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AQUATIC BIOLOGICAL RESOURCE PROCESSING

Summary of the Doctoral Thesis

RIGA TECHNICAL UNIVERSITY
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Institute of Energy Systems and Environment

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**AQUATIC BIOLOGICAL RESOURCE
PROCESSING**

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I hereby declare that the Doctoral Thesis submitted for the review to Riga Technical University for the promotion to the scientific degree of Doctor of Science (Ph. D.) is my own. I confirm that this Doctoral Thesis had not been submitted to any other university for the promotion to a scientific degree.

Kaspars Ivanovs (signature)

Date:

The Doctoral Thesis has been written in English. It consists of Introduction, 4 chapters, Conclusions 20 figures, 22 tables; the total number of pages is 215, including appendices. The Bibliography contains 340 titles.

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INTRODUCTION

We are being compelled by the climatic backdrop to reconsider our production and consuming practises to reduce global environmental harms – rise of global temperature, dwindling biodiversity, scarcity of resources. As consumerism and human needs are growing it is necessary to increase the variety of resources, enhance and modify resource processing methods, and guarantee product availability. Bioeconomy is important to Europe worth about 2.3 trillion euros annually, employs over 18 million people, critical for the environment, food production, and development of rural areas. When the bioeconomy sector and data are sufficiently integrated, it will have a significant impact on the sustainability performance and competitiveness of the bioproducts industry through the processing and analysis of production and other data, enabling accurate and specialised manufacture [1]. According to the Organisation for Economic Co-operation and Development, oceans contribute \$1.5 trillion annually in value-added to global economy and this number could reach \$3 trillion by 2030 [2]. Due to the development of society, there is a need to solve problems related to the more efficient use of fishing and aquaculture resources.

Traditionally, catch and aquaculture have been used for food and residues for soil improvement and animal feed. For some time now, a concept bioeconomy (blue bioeconomy in the context of aquatic resources) has played important role. The concept is widely used in scientific research and is used as a policy framework to include the bioresources part of the circular economy. Sustainable utilization of residues play an important role and could also be feedstock for renewable energy production. Research topic is clearly related to the broader field of renewable energy and sustainable development, as aquatic biomass processing provides a potential source of sustainable energy and materials that could reduce reliance on finite fossil fuel resources.

Topicality

Aquatic biomass processing research contributes to the development of efficient harvesting methods and processing of organic materials. Additionally, research in this field improves our understanding about ecology of aquatic biomass and the potential for sustainable development of resources. Marine environment management, technology, and product development are important in the Baltic Sea region in the blue bioeconomy and effective emission reduction in the European Green Deal policy. A multidisciplinary approach and interdisciplinary research at all levels will facilitate progress in the unattainable direction and will ensure science-based decision-making in research and policy, meeting emission targets, and socio-economic well-being. In context of republic of Latvia, bioeconomy strategy, framework documents, and activity monitoring are important for development, monitoring of the bioeconomy sector is carried out by the smart specialization strategy "Knowledge-intensive bioeconomy".

Over the last two decades, the fisheries and aquaculture sectors have been increasingly recognized for their essential contribution to global food security and nutrition. Expanding this role requires scaling up transformative changes in policy, management, innovation, and investment to achieve sustainable, inclusive, and equitable global fisheries and aquaculture. It is necessary to stimulate the application of biorefinery principles in the processing industry.

The monitoring results of smart specialization show that it is necessary to develop innovative processing technologies and create products, since the application of innovative solutions in the processing industry has a low added value.

Innovative and thoughtful processing technologies with suitable feedstock create added value, diversify the local economy, and contribute to the development of climate-neutral technologies, employment, education, and social welfare. Therefore, it is a very important task for researchers to create good preconditions by analysing the most important components and modelling processing systems, developing the processing industry as a whole and improving its efficiency and potential added value.

Objective of the research

The aim of the Thesis was to research aquatic bioresources and green processing of biomass resources into value-added products to find the best use of aquatic origin feedstocks and to support the transition to a more sustainable circular economy by leveraging renewable water resources. Based on scientific literature research and experiments, the Thesis outlines aquatic bioresources and generally used processing methods, as well as technique for getting products to better the long-term use of Latvia's aquatic bioresources in a technological sense and in the context of decision-making.

