highest volume of freshwater bodies, which has defined a hydropower energy trend in most Canadian provinces. This conforms with the position of Manitoba (97%) and British Columbia (95%) in hydropower contribution to the energy mix against about 77% global adoption and 57-72% for the other selected providers. Europe (~40%) and India (~27%) dominate wind power contribution, probably due to improved infrastructure and good wind speed and density for high energy generation (Zhou et al., 2023). Solar power is less adopted amongst the selected providers, presumably driven by high upfront cost, large land space requirements, and concerns with reliability in energy storage (Basit et al., 2020). Europe has relatively progressed in geothermal energy exploration, with about 2.3% contribution to their electricity mix compared to 0.002% for Manitoba, 0.74% for British Columbia, and 0.76% for India. Recent recommendations emphasize geothermal energy as an environmentally sustainable energy source that could be leveraged to minimize the impacts of fossil energy exploitation (Palomo-Torrejón et al., 2021). Thus, while the global transition to clean energy persists, geothermal energy could be explored amongst other renewable energy alternatives to support sustainable national and industrial energy utilities. However, wide adoption is limited primarily by the high upfront cost, about three to four folds that of solar and wind power infrastructure (energysage, 2021). Other impeding factors are the geographic specificity constraints – mostly requiring areas with reservoirs above 100 °C for large plants, and a possible influx of land catastrophes like earthquakes attached to the deep underground drilling activities (energysage, 2021).

	Hydropower	Solar	Wind	Geothermal
Manitoba (Baseline)	0.97	0.0000027	0.03	0.00002
British Columbia (ES1)	0.95	0.0000043	0.04	0.00741
Global (ES2)	0.77	0.0000348	0.21	0.02741
Europe Without Switzerland (ES3)	0.57	0.0000603	0.40	0.02337
India (ES4)	0.73	0.0000503	0.27	0.00076

Figure 4.7: Contribution of varying energy sources to the regional electricity supply

Key: Deep dark-blue to light dark-blue color shows decreasing order of source contribution

Figure 4.8 a-d highlights the baseline electricity mix as the most sustainable, maintaining its relatively lower contribution across most impact categories, except for high water consumption due to the dependence of hydropower energy generation on freshwater bodies. Notably, reducing the contribution of hydropower was consequent to increasing impact values. For instance, 1.75% (ES1), 23.30% (ES2), 39.32% (ES3), and 23.90% (ES4) reduction in hydropower contribution increased global warming values by 4%, 11%, 6%, and 25%, respectively, relative to the baseline. Likewise, the same reduction trend increased land use values by 9%, 13%, 14%, and 22%, respectively, and human carcinogenic toxicity values by 3%, 7%, 6%, and 16%, respectively. On the other hand, the same reduction trend decreased water consumption by 15%, 41%, 34%, and 43%, respectively, which is quite reasonable due to the linear correlation between hydropower use and water consumption. Another relevant insight is the dynamics presented by the hydro-to-wind power ratio. A lower ratio (hydro and wind are predominant) mostly implied better impact offsets, especially for global warming. For instance, ES3, with a hydro-to-wind power ratio of 1.42, had global warming, land use, and human carcinogenic toxicity values of about 17%, 7 %, and 9% lower than values for ES4, with a ratio of 2.67.

Similarly, the global warming value of ES3 was about 5% lower than ES2, with a hydro-to-wind power ratio of about 3.7. This portrays a critical pursuit that could change the energy paradigm in regions with lower volumes of freshwater bodies for hydropower generation. Here, The opportunity is to improve wind power infrastructure and harness its potential to improve energy generation's overall sustainability, taking favorable conditions. Nonetheless, the reliability of hydropower in achieving better environmental offsets was outstanding, highlighting how regions with high volumes of freshwater bodies could harness these renewable potentials toward addressing climatic crises or minimizing regional environmental footprints from energy generation and consumption, notable of sustainability pursuits in Canadian provinces. Besides, this places Canada in a prime position to expand and possibly lead sustainable SCP integration into the pea industry, considering its leading stance in pea production and the availability of high volumes of freshwater bodies for renewable energy generation.

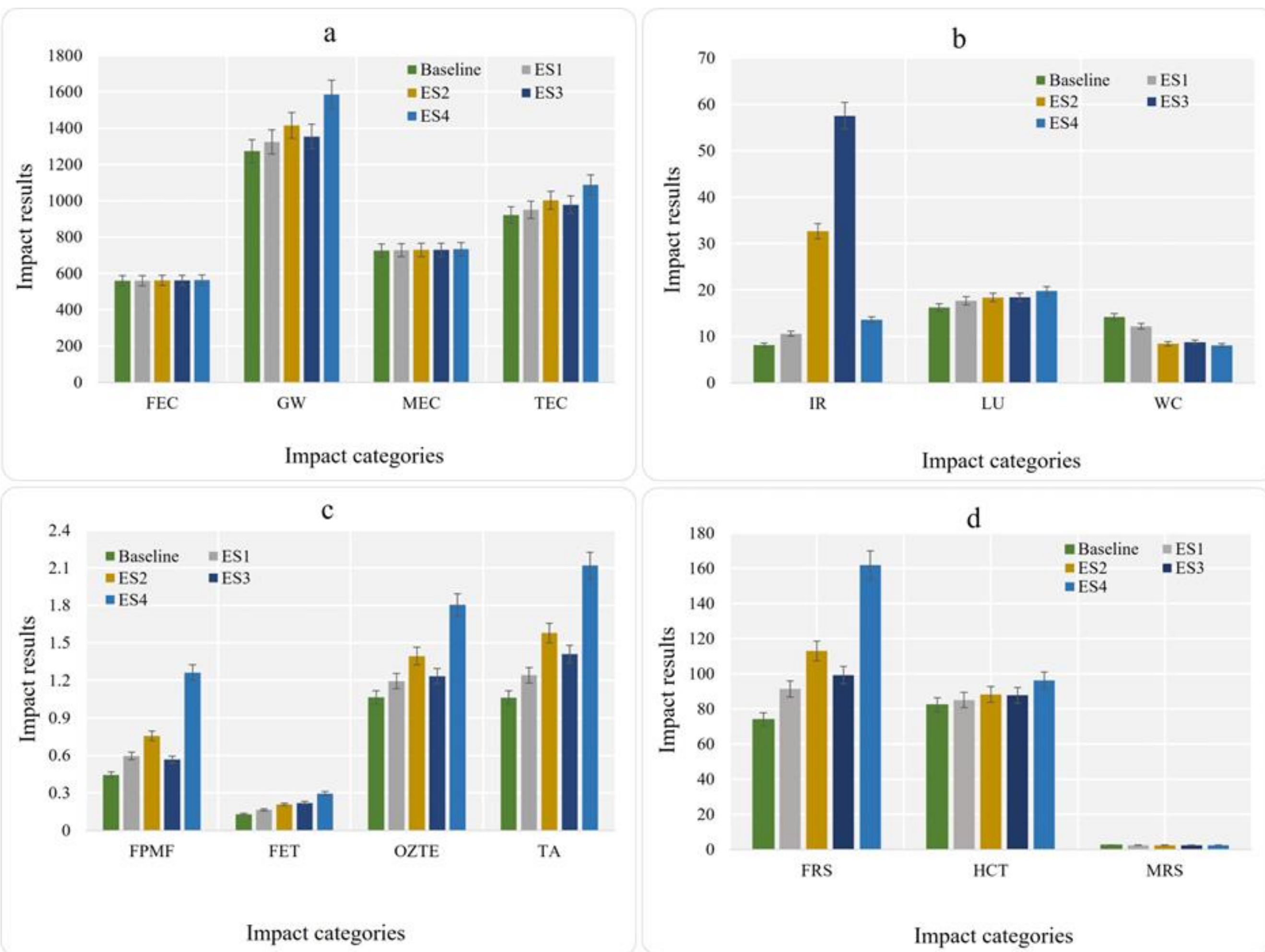

Figure 4.8: Impact dynamics for electricity sensitivity

4.3.2.3 Process Benchmarking

The results for the benchmark analysis (Appendix A, Table A.4) are reported per kg of the benchmark systems as previously described in section 4.2.3.3. The trends in Figures 4.9 and 4.10 highlight some variabilities in the performance of the selected product systems. First, they emphasize that the impact variations were not necessarily linear, with the SCP scenario outperforming the benchmark systems in some impact categories and poorly or similarly in others. For instance, whereas the baseline SCP scenario performed better than all the benchmark scenarios in LU and terrestrial acidification (TA) categories, with about 71-99.97% and 9-98% less impact values per kg of product, a negative performance was recorded in WC, MEC, and FEC categories. Herein, the impact values for the SCP scenario in these categories were about 52-99%, 85-99%, and 80-98% more than the benchmarks, respectively.