The following tasks have been set to achieve the goal:

1. Evaluate local aquatic bioresources as feedstock for value-added bioproducts – economically low-value fishery by-products and other biomass such as macroalgae, and reed, and describe the main bioproducts from aquatic residue.
2. Research literature for sustainable aquatic biomass processing technologies pre-treatment, green extraction methods, and remaining waste treatment method.
3. Describe biorefinery stages and essential components to manage aquatic biomass residue issue using it as feedstock. Recommend processing of three blue feedstocks – fish residue, macroalgae, and common reed.
4. Based on conducted research and literature analysis recommend further research direction in aquatic bioresource management in Latvia.

Theoretical and methodological basis

Literature analysis, experiments in the laboratory, data analysis, and technology description analysis were used in the development of the Thesis. Analysis of broad scope of scientific literature was performed and was the main source of information. In-depth review of literature was performed to assess methodologies for blue-biomass transformation routes. In RTU Biosystems Laboratory, research was conducted where selected resources – round goby, macroalgae, and reed, were studied for processing into bioproducts. Substrates were experimentally converted into oil, protein, biogas, green extracts, and building materials by using a variety of methods, such as chemical and green extraction, anaerobic digestion, and solar energy.

Research experiments and technology analysis are the two main parts of the Thesis and tackle the issue of managing aquatic biomass residue.

Main scientific novelties

There are three main novelties of this thesis, and they are mostly related to use of local aquatic biomass. The use of invasive fish species in the extraction of value-added products was studied. The processing of several aquatic bioresources in one functional unit from pre-processing of the material to disposal of the residues in an environmentally friendly way were researched and analysed. A feasibility study and feasibility analysis of a low-temperature biogas and solar hybrid system on a small scale was performed, the need for the system, socially integrative aspects, scale, opportunities for technology diffusion and integration in the overall renewable energy resource system were examined.

Practical contribution

The research on fish waste has evaluated the round goby biomethane potential for use as a feedstock in the production of biomethane, waste protein utilization has also been proposed. The Thesis research studies have contributed to the EU Blue Growth strategy concept, and smart specialization of bioeconomy. The solutions suggested in the Thesis may be used to design policies and strategies, as well as for designing an aquatic pilot biorefinery. Residual secondary biowaste treatment approach using small-scale low-temperature anaerobic digester has also been reviewed.

Structure of the Thesis

The Thesis is based on a set of 7 publications and focuses on the more complete use of water bioresources, finding applications for different feedstocks based on the analysis of individual bioresources. The research is based on the analysis of international and local scientific literature on aquatic bioresources, innovative processing methods, obtainable products, as well as related concepts of knowledge-intensive bioeconomy in the context of blue bioeconomy. In the practical part, the biomass composition analysis and biomethane potential tests were carried out, a feasibility study was carried out for small-scale processing with a plug-flow digester with solar heating. At the end of the Thesis, the suitability of the biorefinery concept for blue-feedstock is discussed and the author's recommendations for research directions that could be developed are given. Aquatic bioeconomy research was divided into several phases. The classic bioeconomy approach – resource-technology-product analysis, was used in overall research to provide results and rationalize discourse. The Thesis structure defines how to carry out the research analytically, methodologically, and philosophically (Fig. 1).

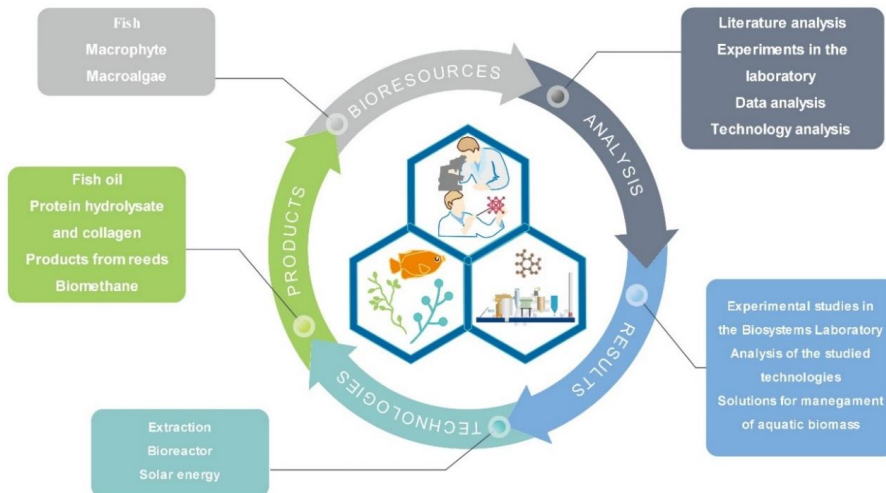


Fig. 1. External layout of the Thesis.

Approbation of the research

The Doctoral Thesis was developed as a thematically unified set of publications and is based on the following studies.

Publications included in the Thesis:

1. **Ivanovs, K.**, Spalviņš, K., Blumberga, D. Approach for Modelling Anaerobic Digestion Processes of Fish Waste. *Energy Procedia*, 2018, Vol. 147, pp. 390–396. ISSN 1876-6102.
2. Gruduls, A., Bāliņa, K., **Ivanovs, K.**, Romagnoli, F. Low Temperature BMP Tests Using Fish Waste from Invasive Round Goby of the Baltic Sea. *Agronomy Research*, 2018, 16 (2), pp. 398–409.
3. **Ivanovs, K.**, Blumberga, D. Extraction of Fish Oil Using Green Extraction Methods: a Short Review. *Energy Procedia*, 2017, Vol. 128, pp. 477–483. Available:
4. Melvere, M., **Ivanovs, K.**, Pubule, J., Blumberga, D. Use of Round Goby (*Neogobius Melanostomus*) Processing Waste in Bioeconomy. *Energy Procedia*, 2017, Vol. 128, pp. 484–490. ISSN 1876-6102.
5. Bāliņa, K., **Ivanovs, K.**, Romagnoli, F., Blumberga, D. Comprehensive Literature Review on Valuable Compounds and Extraction Technologies: The Eastern Baltic Sea Seaweeds. *Environmental and Climate Technologies*, 2020, Vol. 24, No. 2, pp. 178–195. ISSN 1691-5208. e-ISSN 2255-8837.
6. Muižniece, I., Kazulis, V., Žihare, L., Lupkina, L., **Ivanovs, K.**, Blumberga, D. Evaluation of Reed Biomass Use for Manufacturing Products, Taking into Account Environmental Protection Requirements. *Agronomy Research*, 2018, Vol. 6, Special Iss. 1, pp. 1124–1132. ISSN 1406-894X.
7. **Ivanovs, K.**, Blumberga, D. 2023. Plug flow digester with assisted solar heat: feasibility of small-scale system. *Agronomy Research*, 21(X), xxx–ccc.

Other publications of the author:

8. **Ivanovs, K.** Pike Esox Lucius Distribution and Feeding Comparisons in Natural and Historically Channelized River Sections. *Environmental and Climate Technologies*, 2016, 18, pp. 33–41. ISSN 1691-5208. e-ISSN 2255-8837.
9. Priedniece, V., Spalviņš, K., **Ivanovs, K.**, Pubule, J., Blumberga, D. Bioproducts from Potatoes. A Review. *Environmental and Climate Technologies*, 2017, 21, pp. 18–27. e-ISSN 2255-8837.
10. Spalviņš, K., **Ivanovs, K.**, Blumberga, D. Single Cell Protein Production from Waste Biomass: Review of Various Agricultural By-products. *Agronomy Research*, 2018, Vol. 16, Special Iss. 2, pp. 1493–1508. ISSN 1406-894X.
11. Petrauskaite, E., Vaiskunaite, R., Blumberga, D., **Ivanovs, K.** Experimental Study of Droplet Biofilter Packed with Green Sphagnum to Clean Air from Volatile Organic Compounds. 2017. Pp. 373–378. ISSN 1876-6102.