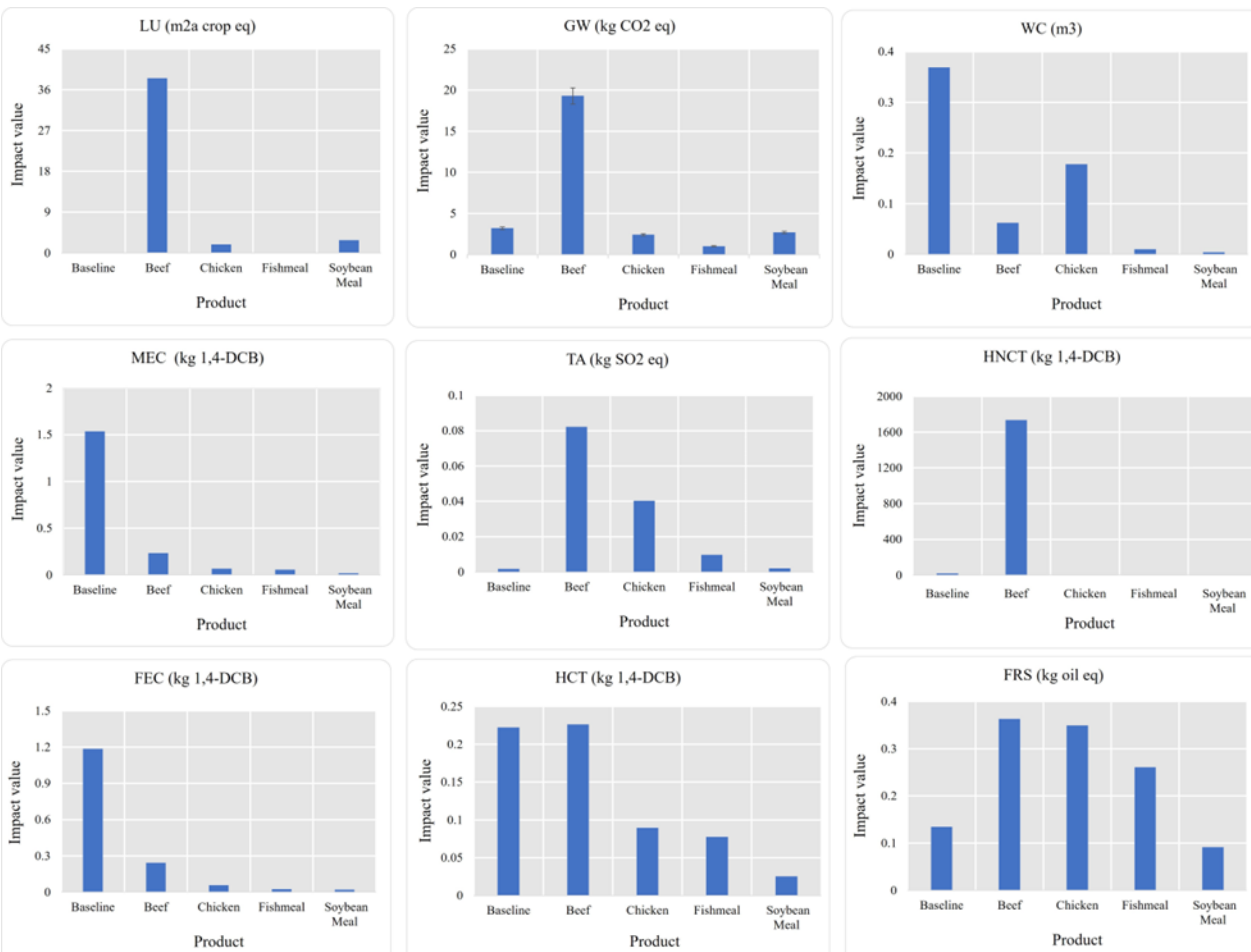

Figure 4.9: Comparison of Benchmark scenarios to Pea Starch-SCP

SCP surpassed the animal-based proteins and fishmeal in the FRS category while performing relatively lower against soybean meal. On the other hand, SCP performed significantly better than beef in the GW and HNCT categories, with about 83% and 99% less impact values, respectively, while performing relatively lower than the chicken and other feed production systems. Whereas inferences about the pre-eminence of chicken and the feed production systems in achieving targeted reductions in the understated categories can be emphasized, the slight deviation between the values of SCP and the chicken and feed systems signifies SCP with substantial relevance in progressing this climate recovery agenda. The eutrophication trend in Figure 10 agrees with the findings of Maiolo et al. (2020), which also noted performance variabilities among feed across impact categories but identified SCP feed to perform favorably in the eutrophication category relative to insect meal, fishmeal, and poultry-by-product. The benefits attached to reducing eutrophication potential are vast, spanning from enabling the environment to sustain aquatic habitats to promoting biodiversity in the long term.

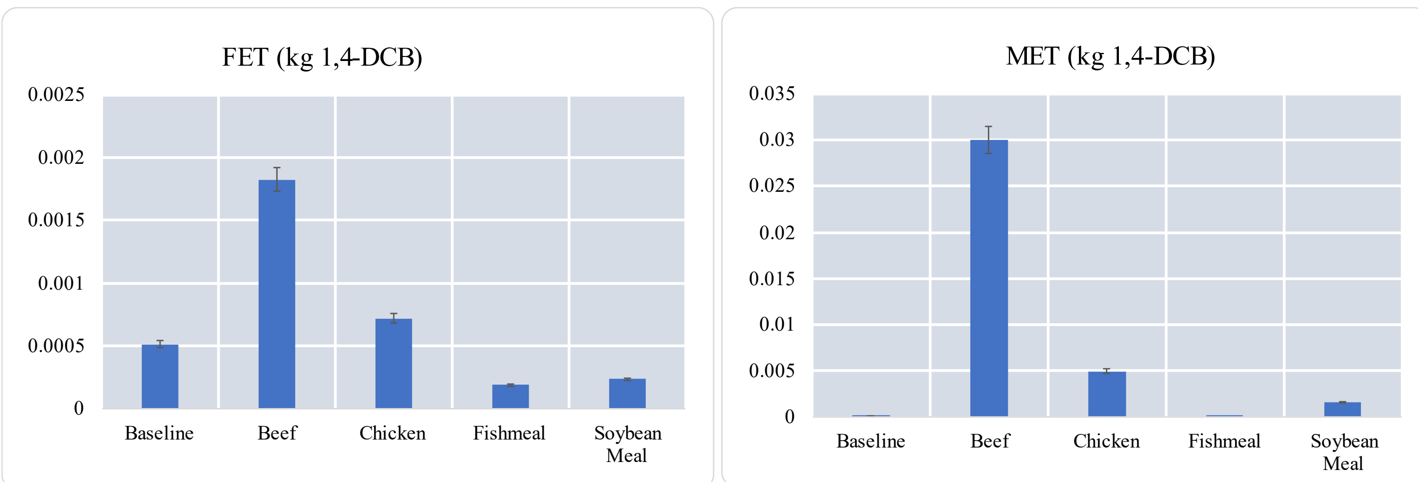

Figure 4.10: Contribution of eutrophication impacts of baseline scenario to benchmark scenarios

Overall, this assessment supports the report by Owsianiak et al. (2022), which indicated limitations in SCP's sustainability benefits, respective to the system's physical flows. Furthermore, the findings caution practitioners against assuming the pea starch SCP strategy as entirely sustainable. Instead, it highlights it as a feasible circular pathway with exclusive sustainability potential, which requires further optimizations, improvements, and radical collaborations to maximize sustainability and commercial benefits.

4.3.2.4 Consequential Impacts

The previous sections have broadly described the implications of the impact results generated for the defined product systems. They pinpointed the alignment of the SCP pathway to sustainable protein production. However, they heightened the need for critical improvements to enhance sustainability benefits. At this point, it is vital to establish that the previously discussed results were in the view of attributional LCA, which only demonstrates an estimated portion of global environmental burdens associated with the physical flows of a given product system (Ekvall, 2019). Thus, despite the inarguable relevance of attributional LCA in interrogating and comparing environmental performances, its preference is limited in advanced cases where results would inform meso and macro-level decisions (Aidoo et al., 2023), presenting some limitations to the earlier analysis. In such situations, consequential LCA (CLCA) is highly recommended as a robust methodology for enhancing decision-making. CLCA progresses the benefits of attributional LCA and places environmental assessment in a context that encapsulates how a given product's production and subsequent use drive variations in global environmental burdens (Finnveden et al., 2009). In this case, results accurately depict the actual environmental transgressions attached to dynamics in utilization decisions which could induce precise utilization or adaptative strategies (JCR-IES, 2011). Following such recommendations, the study dived further into performing a CLCA to measure the outcomes of defined decision cases that involve the utilization of SCP as a feed replacement for soybean meal and fish meal. Such outcomes emphasized the shortfalls of attributional LCA, demonstrating substantial variation in impact trends relative to consequential modeling, also confirming CLCA as a better methodological option for reflecting the actual impacts of processes or products. The following subsections demonstrate these trends per reference amount of 1kg of dried pea-starch SCP.

a. Baseline against soybean meal substitution scenarios

Figure 4.11 highlights changes in impact results for HCT, HNCT, MEC, and FEC categories regarding the variation in soybean displacement levels. While results for HCT (about 20-80% increase) and HNCT (<2% increase) slightly increased with increasing percentage substitution, the contrary was noted for MEC and FEC, which slightly decreased (<2 %) with a relative percentage increase in substitution. These trends attach limitations to the adaptability of such substitution scenarios, especially in geographic regions where HCT and HNCT categories are generally high and are in their danger zones. Also, significant changes in GW and LU categories were identified, with the trends underlining a direct cause-effect relationship between defined decision cases and the burden potential of these categories. An inversely proportional relationship dictating substantial GW and LU offsets relative to an increase in percentage substitution was identified, also asserted by Owsianiak et al. (2022). Herein, SB100 outperformed all other substitution scenarios in both impact categories, including the baseline. For instance, a 99% decrease in GW impacts was noted for SB100 compared to about 75% for SB75, 50% for SB50, and 25% for SB25, relative to the baseline (no substitution). These findings define the magnitude of GW and LU offset awaiting the adoption of pea starch-SCP as a sustainable substitute for soybean meal in feed production (Couture et al., 2019; Elyasi et al., 2021; Tallentire et al., 2018). Moreover, they heighten such consequential adoption as a strategic and adaptable frontline action towards abating the exacerbating climate crises and augmenting sustainable circular bioeconomy practice, especially in pea production and processing regions, together with the parade of emerging sustainable innovation.

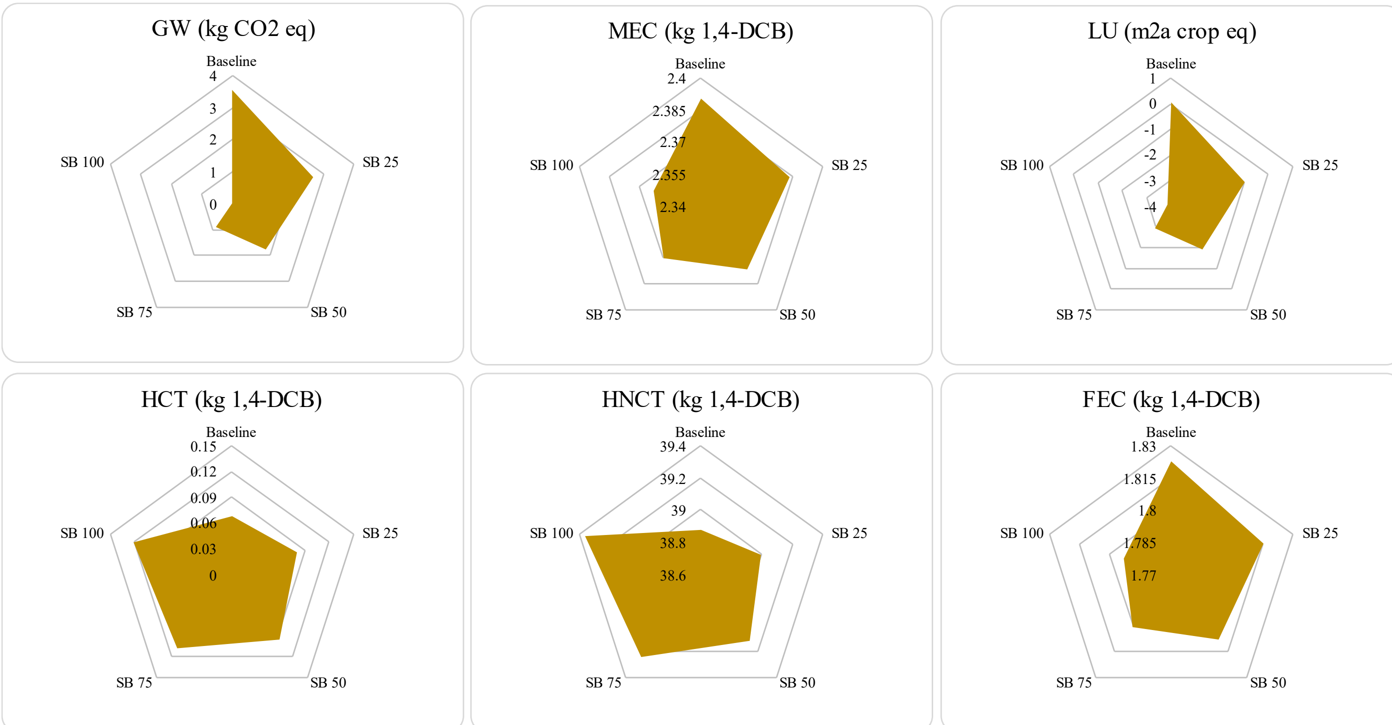

Figure 4.11: Comparison of Baseline System to Soybean Substitution Scenarios

b. Baseline against fishmeal substitution scenarios

While soybean meal substitution increased HCT and HCNT and slightly decreased impacts on MEC and FEC, Figure 4.12 displays a contradictory trend for fishmeal substitution, wherein impact offset was achieved across the displayed impact categories relative to the baseline, except for land use. About 0.9-3.7% (FEC), 1.1-4.5% (HNCT), 1.3-5.1% (MEC), 18-73% (GWP), and 70-280% (HCT) offsets were achieved for the defined substitutions, with higher substitution rates enabling higher offsets (refer to Appendix B, Table B.2). Conversely, LU values for fishmeal meal substitution scenarios were more than double fold of the baseline values. This trend highlights the requirement of a relatively high land area for substituting fishmeal with pea-starch SCP, informing the potential exacerbation in land footprint for fishmeal substitution compared to soybean meal substitution. Explicably, being a water-based industry, fishmeal production ideally requires less land space than SCP, prompting a dramatic increase in the land area required to produce adequate amounts of SCP to satisfy the needed substitution. On the other hand, the SCP requires relatively low land space than soybean meal, resulting in a more substantial land use offset for soybean meal substitution, as shown in Figures 4.11 and 4.12.