Participation in scientific conferences:

1. **Ivanovs, K.**, Blumberga, D. Extraction of Fish Oil Using Green Extraction Methods: a Short Review. The Conference of Environmental and Climate Technologies CONECT, 2017, October 12–14, 2017, Riga, Latvia.
2. **Ivanovs, K.**, Spalviņš, K., Blumberga, D. Approach for Modelling Anaerobic Digestion Processes of Fish Waste. The Conference of Environmental and Climate Technologies CONECT 2018, Vol. 147, pp. 390–396. ISSN 1876-6102.
3. Bāliņa, K., **Ivanovs, K.**, Romagnoli, F., Blumberga, D. Comprehensive Literature Review on Valuable Compounds and Extraction Technologies: The Eastern Baltic Sea Seaweeds. The Conference of Environmental and Climate Technologies CONECT 2020, May 13–15, 2020, online.
4. **Ivanovs, K.**, Blumberga, D. Plug flow digester with assisted solar heat- feasibility of small-scale system. 13th International Conference “Biosystems Engineering 2023”, May 10–12, 2023, Tartu, Estonia.

Supervised Bachelor and Master’s Thesis

1. Maira Melvere. Use of Round goby processing residues in the bioeconomy. Master Thesis. RTU, 2017 (in Latvian).
2. Erika Petrauskaite. Analysis and assessment of droplet biofilter packed with sphagnum load. RTU, 2017 (in English).
3. Laura Graudumniece. Utilization of fish processing residues containing connective tissue protein for collagen extraction. RTU, 2019 (in Latvian).

1. LITERATURE REVIEW

1.1. Aquatic bioresources and biomass processing

Aquatic biological resources are a set of organisms (hydrobionts) living in water whose life is not possible, permanently or at certain stages of development, without remaining in water. Aquatic resource management refers to the management and conservation of the aquatic resource base in the context of aquaculture, the concentration and capture of wild fish, and the search and harvest of other aquatic resources such as crabs, shrimps, snails, insects, aquatic plants, and seaweed [3]. Intensification of natural resource management has created interest in biomass substrates that have not been widely used in the national economy, and promoted research about macroalgae [4] and macrophytes [5], identification of resources and their possible application in the bioeconomy, for example to produce high added value products, or for use in the energy sector as fuel. Processing of fish and shellfish into food and other value-added products involves many sequential operations, and the main processing stages are primary processing, food processing, and preparation, pre-treatment of by-products, and extraction of value-added products. In the food industry, fish, shellfish, and edible algae are referred to as seafood and in non-food food industries as by-products, discards, residue, waste, surplus, biomass, excess, etc. The priority is always to use freshwater or marine biomass in food production first, then – for non-food production of feed, materials, and energy. Main task of the industry is to meet the demand for seafood products, ensuring their safety and quality. Fish processing involves preparing fish and seafood for delivery to consumers. Seafood can be preserved in several ways by curing – drying, salting, smoking, pickling, marinating, or combinations of these methods.

Fish used for non-food purposes is chilled or frozen before further processing, providing fresh feedstock. Feedstock composition depends on species, processing method, type of product; bycatch is also used as feedstock [6]. Monitoring the quality of processing species is essential for products to reach consumers, and high traceability also helps to ensure quality. However, if the product is damaged or discards have nowhere to be used, it is possible to use it to create added-value products by applying different processing technologies. Seafood products have a high nutritional value regarding protein, lipids, and essential micronutrients. Proximate composition of shellfish and finfish are provided in European and international databases [7]–[9]. Seaweed in a global sense is a new branch in seafood sector. In many parts of the world seaweed is used as food source because it is distributed in diverse environments. Since ancient times until the beginning of the 19th century people in the East regarded seaweed as a food of great delicacy [10]. Reed is used in eco-buildings, production of extracts and feedstock in fermentation. In hierarchy for aquatic food recovery the priority is to maximize edible yield, and the least preferred lowest value is given to incineration or landfilling [11].

Waste, discards, and residue from aquatic resources are produced throughout the fishing and processing phases. Sustainable utilisation of waste has improved recently. Non-edible components of finfish processing account for 10–50% of the overall weight and comprise the head, viscera, skin, bone, and flesh that is still attached to the bone. Non-edible components of shellfish, particularly those of crustaceans, such as the head, shell, viscera, and appendages, can make up to 85% of the raw material. A significant portion of these by-products are

underutilised, wasted, or discarded. Like any feedstock, biomass has its own specifics – location, seasonality, species (diversity and adaptations of biological organisms to different environmental conditions determine heterogeneous and complex composition), microhabitat conditions, harvest and storage conditions, relatively low energy density, and ambiguity of the market (demand, price, suppliers, distributors). Therefore, ability to measure biomass properties consistently and accurately is critical when planning the processing operations. The biomass studied in the Thesis falls into two classification groups – aquatic biomass (fish, seaweed) and herbaceous biomass (reed). The most important parameters determining the production process are renewable end-product required, quality and quantity of biomass, and the cost of the process [12]. Fish, shellfish, and macrophyte in wet weight all show similar water content from 60 to 80% and seaweed – 80 to 90%. This means that reduction of moisture content is an indispensable part of aquatic bioresource processing.

Biomass can be converted into two main types of energy carriers – electrical/heat energy and transportation fuels. Physicochemical characteristics that play a crucial role in directing the available feedstock into both or either of these domains are moisture content, caloric value, proportions of fixed carbon and volatile substances, ash content, alkali metal content, and cellulose/lignin ratio. Common processes involved in biomass conversion into energy are thermochemical conversions, biochemical conversions, and physicochemical conversions. The main pre-treatment methods of lignocellulosic biomass are: mechanical – milling, ultrasonic [13], [14]; chemical – liquid hot water, acid hydrolysis, alkaline hydrolysis, organosolv, oxidative, ionic liquids [15], [16]; chemical/mechanical – steam explosion, ammonia fibre expansion, CO₂, mechanical alkaline pre-treatment [14], [17]; and biological – biological hydrolysis [12]. After biomass pre-treatment and reduction of water content the main process is recovery of substances from the pre-treated matrix called extraction. Seafood waste biomass matrix is characterized by the substance content if it is nitrogen, lipid, polysaccharide, mineral, lignin based. Quintessential inputs and outputs of extraction process related to the six principles of green extraction are [18]:

- 1) selection of renewable raw resource;
- 2) use of water or agrosolvents;
- 3) reduction and recovery of energy using innovative technology;
- 4) production of co-products;
- 5) development of controlled process and reduction of operations;
- 6) aim for clean green bioactive extract.

Common innovative approaches for the extraction of bioactive compounds are: instant controlled pressure drop (DIC) technology [19], [20]; pulsed electric fields (PEF) treatment [21]; accelerated solvent extraction (ASE) [22], [23]; negative pressure cavitation (NPC) [24]; sub-critical water (SBW) [25], [26]; and ionic-liquid-mediated extraction (ILE) [27], [28]. Capability to control bioprocesses automatically and accurately in their optimal state is extremely important and allows to reduce or limit production costs and increase yields while maintaining the product quality. Regardless of biomass processed it is essential to choose a suitable analytical method for the specific biomass, reaction, and extracts. Most popular are sensor methods based on mathematical models, as real-time data is obtained based on sensor readings.

1.2. Intermediate products from aquatic biomass

In recent years, there has been a significant increase in the interest in marine compounds, source organisms chemical composition and their biological activities. Carroll et al. in 2022 presented a review on natural marine products – 1470 new compounds have been described in 2020, and overall, about 39 000 compounds are described in the *MarinLit* database [29]. Marine by-products from the fish processing industry and fishery by-catch are an important source of bioactive compounds – proteins, amino acids, peptides, enzymes, collagen, gelatine, lipids, ash, chitin, vitamins, and others are of great interest for their high market value. [30]. Fish skin, tendons, cartilage, bone, and connective tissue contain both collagen and gelatine, which can be extracted and used in food and pharmaceutical products [30], [31]. Fish waste represents a huge and cheap source of collagen for the industry [32]. By-products of fish processing is a great potential source for good quality fish oil, which can be used for human consumption, feed, and production of biodiesel [32]. Fish viscera containing digestive enzymes exhibit high catalytic activities at relatively low concentrations and high stability in a wide range of pH [32]. Chitin is a structural component in shrimp and crab shells and squid pens. Marine chitins have been utilized to produce vast array of bioactive products, including chitoooligomers, chitinase, chitosanase, antioxidants, antidiabetic compounds, and prodigiosin [30], [33].