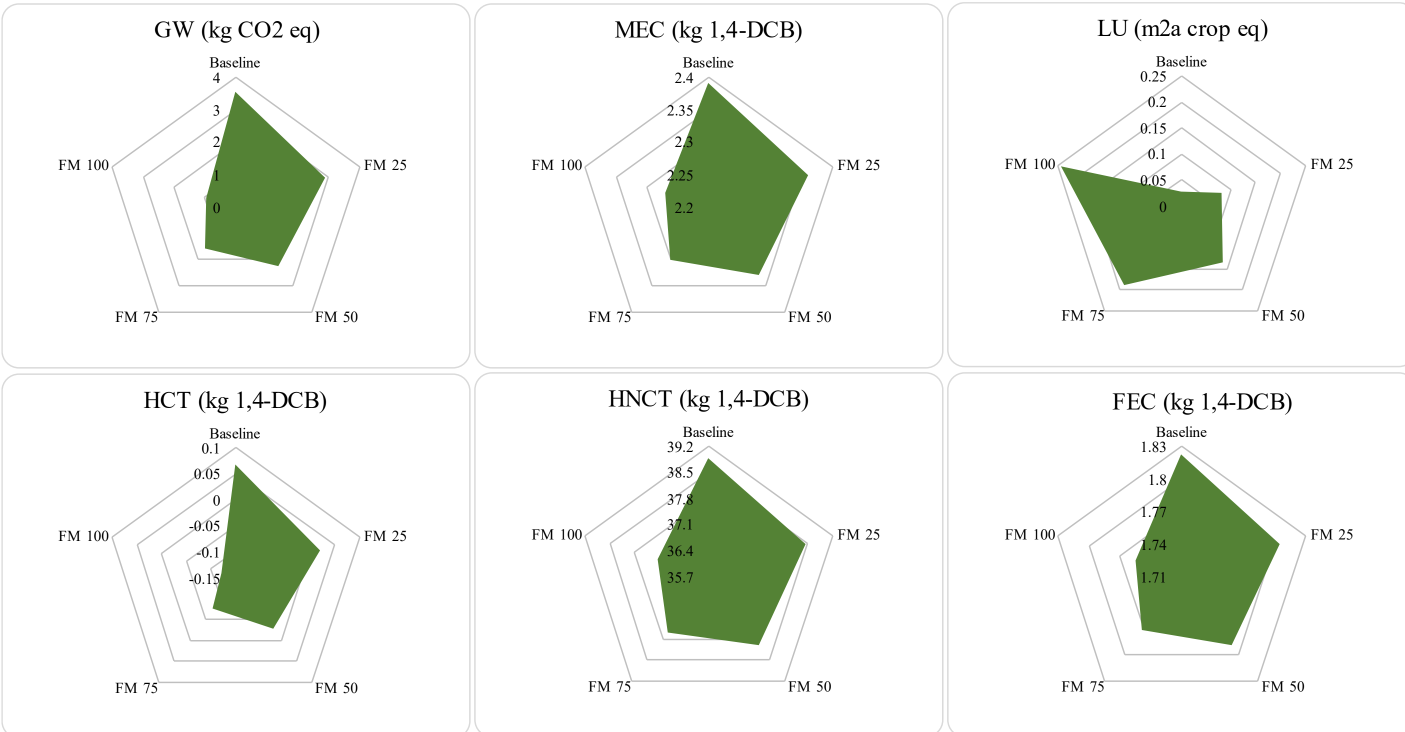

Figure 4.12: Comparison of Baseline System to Consequential Fishmeal Substitution Scenarios

While many assertions lie within the understated trends, particular interest is found in how they depict the distinction between soybean meal and fishmeal substitution in SCP utilization. First, they delineate the diversities in environmental incentives attached to the two substitution scenarios, encouraging the prioritization of local sustainability outlooks in their adoption. However, they underscore the environmental benefits of fishmeal substitution over soybean meal substitution, noted in its high impact saving potential toward FEC, MEC, and HCT, out of five impact categories considered, as shown in Figure 4.13. Overall, the consequential scenarios depicted an interesting correlation between product characteristics, level of substitution, and potential impacts, relating the level of offset or impact exacerbation to the relative variation in characteristics of the substituting product and the potential substitution capacity (Khoshnevisan et al., 2020; LaTurner et al., 2020).

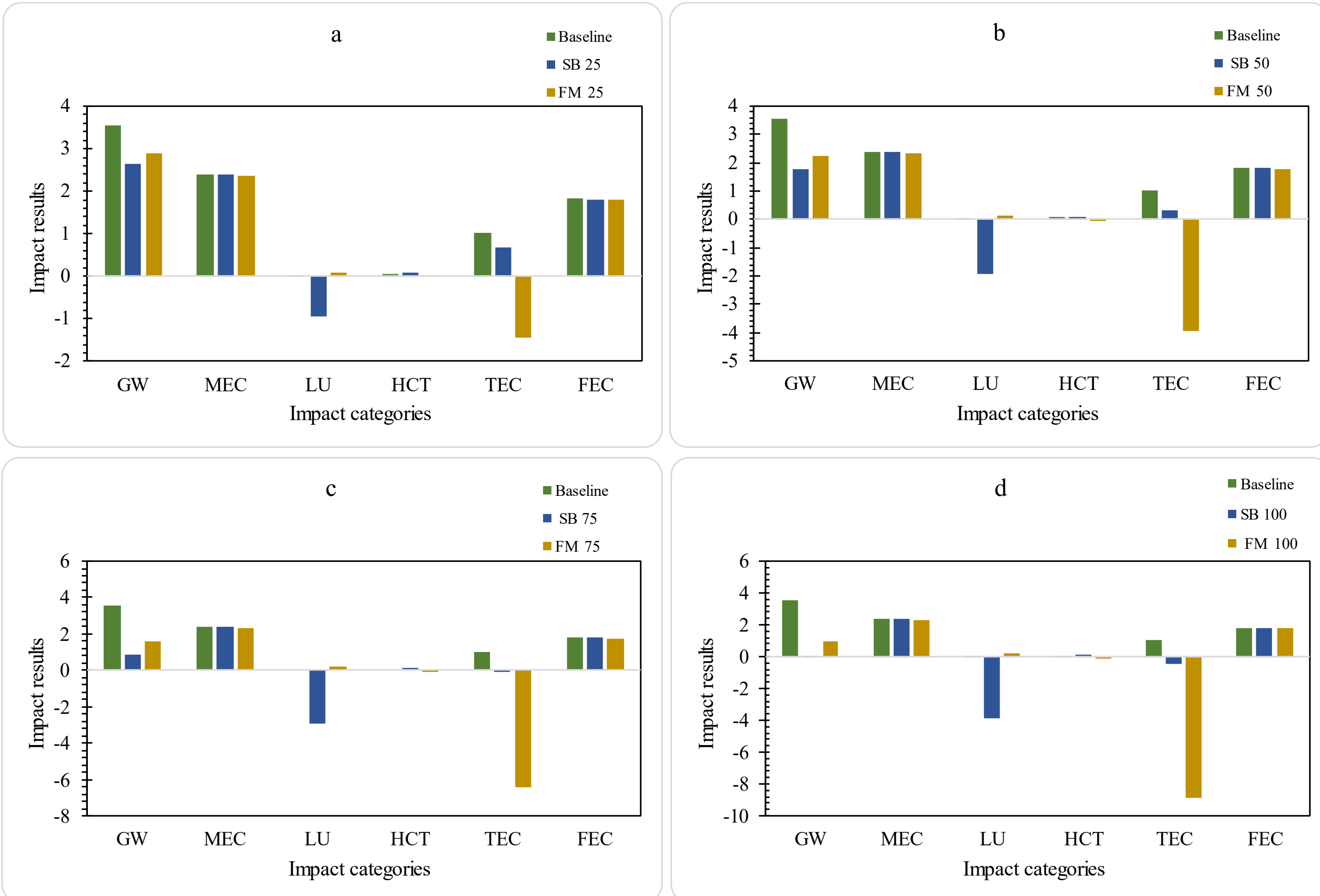

Figure 4.13: Impact variation between baseline system and consequential scenarios

4.4 Conclusions

Crude pea starch is emphasized as a valuable bioresource with vast potential for SCP production in sustainable pea processing regions. The sustainability disposition of its utilization in SCP production is established through the LCA, emphasizing significant impact offsets relative to conventional protein food and feed. The evidence suggests pea-starch SCP as a sustainable pathway for reinforcing the pea industry's engagement toward carbon neutrality and aligning protein food and feed supply with local and global sustainability targets. Particular attention is drawn to the relevance of adopting novel technological and transportation options, with energy-efficient centrifugation technologies, train or air freight transportation, and renewable energy innovations heightened among the recommendations for improving system performance. The impacts of utilizing pea-starch SCP as a protein-feed alternative is highlighted through the consequential LCA, which enabled substantial global warming and land use offsets for replacing soybean meal and fishmeal. Aside from the relevance of the consequential LCA in defining the orientation of pea-starch SCP as a climate-adaptative feed option, the study's findings demonstrate its key strength in enhancing true environmental impact accounting in micro and macro-level sustainability decisions. Although the current study has shown a clear-cut benefit of pea starch upcycling for SCP production, the analysis presented is limited by the dependence on secondary data. The uncertainties surrounding such limitations could be addressed by a progressive in-situ implementation and assessment of the proposed design scenarios at varying experimental scales. Overall, pea-starch SCP production is suggested as an adaptable technical solution for improving sustainable practices in the pea industry while augmenting local and global protein security. However, promoting such a trajectory would require restructuring the pea value chain to include pea starch upcycling, backed by enabling policies and a radical collaboration between governments, private sectors, industry, researchers, and other stakeholders of the pea value chain.

CONNECTING TEXT IV

The manuscript for Chapter five is under internal review for final revision before submission.