Main fields of application of seaweed are food industry, biofuel production, bioactive antioxidant, antimicrobial compounds, healthcare, cosmetic industry, biofertilizer, and wastewater treatment [34]. Foremost use of seaweed polysaccharides is in food industry – alginate, carrageenan, and agar as food additives with emulsifying, stabilizing, foaming, filler, gelling, binder properties are used in ice-cream, meat, soft drinks, dairy, low fat products, beer, and wine products [10], [35]. Aquatic invertebrates are a source of natural products that can find applications as pharmaceuticals, cosmetics, antibiotics, antifouling products, and biomaterials [36]. Groups of marine invertebrates and products derived from them are: sponges – hydroxyapatite, calcium carbonate, bio-silica, chitin, collagen; cnidarians – hydroxyapatite, collagen; mollusks – proteins for marine glues, calcium carbonate; echinoderms – collagen, proteins, magnesium calcite; tunicates – tunicin – a highly crystalline cellulose nanofiber, proteins. Given the unique and particular characteristics of these organisms, most developed applications aim at bone tissue engineering and other innovative biomedical applications – scaffolds for regenerative medicine, dentistry, and bioadhesives [36].

Reed biomasses are used both fresh and dry; fresh shredded and mulched are used in agriculture for soil improvement and dry reed with moisture content below 20% in construction [37]. Reed biomass is used in variety of added value products: in construction as sound and thermal insulation [38], roofing, combustion [39], ethanol [40], fertilizer [41], biogas, paper and pulp, and feedstock for other products – organic acids, pharmaceuticals, and commodity chemicals [37].

1.3. Blue bioeconomy concepts contributing to sustainability

Internationally used general term “bioeconomy” is crosscutting, encompassing multiple sectors and refers to the share of the economy based on processes, products, and services derived from biological resources. Bioeconomy is one of key components of the sustainable

future economies – development of and transition to predominantly a bioeconomy to address climate change, food security, energy independence, and sustainability of environment. Advancements in science of bioeconomy have opportunity to diversify the industries and jobs, improve human health, and boost rural development [42]. Blue bioeconomy is the part of bioeconomy based on the use of organisms in oceans, seas, lakes, rivers, and aquaculture facilities. In comparison, the term "blue economy" covers all maritime sectors, including, offshore energy, shipping, mining, etc., in addition to the blue bioeconomy sectors [43]. Small-scale fisheries and coastal communities that feed people in need for protein are the most important for bioeconomy strategies and concepts. Blue Justice concept emerged as response to concerns about injustices against small-scale fisheries in Blue Growth agendas. Justice includes a temporal dimension and can include demands for recognition and remediation of past harms. Blue Justice for small-scale fisheries requires information and strategies and, to this end, transdisciplinary research to develop new vocabularies that disrupt dominant discourses on what ocean sustainability is and what it entails. Blue Growth is underpinned by a discourse that frames a trajectory of development that can realize greater revenues from marine resources while at the same time preventing degradation, overuse, and pollution [44]. Blue economy and blue growth concepts are at the heart of most maritime policy initiatives. Blue growth is not a one-size-fits all concept, it is an adaptable framework that can be customized and applied differently across regions and to provide the most benefit to the stakeholders in each case. The global challenges that threaten humanity cannot be solved by addressing climate change alone. This is the correct way to proceed, but on its own, will be insufficient to tackle other key challenges facing mankind [44]. By establishing the common ideals that serve as the cornerstones of biodiplomacy, Europe is taking the lead in the movement toward an integrated and inclusive response to global challenges. In the context of sustainable bioeconomy principles, appropriate monitoring indicators have been found from FAO programs. These indicators will aid in monitoring and assessing the sustainability of policymakers' bioeconomy initiatives and interventions as well as those of producers and manufacturers. Literature demonstrates that the relationship between the bioeconomy and sustainable development goals (SDGs) can vary greatly depending on the strategic goals that a nation chooses for its bioeconomy. The country context will therefore be particularly important for developing bioeconomy plans to promote progress in linked SDGs, as it may modify the nation's primary sustainability goals, and vice versa [45].