Chapter three focused on developing the circular bioeconomy accounting tool to enable sustainable circular bioeconomy practice in biosystems. Life cycle assessment was emphasized in the theoretical framework as a relevant component in the performance analysis. Thus, chapter four explored developing varying pea-starch single cell protein scenarios and ascertaining their environmental impact performance. This was to identify operational hotspots for further system optimizations and decisions. The LCA showcased three process scenarios, SCP Baseline, SCP NME (production without media enrichment), and SCP ONME (production where synthetic carbon source is substituted with organic sugarbeet molasses in inoculum production and no media enrichment is involved) as environmentally competitive. In the following chapter (Chapter five), other components of the C-BEAT are estimated for these process scenarios following the prescribed steps in Chapter 3, and their respective CBI is calculated to emphasize general trends in their bioeconomy performance. Also, the respective ranks of the scenarios are ascertained following the BWM-CoCoSo methodology described in Chapter three to identify their respective ranks relative to defined economic, environmental, and social indicators. Chapter five is the first validatory study that utilizes the C-BEAT framework in circular bioeconomy performance assessment.

5 CHAPTER FIVE: C-BEAT APPLICATION

Case Study: Application of the C-BEAT in Industrial Decision Modelling: A Case Study for Pea Starch-Based Single Cell Protein

Abstract

The integration of comprehensive decision and sustainability tools into circular bioeconomy is becoming evolvingly significant for driving sustainable practice. In this study, the authors leveraged the novel Circular Bioeconomy Accounting Tool (C-BEAT) as an adaptable and robust framework to select an optimal scenario for converting crude pea starch into valuable single cell protein (SCP). Also, the study seconds as a validation of the C-BEAT. Following described steps, three production scenarios were identified and modeled, assessed for their economic, environmental, and social performances, and further tested against the novel Circular Bioeconomy Index (CBI) and the BWM-CoCoSo multicriteria decision method. A CBI value of one (1) highlighted no substantial distinction in performance, placing all the scenarios in a common sustainability region. However, multicriteria decision analysis revealed an explicit distinction between the scenarios, identifying the production of pea starch SCP using organic nutrient substrates for inoculum production and without further media enrichment (SCP ONME) as a relatively sustainable option. The finding supports the global concession to minimize using synthetic chemicals in agricultural and agro-industrial practices, marking the potential of SCP ONME to improve the overall sustainability of the pea industry. Regarding CBI and BWM CoCoSo, the former is particularly useful in cases where indicator values have higher deviations. However, CBI portrays strength as an index for concise circular bioeconomy communication. The latter is presented as a comprehensive tool for detecting minute differences between biocircular scenarios and is recommended for use when decisions are expected to guide policies or strategies.

Keywords: Circular Bioeconomy Accounting, Stakeholder, Multicriteria Decision, Single Cell Protein, Crude Pea Starch

5.1 Background

This thesis reiterates circular bioeconomy as a turnkey pursuit for revolutionizing biosystems through a regenerative value recovery approach and progressing the age-zero waste concept (Saha & Mathew, 2022; Smol, 2022). This stance has advanced interest and is navigating a smooth but gradual transition of the circular bioeconomy concept into local and global sustainability and waste management strategies (Lin, 2020; Pereira da Silva et al., 2021). Through the evolution of concepts, we have understood an explicit distinction between sustainability and circular bioeconomy (Morseletto, 2023; Yadav et al., 2020). There is an articulate explication of how these concepts diverge in actual principles but could be manipulated to intercept at a break-even point (Mohanty et al., 2022). Drawing on these perspectives, several ideas have evolved to emphasize the need to spare the generic assumption that circular bioeconomy is entirely sustainable (Abad-Segura et al., 2020; Avellán et al., 2022). In the best of construction, such a conceited assumption would frown on the exclusiveness of the circular bioeconomy concept and deny global access to its benefits in building regenerative economies by invading systems with circular but unstainable innovations (Morseletto, 2023; Yadav et al., 2020). These presumptions have revamped the need to reconceptualize the circular bioeconomy framework in such a way that aligns with and progresses its actual intent in achieving global sustainability goals, which in turn, could enable the outstanding benefits such transformative concept hold (Robinson, 2022; Saha & Mathew, 2022). On this premise, a body of work is rapidly evolving to identify, integrate, and adopt theories and models to enhance conceptual robustness and augment its adaptability as a decision support system for sustainable circularity practice. One such study is that of Köhler et al. (2022), which informed the relevance of considering cross-sectional collaboration in systems for maximizing circular economy practices. The study addressed a critical but usually underemphasized concept of system scrutiny for ascertaining relevant networks and accentuated how such system interlinkages could be rightly deployed in promoting circular practices. Others have highlighted deficiencies of the circular bioeconomy framework in demonstrating the environmental, economic, and social performances of proposed solutions (Mannan & Al-Ghamdi, 2022; Medina-Salgado et al., 2022), recommending their integration as an essential compliment to the capacity of this vital concept. Regarding decision-making, Romero-Perdomo and González-Curbelo (2023) also identified a significant hitch. They concluded the integration of multicriteria decision analysis as a unique path for aligning circular bioeconomy propositions for micro, meso, and macro-scale adoption. The pool of these ideas coupled with the urgency of providing an actionable framework that unifies the diversity of exceptional thoughts drove the development of the circular bioeconomy accounting tool (C-BEAT). The tool consolidates stakeholder inclusion, environmental, economic, and social assessment, and multicriteria decision analysis into a followable procedure for identifying circular bioeconomy pathways and ascertain their sustainability relevance to aid research, industrial, and governmental decisions that align with defined goals. In this study, we take a validatory step through applying C-BEAT in a case-study, wherein all steps are followed for ascertaining the circularity potentials of various process scenarios designed for transforming underutilized industrial crude pea starch into single cell protein. The study will ultimately help in identifying the optimal scenario that meets current market needs and bears the capacity to support sustainable production.

5.2 Methodology

This section intricately describes the steps followed to achieve the objectives of this study. Herein, steps already described for the C-BEAT framework in chapter three were followed with slight assumptions where necessary. The following subsections describe the steps and clarify the assumptions made for this case-study. All equations for variable estimations can be found in chapter three.

5.2.1 Phase 1: Stakeholder Engagement

The stakeholder engagement stage is the prime step in the framework, which involves the selection of a team of experts to collaboratively identify pathways within the interest and goals of the focus company or system. In this case, stakeholders of the pea protein extraction industry must be considered. This phase also involves a market survey to collect ideas on the patronage potentials of the internally selected pathways and adopt consumer insights to design innovative products that align with profit-making goals. The limitations of this case-study in executing this phase in situ necessitated the assumption of single cell protein as a feasible and market-ready pathway taking the plethora of scholarly insights on its potential and market share, comprehensively captured in chapter two.

5.2.2 Phase 2: Pathway Description

This phase involves conceptualizing the validated pathway(s) considering the scope of operation or system boundary, potential scenarios, process assumptions, and characteristics of the reference product, amongst other relevant attributes that can inform variations in system performance. The case study understudies the baseline pea-starch SCP scenario in chapter four as the primary pathway. It considers two other process scenarios, SCP NME, and SCP ONME, for further analysis taking their competitive performances in the environmental impact assessment. The baseline SCP scenario involves the production of SCP from crude pea starch slurry generated on-site. The upstream process considers a water addition step to increase the moisture content of the slurry to 70 %, suitable for solid-state fermentation, microbial preculturing (inoculum production), which involves the on-site production of inoculum, a media enrichment step to augment the nutritional quality of the substrate and an aerobic fermentation step that allows microbes to multiply through the utilization of the nutrient-rich crude pea starch substrate. Following fermentation is the harvesting procedure that deploys centrifugal technologies to separate the microbial biomass from the other biomass components, which is then dried to a 10% moisture content, stored at 4°C, and transported to the uptake site. The SCP NME and SCP ONME scenarios followed the same steps, with the exception that there was no media enrichment for both scenarios and synthetic carbon was substituted for organic molasses in the microbial preculturing step for the SCP ONME scenario considering the rising abundance and continuous generation of sugar beet molasses in the Canadian sugar beet industry. The baseline and process scenarios were modeled to represent the ideal situation in Canada. Thus all physical flows were assumed to be sourced from Canada, except for flows that lacked local data, in which case they were represented with other regional and global data. The process flow was designed using the draw.io software, and physical flows and system boundaries are explicitly shown in Figure 4.1.

5.2.3 Phase 3: Simulation of Pathways

Based on the assessment, the C-BEAT framework recommends deploying trusted simulation software for modeling the various processes described. Following the steps described, the required assessments include life cycle assessment, life cycle costing (LCC), and social life cycle assessment (SLCA). For life cycle assessment, the processes were simulated in the OpenLCA software. This free LCA software provides a user-friendly interface and gives the luxury of efficiently performing environmental assessments. For LCC and SLCA, Microsoft Excel was used to design separate adaptable dashboards for the assessment (refer to supplementary materials: SM5.1 and 5.2).

5.2.4 Phase 4: Sustainability Assessment

5.2.4.1 Life Cycle Assessment (LCA)

The methodology description and actual assessment for the baseline and process scenarios have been done in chapter four. Summarily, LCA followed the ISO 2006: 14040/44 standards, considering the definition of the goal and scope of the assessment, life cycle inventory, life cycle impact assessment, and interpretation. It is, however, important to emphasize that the functional unit used for all assessments in the case study was estimated as per the total annual output from processing about 34,000 tons of pea-starch slurry, quantified to be about 4.08kt per annum.

5.2.4.2 Life Cycle Costing

The relevance of life cycle costing has been reiterated in the earlier chapters of this thesis. Also, costs considered for the assessment were generated by Peters et al. (2003) with slight modifications. The assessment considered total capital investment and total product costs. An annualized capital charge ratio of 11.7% was used to quantify the total annualized operating cost. Gross profit, net profit, and net present value (at a depreciation rate of 10%)

were estimated accordingly. A consummation of fixed capital investments, including direct and indirect costs, and the working capital amounted to the total capital investment. Total product costs included manufacturing and general costs like administrative, research and development, and distribution and marketing costs. In accounting for the cost associated with emissions from annual production, the cost of environmental emissions and disposal were included in the total capital investment calculation. Environmental emissions and disposal cost was assumed to be 1% of fixed capital investment. It is important to emphasize that environmental monetization is not necessarily synonymous with emission cost, which informs why monetized environmental impacts were not treated as the actual emission costs. Instead, it is the monetary equivalence of LCA results in this case study. A complete list of cost considerations for the LCC is outlined in Tables 5.1 and 5.2.

Table 5.1: Considerations for Total Capital Investment

Fixed Capital Investment (FCI)		
Direct cost	% of FCI	Normalized %
Purchased equipment	25	21.19
Equipment installation	12	10.17
Instrumentation and controls	11	9.32
Piping	15	12.71
Electrical systems	5	4.24
Service facilities	15	12.71
Yard improvements	2	1.69
Emissions and Disposal Cost	1	0.85
Indirect		
Engineering and supervision	10	8.47
Contractor’s fee	2	1.69
Legal expenses	2	1.69
Contingency	8	6.78
Construction expense	10	8.47
Working Capital	0.15 of Total Capital Investment	

Table 5.2: Considerations for Total Product Cost

Manufacturing Cost	
Variable Cost	Factor
Raw materials	Calculated
Utilities	Calculated
Operating labor	Calculated
Operating supervision	0.15 of operating labor
Maintenance and repairs (M& R)	0.07 of FCI
Laboratory charges	0.15 of operating labor
Operation supplies	0.15 OF M & R

Nutrients and solvents	Calculated
Royalties and Patents	0.01 of FCI
Fixed Cost	Factor
Depreciation	Calculated
Taxes	0.02 of FCI
Insurance	0.05 of FCI
Total Plant Overhead Cost	0.45 of operating labour + operating supervision
General Expenses	
Administrative cost	0.3 of operating labor
Research and Development	0.15 of Manufacturing cost
Distribution and Marketing	0.15 of Manufacturing cost

5.2.4.3 Social Impact Assessment

Due to existing difficulties in quantifying the social burdens attached to a given system, the C-BEAT took a novel approach to develop an easily measurable employment index to satisfy social performance circular bioeconomy accounting. Employment and labor values for the estimation were generated from recognized Canadian databases and the FAOSTAT. The concept and associated steps for estimating the employment index have been comprehensively described in Chapter 3, and an Excel sheet has been developed to ease estimation (attached as supplementary material, SM.5.1).

5.2.5 Circular Bioeconomy Index

The estimation of the unified circular bioeconomy index follows a four-stepwise procedure, including indicator monetization, indicator scaling, indicator weighting using the values provided in Table 3.1, and performance prediction using the CBI scale shown in Figure 3.4. Since the economic indicator, net profit, was already monetary, the monetization step involved the environmental and social indicators. Environmental impact monetization used the factors estimated in the Environmental Priority Strategies 2015d (EPS 2015d) monetization method, which deploys market values for monetary quantification contrary to other regionalized or localized monetization methods (Arendt et al., 2020; Steen, 2016). This validation results from this method as a representative of the global market case, allowing generalizability of values. For employment index monetization, we estimated the monetary gains attached to the number of employees for each scenario by calculating the unemployment cost reclamation using published data on the per labor cost of unemployment (USD 7742) (Pettinger, 2019). The step-by-step methodology for unemployment cost reclamation (Ucr) has also been described in chapter three.

5.2.6 Multicriteria Decision Analysis (MCDA)

Decision analysis has progressively advanced precision in decision-making at the micro, meso, and macro scales and continues to offer relevance in selecting optimal solutions for proposed ideas. In cases where multiple criteria are involved, the MCDA framework suffices competence and facilitates decisions that align with predefined maximization and minimization interests of stakeholders for strategic propositions. MCDA has dramatically evolved, wherein objective and subjective mathematical modeling has played exclusive roles in recent developments and enabled consistency and accuracy in existing models (Agyemang, Prince et al., 2022; Javaid et al., 2022; Soniya et al., 2022). Amongst the plethora of MCDA models, the C-BEAT, described in chapter three, found BWM-CoCoSo, a combined and structured framework for sustainable decisions that address the predominant consistency issues in multicriteria decision modeling (Ecer & Pamucar, 2020; Hashemkhani Zolfani et al., 2019), as a robust and reliable approach for biocircular decisions involving multiple criteria. Herein, we followed the methodology described in Chapter 3, Figures 3.5 and 3.6, without any modification. Three scenarios were formulated for the MCDA, as shown in Table 5.3, and details of the calculations are provided in the supplementary material (SM.5.3).

Table 5.3: Description of Scenarios for MCDA

Scenario	Description
1	Net profit and GWP were considered the best and worst criteria, respectively, with varying weights for all criteria
2	Employment Index and GWP were considered the best and worst criteria, respectively, with varying subjective weights for all criteria
3	Net profit and GWP were considered the best and worst criteria, respectively, with the same subjective weights assumed for all criteria

5.3 Results and Discussion

5.3.1 Environmental Impact Assessment

The environmental impact assessment singled out the global warming potential of the various scenarios for display in Figure 5.1, taking its prime consideration in the CBI calculation and multicriteria decision analysis. Like the trend discussed in chapter four, the baseline scenario demonstrated a high contribution to the global warming category, with an annual CO2 equivalence of 13 kt. On the other hand, SCP ONME showed the least global warming impact, offsetting about 15.41% of baseline impacts. It, however, compared closely with the

15.36% offset of the SCP NME scenario, highlighting a slight distinction in their performances. Furthermore, the baseline scenario portrayed a relatively high undesirability in achieving the net-zero carbon emission goal of the food system, presumably driven by the extra energy demand and greenhouse gas emissions attached to manufacturing synthetic nutrients for the media enrichment and microbial preculturing process. Thus, the SCP ONME scenario is zoned as a more sustainable preference to the other scenarios, especially for addressing the persisting climate emergency. By implication, the findings inform the need to engage substrate optimization strategies that can enhance their capacity to satisfy the nutritional needs of the microbes during the incubation period. Also, they highlight an opportunity to interlink pea-starch SCP production with other industries that produce second-generation carbon biomass, posing as a remediative strategy for promoting cross-system circular bioeconomy networks and collaborations to enhance practice (Köhler et al., 2022). In all these, it is important to reemphasize the urgency of adopting energy-efficient harvesting technologies to augment overall system performance.

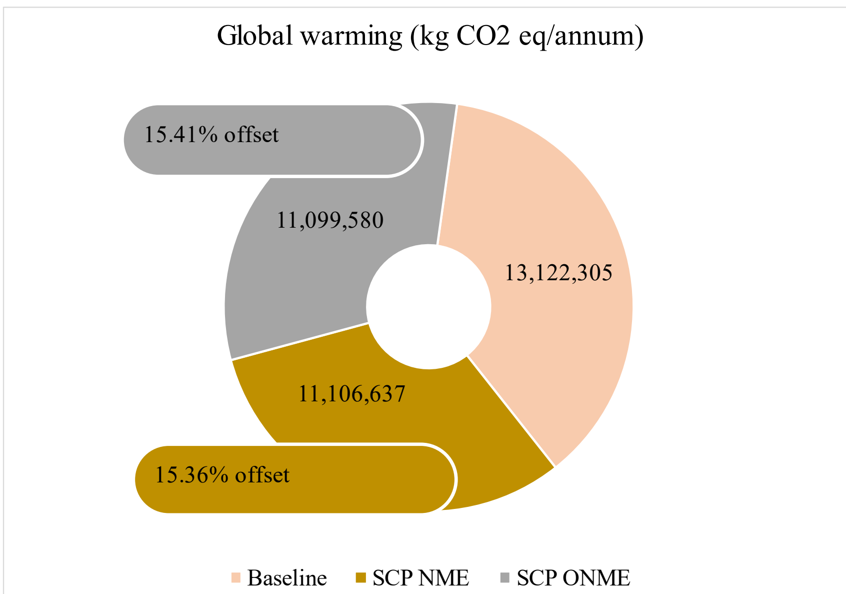

Figure 5.1: Annual Global Warming Impact for Selected Scenarios Per Annual Production Output

5.3.2 Life Cycle Costing

The economic evaluation resulted in a total annual operating cost (TAOC) of about $ 36,705,480 for the baseline scenario and about $35,855,925 and $35,855,916 for SCP NME and ONME scenarios, respectively, graphically shown in Figure 5.2. TAOC is the sum of annualized total capital investment and total product cost required for the annual operation of each scenario. To achieve an annualized total capital investment, a predefined annual capital cost ratio of 0.117 for an interest rate of 10 % and a project life of 20 years was applied to estimate the annual monetary equivalence of the total capital investment. Thus, the results corroborate the trend described for global warming, also showing the baseline and SCP ONME as the worst and best-performing scenarios, respectively. Again, SCP NME compared closely with SCP ONME. However, the slight monetary variation may bear economic prudence in the long term, which still places SCP ONME as the preference with better economic benefits. NPV was positive for all scenarios, demonstrating vitality in investing in the pea-starch SCP pathway. Nonetheless, again, SCP ONME ($77,415,664) had the highest NPV, while SCP NME ($77,415,611) closely matched the SCP ONME value. Likewise, SCP ONME demonstrated higher net profit ($11,513,335) at a unit selling price of $13, compared to slightly varying values of about $10,944,128 and $11,513,330 for the baseline and SCP NME scenarios, respectively. From these trends, it can be generally asserted that endeavoring to minimize the use of synthetic nutrients in pea-starch SCP production and optimizing inoculum production would offer substantial economic gains while promoting actions toward climate remediation.

An interesting part of the economic evaluation is creating a control scenario to ascertain the consequences of outsourcing inoculum for the production process. It is a widespread practice that due to time and resource demand, fermentation-based industries usually commonly outsource base microbes from other providers. The SCP Control scenario shown in Figure 5.2 demonstrated this scenario as economically draining, showing the lowest scores for net profits ($10,460,345) and NPV ($67,072,390) and the highest score for TAOC. However, a positive NPV infers economic prudence in investing in such a scenario. Such a decision could be justified with a calculated trade-off between time, labor, and resource demand.

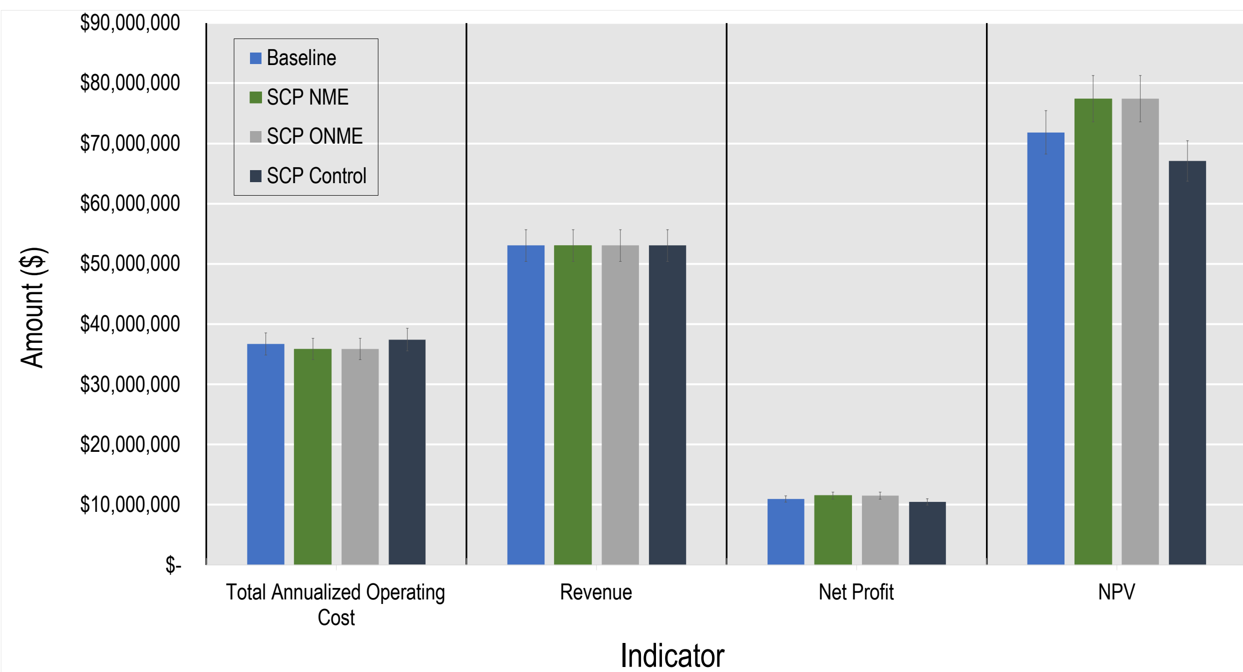

Figure 5.2: Life Cycle Costing Results for Selected Economic Indicators

5.3.3 Social Performance

The employment index and unemployment cost reclamation results for the various scenarios are presented in Figure 5.3. The employment index measures a system's potential to minimize unemployment within a given geographical region, respective of the labor force and population within such region. A higher employment index shows a high requirement for operating labor and a high propensity for reducing unemployment. On this basis, the baseline scenario (2.95×10^{-6}) offers a high employment rate compared to the SCP NME and ONME scenarios (2.76×10^{-6}) due to eliminating the media enrichment step in the latter scenarios. Whereas this portrays a subjective preference for the baseline scenario toward improving employment opportunities, such preference can only be validated by the overall goals and predilection of the focus industry and how they measure creating employment opportunities against profitability and environmental impact remission in their circular endeavors.