Blue Transformation works to advance improved aquatic value chains, sustainable aquaculture expansion, intensification, and efficient management of all fisheries. To boost equal access to profitable markets and increase output, proactive public and commercial collaborations are required. To expand availability and improve access, aquatic foods must also be included in national food security and nutrition programmes along with campaigns to raise consumer awareness of the benefits [46]. Finally, people's perceptions of how aquatic resources should be utilised must be altered. The continued development of multi-stream biorefineries will boost aquatic food production while increasing the economic value of aquatic biomass, so contributing to the improvement of the blue bioeconomy [47], [48]. The global bioeconomy is structured into a number of high-level fora and organisations. With the maturing of the bioeconomy and its growing influence on the industry's transition to a sustainable and climate-

neutral economy, it is critical to discuss strategy alignment, consolidate roadmaps, and link activities [49].

2. METHODOLOGY

2.2. Empirical studies and data analysis

2.2.1. Extraction of lipids from fish waste of round goby

Lipid extractions from fish and fish residues were carried out in the following steps: first, preparation of biomass, then analytical lipid content determination, extraction of lipid from round goby with heat and microwaves [54], indication of lipid quality, and analysis of nutritional value. Homogenization was performed prior to lipid extraction. Total lipid content was determined using the Bligh/Dyer method, which was compared to the alternatives in [50]. Lipid quality was compared using the amount of lipids obtained, colour and viscosity, saponification value, and oxidative quality of the oils (acid value, content of free fatty acids). Saponification value is an important lipid analysis to consider when evaluating the subsequent manufacturing process. It was determined according to the official methodology of the American Oil Chemists' Society (AOCS) [51]. Content of free fatty acids (%) and the amount of acids were determined according to the official method of AOCS Ca 5a-40. Protein content was determined using the Kjeldahl method. By determining the content of protein, fat, water, and ash in fish. This calculation was performed according to the official methodology of AOAC, 2002. Moisture and ash content of the body and head of the goby were also determined. Moisture content in fish was determined by calculating changes in body weight before and after heating. In total, the test was carried out for 20 hours, a temperature of 105 °C was maintained for drying. Ash content was obtained according to the AMC (Royal Society of Chemistry Committee for Analytical Methods) modified method without the addition of magnesium acetate [51]. A drying oven *Ecocell 55* was used to determine the moisture composition. The drying process took an average of 5 h. The analysed sample was weighed every 1 h, after 4 min of cooling in a desiccator, until mass stabilization was achieved. A comparison was also made between head and carcass oils. Free fatty acid content (%) was determined according to the AOC Official Method Ca 5a-40. Oil oxidation can be indirectly determined by the acid value. The acid content was calculated according Formula (2.2.1.1):

$$AV = 1.99 \times FFA, \quad (2.2.1.1)$$

where AV is acid value (mg KOH/g) and FFA is free fatty acid content (%).

The protein content was determined according to the original Kjeldahl method at scientific institute *IFSAHE "BIOR"* [52]. The nitrogen content was calculated according to Formula (2.2.1.2):

$$N = \frac{0.7(V1-V0)}{M}, \quad (2.2.1.2)$$

where

N – nitrogen (%);

$V1$ – 0.1 M sulfuric acid consumed in sample test (mL);

$V0$ – 0.1 M sulfuric acid consumed for the base test (mL);

M – sample mass (g).

The amount of protein was calculated according to Formula (2.2.1.3). Percentage was determined from the total sample, incl. amount of moisture [53].

$$P \% = 6.25 \times N \quad (2.2.1.3)$$

where N is nitrogen (%).