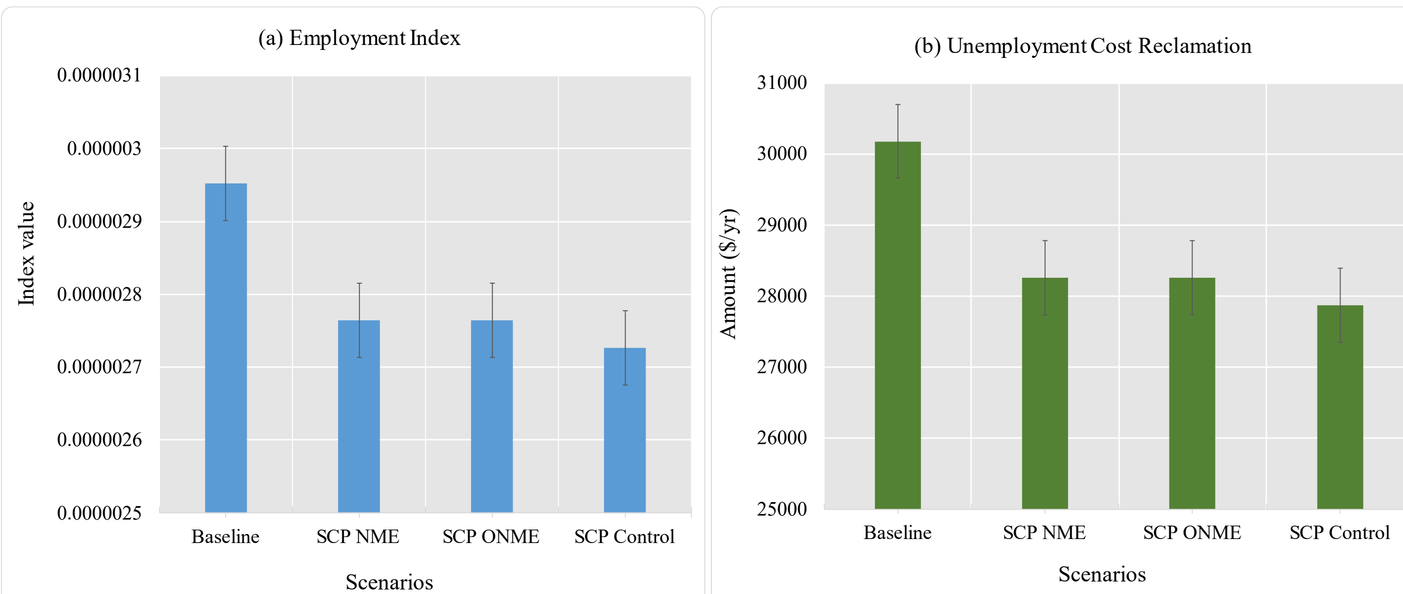

Figure 5.3: Comparison of the social performance of process scenarios

Unemployment cost reclamation denotes the monetary recovery for total operating labor resulting from the actual operation of the scenarios. It is government-oriented and stands to underline the economic savings in terms of 'avoided government expenditure' on unemployed labor. Like the employment index trend, the baseline system that demanded more operating labor enabled the reclamation of high unemployment cost, about $30,179 per annum, compared to $28,255 for SCP NME and ONME. This demonstrates its precedence in minimizing government spending on unemployed labor and reinstates how pursuing such a circular approach transcends benefits to the industrial economy to buffering the entire economy. Moreover, the findings reiterate the need to double efforts towards pushing for the integration of SCP into the pea protein extraction industry, considering the massive environmental and socioeconomic benefits that would enable. The SCP NME and ONME had proximal but substantially varied values compared to the baseline, implying considerable economic benefits concerning minimizing government expenditure on unemployment. However, a thorough analysis of the tradeoff between the scenarios taking the multiple criteria described would be a sound basis for justifying scenario preference.

5.3.4 CBI Estimation

As described earlier, C-BEAT deploys two performance assessment perspectives to enhance biocircular decisions. The first perspective, Circular Bioeconomy Index, which aggregates values from environmental, economic, and social impact assessment through a logical scaling and weighting procedure, is estimated for each scenario and presented in Table 5.4. The scaled values denote the percentage proximity of each indicator value to the maximum indicator value across the given scenarios. Also, the weighted values represent the numerical equivalence of the scaled values within the predefined weights of 0, 0.5, and 1 (refer to Chapter 3 for more details). All scaled values for the GWP indicator were above 80 for all scenarios, with corresponding weights of 1. This places all the scenarios within the same environmental sustainability region, marking a little distinction between the baseline's and proposed scenarios' environmental performances. A similar trend was found for the net profit and unemployment cost reclamation, wherein all scenarios were drawn within the same sustainability region. The CBI values show that all scenarios have an index of 1, which can be described following the CBI scale as within a sustainable region. However, the scale does not allow selective judgment, limiting the distinction of the various scenarios regarding their circular bioeconomy performance. In this case, multicriteria decision analysis would be preferable for ascertaining such distinctions for a more definitive judgment. The following section presents and discusses the results of the multicriteria decision analysis using the BWM-CoCoSo method.

Table 5.4: CBI Calculation for Process Scenarios

Indicator	**S1 Value**	**Scaled S1**	**Weighted S1**	**S2 Value**	**Scaled S2**	**Weighted S2**	**S3 Value**	**Scaled S3**	**Weighted S3**
$GWP	1,771,511	100	1	1,499,396	84.64	1	1,498,443	84.59	1
Net Profit	9,167,177	94.15	1	9,736,490	99.99	1	9,736,496	100	1
Unemployment Cost Reclamation	30,180	100	1	28,256	93.62	1	28,256	93.62	1
CBI Value	CBI_{S1} = 1+1-1 = 1			CBI_{S2} = 1+1-1 = 1			CBI_{S3} = 1+1-1 = 1		

5.3.5 Multicriteria Decision Analysis

5.3.5.1 Weight Estimation – BWM

The estimated criteria weights for the various MCDA scenarios using the BWM pairwise comparison method are shown in Table 5.5. The first scenario was formulated following the business-for-profit ideology, which positions businesses, including the agroindustry, as profit-making entities with secondary responsibilities of improving socio-economic status and minimizing environmental burdens. Thus, the weighting procedure was manipulated to quantify the highest weight of 0.67 and lowest weight of 0.08 for the net profit and GWP criteria, respectively, considering net profit as the best criterion and GWP as the worst or minimization criteria in the objective function. The second scenario assumed the employment index as the best criterion, intending to forecast the respective ranks of the SCP scenarios if social benefits (employment index) and GWP were considered the best and worst criterion, respectively. In addition, it sought to answer the question: What if organizations or governments prioritize social benefits over economic benefits while maintaining their aversion towards global warming? In this vein, the criteria weights were estimated as 0.36, 0.56, and 0.08 for Net Profit, Employment Index, and GWP, respectively. These criteria weights were used in the CoCoSo model to estimate the ranks of the various SCP scenarios and identify optimal scenarios.

Table 5.5: Criteria Weights for Various Scenarios

Scenarios	Criteria Weight		
	Net Profit	Employment Index	GWP
Scenario 1	0.67	0.25	0.08
Scenario 2	0.36	0.56	0.08
Scenario 3	0.33	0.33	0.33

5.3.5.2 Ranks for SCP Scenarios in varying MCDA Scenarios

The ranks for the SCP scenarios respective to MCDA assumptions are presented in Figures 5.4 and 5.5. The scenarios' ranks were estimated through complex appraisal algorithms (Ka, Kb, and Kc), which quantified an appraisal value and rank for each scenario. These algorithms employed the weighted sum (Si) and weighted product (Pi) values to model complex equations that cumulatively minimized inconsistencies and uncertainties in the final appraisal and ranking. Ki was modeled from the Ka, Kb, and Kc values to provide final appraisal outputs and ranks for the various scenarios- "one (1)" is the highest rank. Figures 5.4 and 5.5 show that SCP ONME had the highest appraisal value and rank across all the appraisal algorithms, signifying its preeminence over the SCP NME and the baseline. Similarly, it outweighed the other scenarios across the MCDA assumptions, confirming certainty in its position as the most preferred alternative for achieving optimal environmental, economic, and social benefits. While the position of SCP ONME was quite distinct, SCP NME and SCP Baseline played contrarily across the various appraisal algorithms and assumptions, making it challenging to assign a definite rank for the two. For instance, when net profit was assigned the highest weight and similar weights with the other criteria, SCP NME scored a higher rank than the baseline, ranking 2nd after the SCP ONME scenario. On the other hand, the SCP baseline ranked 2nd after the SCP ONME when employment index was assigned the highest weight. Therefore, while the overall trend directs attention towards SCP ONME, SCP NME, and SCP baseline as the decreasing order of circular bioeconomy preference, the relativities in ranks concerning change in best criterion reinstates the need to prioritize weighting and priority sets in multicriteria decision. It emphasizes a mutual dependence between scenario ranks and the preferences of the analyst or stakeholders, commending a comprehensive representation of stakeholder perspectives in criteria prioritization and weighting to ensure a fair and reliable appraisal. On the action side, the findings call for the need to support innovations toward identifying second-generation enrichment options or drastically reducing the use of synthetic nutrients in SCP production, taking its prospects of magnifying sustainability benefits. Furthermore, it supports the adoption of second-generation pea starch SCP as a frontline climate action and a reliable bioeconomy option for building economic wealth and improving socioeconomic well-being.

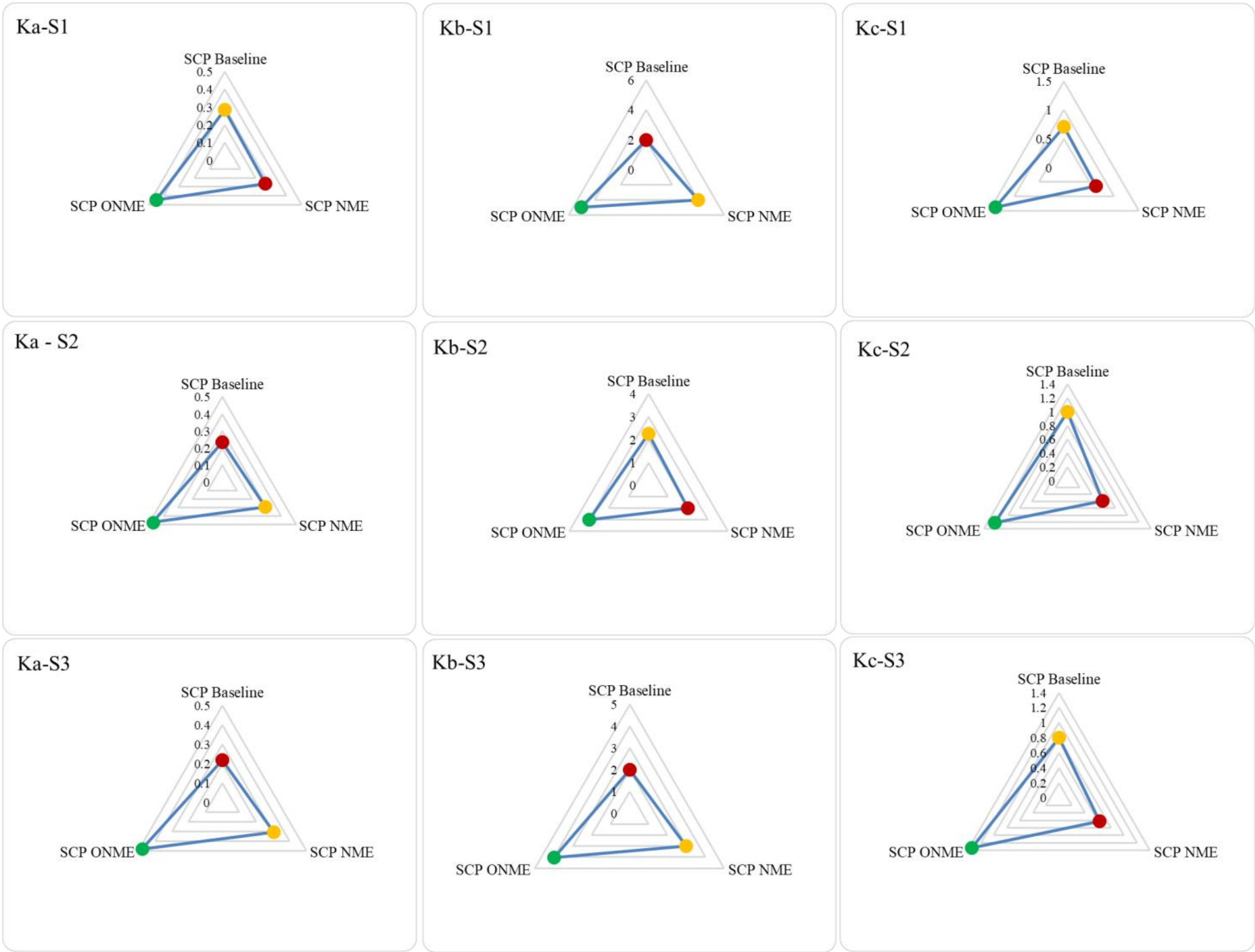

Figure 5.4: Appraisal values for various MCDA Scenarios

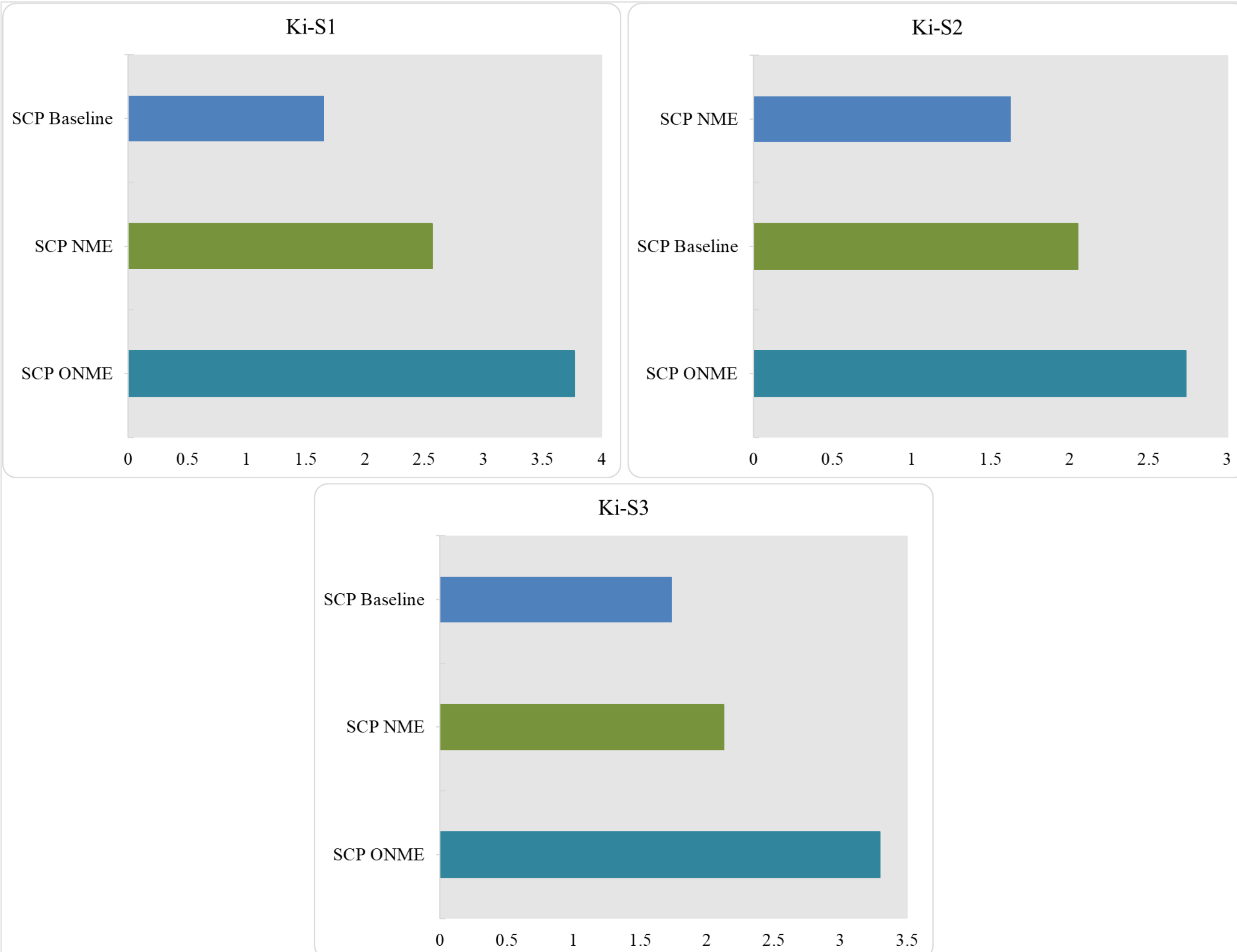

Figure 5.5: Final Appraisal Values (Ki) and ranks of SCP Scenarios

5.4 Future Perspectives and Conclusions

Throughout the study, circular bioeconomy has been emphasized as an actionable regenerative practice that should be adopted to improve sustainability in the pea industry through crude starch upcycling. The study has refocused the vitality of engaging comprehensive frameworks that leverage subjective and objective stakeholder perspectives and robust life cycle sustainability and decision analysis tools in advancing circular bioeconomy practice. The study also underlined the capacity of such frameworks in contextualizing biocircular decisions for sustainable deployment at all decision scales, marking the C-BEAT as an outstanding tool to spearhead circular bioeconomy accounting. In the deployment of the C-BEAT for assessing the performance of pea-starch SCP scenarios, the SCP ONME, which involved no media enrichment, and the substitution of synthetic carbon sources with second-generation carbon-rich feedstock (sugar beet molasses) for inoculum production, demonstrated high potentials across the accounting indicators, showing potentials in improving profitability, social benefits, and reducing the environmental repercussions associated with SCP production. However, the distinction between SCP ONME, SCP NME, and SCP baseline was quite comparable, generally asserting the pea starch SCP pathway as a recommendable action for improving sustainability in the pea industry. The next line of action is for governments, industries, and researchers to consolidate efforts toward advancing current technologies to a readiness level that would enable commercial production. This will demand monetary investment, policy formulations, and economic remodeling to successfully engulf the emerging sustainable economy trends, SCP production inclusive. Finally, whereas the CBEAT framework is reiterated as a robust methodology for circular bioeconomy accounting, it is vital to reemphasize some limitations to the accounting perspectives. First, the CBI method only measures the position of the scenarios in limited sustainability regions, constraining the distinction of scenarios in cases where indicator values are close to each other. However, it demonstrates strengths in enhancing circular communication using aggregated and easily interpretable values. The multicriteria decision analysis builds on the limitations of the CBI method, allowing the identification of slight distinctions based on criteria weighting and prioritization. Despite such strengths, it is exposed to the influence of stakeholder perspectives and subjective weightings, emphasizing the need to allow comprehensive stakeholder inputs to allow decision outputs respecting the core goals of the focus system and global sustainability.

6 CHAPTER SIX: GENERAL CONCLUSION AND RECOMMENDATIONS

The circular bioeconomy model is explicitly a promising trajectory for enhancing the sustainability performance of current economic pursuits while maximizing economic growth. Beyond sustainability benefits, it appears to be a reliable approach for transforming the extravagant amount of waste generated from agro-industrial processes into high-value, commercially viable products to support global food and nutritional needs. As the concept progresses, the C-BEAT demonstrates novelty as a timely input that will augment practice and place biocircular strategies within a frame that benefits the global environmental, economic, and social sustainability goals. Stakeholders are critical elements of every system, and their interests define the performance of most projects or pursuits. Thus, the consideration of multiple stakeholders, especially consumers, in the model stands to nudge a collaborative drive toward successfully pursuing circular bioeconomy in the agro-industry and ensuring commercial acceptance of evolving circular products. The SCP pathway showcases an opportunity to circularize crude pea starch for protein production, offering advantages in enhancing sustainability in the pea industry while increasing protein availability to satisfy growing demand. It shows substantial environmental benefits over conventional animal proteins like beef, informing its relative preference in cases where substituting beef with microbial protein is possible. Moreover, it portrays as a sustainable substitute for protein feeds like soybean meal and fishmeal, attached with significant land use and global warming offsets. Here, a viable opportunity is emphasized in exploring pea starch SCP as a sustainable alternative to conventional aquaculture feed, based on its advantages in decoupling protein production and utilization from overexploitation of forest land areas and the climate crisis. Regarding process optimization, adopting energy-efficient harvesting technologies and establishing the SCP plant in areas where renewable energy is dominant would be essential for achieving maximum sustainability performance. This subtly urges private and public efforts toward developing energy-efficient and smart technological innovations to minimize energy demand for SCP harvesting and the entire production process. Furthermore, it is advised from a sustainability purview that SCP upcycling is done at the site of co-product generation, considering the high environmental impacts attached to the transportation of high volumes of feedstocks contrary to the favorable impacts of distributing the final products instead. In terms of optimal designs, sole dependence on nutrient-dense second-generation feedstock without synthetic nutrient supplementation is identified as the ideal pathway for achieving optimal economic, social, and environmental benefits, which provides a clear starting point for further exploration. Subsequent actions should prioritize advancing the integration of pea-starch SCP into regional pea protein processing, which could frontline strategic actions toward augmenting value recovery in the pea industry. It would also play socioeconomic significance in redeeming additional revenue that would have been lost if co-products were discarded. Additionally, such a pursuit would create several job opportunities to balance regional labor force with employment rates. Therefore, governments, the private sector, industrial stakeholders, and researchers in regions where pea production and processing are prominent are encouraged to direct a common interest in pursuing this opportunity. Holding this as a strategic climate action will not only benefit improving the sustainability of the pea industry but will prolong the emerging drive toward designing regenerative economic models and boosting economic growth.

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APPENDICES

Appendix A: Percentage increase and offset in impact values for interpretational analysis

Table A.1: Impact offset or increase by baseline relative to process scenarios

Impact category	Baseline	SCP NME	SCP ONME	SCP OME
Fine particulate matter formation	Baseline	3.02	2.66	34.42
Fossil resource scarcity	Baseline	25.94	25.78	41.81
Freshwater ecotoxicity	Baseline	-1.34	-1.35	0.49
Freshwater eutrophication	Baseline	-20.52	-20.86	7.88
Global warming	Baseline	4.59	4.53	10.74
Human carcinogenic toxicity	Baseline	0.66	0.57	9.50
Human non-carcinogenic toxicity	Baseline	-0.70	-0.70	0.81
Ionizing radiation	Baseline	-14.97	-15.79	25.93
Land use	Baseline	-33.28	-37.99	33.64
Marine ecotoxicity	Baseline	-1.17	-1.18	0.65
Marine eutrophication	Baseline	-8.12	-11.27	1.22
Mineral resource scarcity	Baseline	-229.68	-231.44	8.95
Ozone formation, Human health	Baseline	37.56	37.48	45.79
Ozone formation, Terrestrial ecosystems	Baseline	37.78	37.71	46.00
Stratospheric ozone depletion	Baseline	3.44	3.18	5.61
Terrestrial acidification	Baseline	-3.59	-4.23	32.77
Terrestrial ecotoxicity	Baseline	67.98	67.96	71.23
Water consumption	Baseline	-23.56	-23.73	1.77

Key: Positive values represent percentage offset and negative values represent percentage increase

Table A.2: Impact offset or increase by baseline relative to transportation scenarios

Impact category	Baseline	TS1A	TS1B	TS1C	TS2A	TS2B	TS2C
Fine particulate matter formation	Baseline	85.44	98.36	92.64	34.58	27.37	64.22
Fossil resource scarcity	Baseline	92.87	84.36	96.12	41.91	18.96	76.21
Freshwater ecotoxicity	Baseline	10.93	10.56	2.74	0.50	0.31	0.79
Freshwater eutrophication	Baseline	66.14	87.77	40.43	8.07	9.01	13.53
Global warming	Baseline	69.48	58.16	81.59	10.79	4.70	36.11
Human carcinogenic toxicity	Baseline	69.67	73.09	36.78	9.58	16.86	14.20
Human non-carcinogenic toxicity	Baseline	15.46	15.61	10.01	0.80	0.20	1.89
Ionizing radiation	Baseline	85.65	68.60	87.65	26.27	23.48	51.66
Land use	Baseline	86.10	72.38	43.09	34.75	13.48	37.93
Marine ecotoxicity	Baseline	12.75	11.13	3.61	0.66	0.32	1.04
Marine eutrophication	Baseline	44.55	68.13	26.50	3.71	4.01	6.98
Mineral resource scarcity	Baseline	71.19	50.48	37.87	9.08	8.91	13.93
Ozone formation, Human health	Baseline	81.11	87.62	96.31	45.84	38.86	77.63
Ozone formation, Terrestrial ecosystems	Baseline	81.71	87.55	96.31	46.06	38.90	77.65
Stratospheric ozone depletion	Baseline	62.52	20.50	54.91	5.86	2.57	15.59
Terrestrial acidification	Baseline	83.93	97.43	93.99	33.03	25.83	67.35
Terrestrial ecotoxicity	Baseline	97.01	70.46	90.95	71.25	0.96	77.76
Water consumption	Baseline	26.57	14.01	14.84	1.88	1.72	3.54

Key: Positive values represent percentage offset and negative values represent percentage increase

Table A.3: Impact offset or increase by baseline relative to electricity scenarios

Impact category	Baseline	ES1 (%)	ES2 (%)	ES3 (%)	ES4 (%)
Fine particulate matter formation	Baseline	25.52	41.27	21.62	64.74
Fossil resource scarcity	Baseline	19.03	34.47	25.45	54.24
Freshwater ecotoxicity	Baseline	0.20	0.45	0.48	0.93
Freshwater eutrophication	Baseline	21.64	37.63	41.20	56.17
Global warming	Baseline	3.88	10.03	5.99	19.69
Human carcinogenic toxicity	Baseline	3.21	6.73	6.22	14.41
Human non-carcinogenic toxicity	Baseline	0.30	0.68	0.72	1.43
Ionizing radiation	Baseline	23.03	75.12	85.87	40.12
Land use	Baseline	8.34	11.81	11.94	17.99
Marine ecotoxicity	Baseline	0.21	0.48	0.51	0.99
Marine eutrophication	Baseline	7.43	16.60	18.96	26.69
Mineral resource scarcity	Baseline	0.39	1.67	2.43	1.46
Ozone formation, Human health	Baseline	10.94	23.74	13.67	41.01
Ozone formation, Terrestrial ecosystems	Baseline	10.93	23.69	13.66	40.95
Stratospheric ozone depletion	Baseline	1.35	3.88	2.53	5.06
Terrestrial acidification	Baseline	14.34	32.64	24.61	49.83
Terrestrial ecotoxicity	Baseline	3.09	8.11	5.81	15.32
Water consumption	Baseline	-17.01	-69.35	-62.43	-75.97

Key: Positive values represent percentage offset and negative values represent percentage increase

Table A.4: Impact values of baseline and benchmark scenarios per kg of product

Impact category	Reference Unit	Baseline	Beef	Chicken	Fishmeal	Soybean Meal
Fine particulate matter formation	kg PM2.5 eq	0.0016	0.0135	0.0071	0.0032	0.0016
Fossil resource scarcity	kg oil eq	0.1349	0.3633	0.3493	0.2611	0.1349
Freshwater ecotoxicity	kg 1,4-DCB	1.1870	0.2420	0.0578	0.0242	1.1870
Freshwater eutrophication	kg P eq	0.0005	0.0018	0.0007	0.0002	0.0005
Global warming	kg CO2 eq	3.2163	19.2889	2.4241	1.0410	3.2163
Human carcinogenic toxicity	kg 1,4-DCB	0.2224	0.2262	0.0895	0.0775	0.2224
Human non-carcinogenic toxicity	kg 1,4-DCB	19.8183	1734.4972	0.7961	0.6757	19.8183
Ionizing radiation	kBq Co-60 eq	0.0337	0.0418	0.0717	0.0147	0.0337
Land use	m2a crop eq	0.0118	38.5408	1.8564	0.0408	0.0118
Marine ecotoxicity	kg 1,4-DCB	1.5378	0.2345	0.0654	0.0561	1.5378
Marine eutrophication	kg N eq	0.0001	0.0301	0.0049	0.0000	0.0001
Mineral resource scarcity	kg Cu eq	0.0027	0.0125	0.0052	0.0017	0.0027
Ozone formation, Human health	kg NOx eq	0.0021	0.0087	0.0059	0.0083	0.0021
Ozone formation, Terrestrial ecosystems	kg NOx eq	0.0021	0.0090	0.0060	0.0084	0.0021
Stratospheric ozone depletion	kg CFC11 eq	0.0000	0.0002	0.0000	0.0000	0.0000
Terrestrial acidification	kg SO2 eq	0.0018	0.0822	0.0403	0.0096	0.0018
Terrestrial ecotoxicity	kg 1,4-DCB	1.6589	6.3360	4.1930	2.6476	1.6589
Water consumption	m3	0.3692	0.0619	0.1780	0.0100	0.3692

Appendix B: Consequential LCA Results

Table B.1: Impact offset or increase by soybean meal substitution scenarios relative to baseline

Impact category	Baseline	SB 25	SB 50	SB 75	SB 100
Fine particulate matter formation	Baseline	62.11	124.21	186.32	248.43
Fossil resource scarcity	Baseline	27.91	55.83	83.74	111.66
Freshwater ecotoxicity	Baseline	0.40	0.81	1.21	1.62
Freshwater eutrophication	Baseline	412.71	825.41	1238.12	1650.82
Global warming	Baseline	24.95	49.90	74.85	99.80
Human carcinogenic toxicity	Baseline	-20.18	-40.36	-60.54	-80.73
Human non-carcinogenic toxicity	Baseline	-0.31	-0.63	-0.94	-1.26
Ionizing radiation	Baseline	20.33	40.67	61.00	81.33
Land use	Baseline	3550.44	7100.88	10651.32	14201.76
Marine ecotoxicity	Baseline	0.29	0.57	0.86	1.15
Marine eutrophication	Baseline	17.33	34.65	51.98	69.30
Mineral resource scarcity	Baseline	129.60	259.20	388.81	518.41
Ozone formation, Human health	Baseline	38.91	77.82	116.73	155.64
Ozone formation, Terrestrial ecosystems	Baseline	40.88	81.75	122.63	163.51
Stratospheric ozone depletion	Baseline	472.93	945.87	1418.80	1891.73
Terrestrial acidification	Baseline	28.88	57.76	86.64	115.52
Terrestrial ecotoxicity	Baseline	35.14	70.28	105.42	140.55
Water consumption	Baseline	37.68	75.35	113.03	150.70

Key: Positive values represent percentage offset and negative values represent percentage increase

Table B.2: Impact offset or increase by fishmeal substitution scenarios relative to baseline

Impact category	Baseline	FM 25	FM 50	FM 75	FM 100
Fine particulate matter formation	Baseline	280.39	560.78	841.17	1121.56
Fossil resource scarcity	Baseline	102.53	205.06	307.59	410.12
Freshwater ecotoxicity	Baseline	0.93	1.86	2.79	3.72
Freshwater eutrophication	Baseline	372.74	745.48	1118.23	1490.97
Global warming	Baseline	18.29	36.58	54.87	73.16
Human carcinogenic toxicity	Baseline	70.44	140.88	211.32	281.76
Human non-carcinogenic toxicity	Baseline	1.12	2.24	3.35	4.47
Ionizing radiation	Baseline	-119.93	-239.87	-359.80	-479.73
Land use	Baseline	-195.47	-390.94	-586.41	-781.88
Marine ecotoxicity	Baseline	1.26	2.53	3.79	5.06
Marine eutrophication	Baseline	-14.38	-28.76	-43.14	-57.52
Mineral resource scarcity	Baseline	56.53	113.06	169.60	226.13
Ozone formation, Human health	Baseline	211.43	422.86	634.29	845.72
Ozone formation, Terrestrial ecosystems	Baseline	206.63	413.26	619.89	826.52
Stratospheric ozone depletion	Baseline	-45.34	-90.68	-136.03	-181.37
Terrestrial acidification	Baseline	275.39	550.78	826.16	1101.55
Terrestrial ecotoxicity	Baseline	239.44	478.89	718.33	957.77
Water consumption	Baseline	73.21	84.53	89.13	91.62

Key: Positive values represent percentage offset and negative values represent percentage increase