2.2.2. Biochemical methane potential from round goby

Processing waste – heads, intestines, and skin/bone mixture were used for further biochemical methane potential (BMP) testing. Fish waste fractions were separately homogenized using 1500 W kitchen blender. Values of total solids (TS) and volatile solids (VS) were determined prior to the experiments based on ISO Standards. TS was obtained by placing a sample into an oven for 18 hours at 105 °C, then the dry sample was finely ground and placed into an oven for 5 hours at 105 °C. VS was obtained by placing 5 g of totally dry sample into an oven for 11 hours with a heating step 50 °C and then kept at 550 °C for 3 hours to be able to obtain the VS content as a fraction of TS (% of TS). Sewage sludge was collected from wastewater treatment plant “Daugavgrīva” (Riga district, Latvia) directly from biogas reactors. Before BMP experiments, inoculum was incubated for 6 days at 37 °C, with regular degassing. Inoculum was always evaluated for TS and VS content using ISO standards: ISO 14780:2017, [55], ISO 18134 2:2017 [56], ISO 18134-3:2015, ISO 18122:2015 [57]. BMP tests were conducted in a batch mode using 100 mL ND20 vials with a working volume of 50 mL. Each bottle was filled with 30 mL of distilled water, 20 mL of inoculum and 1mL of 0.7M NaHCO₃ buffer basal solution to maintain a neutral pH. Different amount (fresh weight) of different fish waste fraction was added to specific samples based on TS content to maintain ISR around 3. Reference samples containing only inoculum were prepared both for high and low temperature conditions to account for the methane production solely from the fish waste biodegradation. Sample headspace was flushed with N₂ for 30 seconds at a flow rate around 2 L/min before sealing them with butyl rubber stoppers and aluminium crimps. The tests were carried out in dark conditions at a mesophilic temperature of 37 °C in *EcoCell LSIS-B2V / EC 111* incubator and at 23 °C for 31 days. Batches were manually shaken one time per day on average, and all batch tests were prepared in triplicates. In total, three experiments were performed. The tested samples contained heads, skin/bone mixture, and intestines. In total 90 samples were analysed for 6 different feedstock's and two AD temperature conditions (Fig. 2.1). Depending on the type of biomass, the assessment of BMP can eventually require time of up to 90 days [58]. For a more rapid estimation, a theoretical biomethane potential (BMP_{theo}) can be used from the Buswell equation. Chemical composition of fish waste fractions was analysed by the Latvian State Institute of Wood Chemistry.

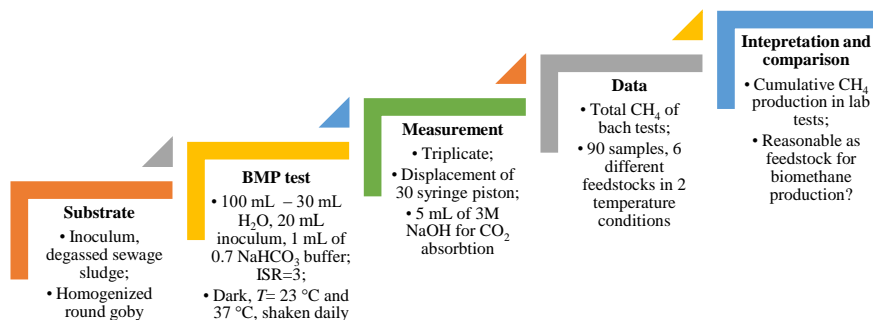


Fig. 2.1. Workflow of biochemical methane potential test.

2.2.3. Multicriteria analysis of common reed use in bioeconomy

Multiple-criteria decision making method was used to evaluate products from reed [59]. It is one of the most commonly used methods in studies that uses both quantitative data (e.g. consumed electricity, emissions, etc.) and qualitative data (interviews, audience opinions, expert testimony) or a mix of both. In the case of lack of data, an environmental engineering assessment, which is based on information on similar products, was considered. To determine the most promising products from reeds using TOPSIS method in accordance with the requirements of environmental protection, the main factors, which are affecting the research issue, were defined as 11 indicators (Table 2.1). Weight of each of factors was determined by nature conservation experts. The value of the qualitative indicators was expressed in a descriptive form and quantified on a decimal scale from 1 to 10.

Table 2.1

Criteria Used for Multi-criteria Analysis

Type of sustainability indicator	Sustainability indicator	Description, quantitative (Q _N) or qualitative (Q _L) examples of indicators
Climate and environmental	Consumption of resources	Consumption of resources in production process of the product – energy, water, chemicals – m ³ H ₂ O, kWh electricity and heat, kg metal, kg fossil or chemicals, kg bioresources, kWh RES, in kilograms of final product
	CO ₂ emissions	Amount of CO ₂ emissions arisen in the production process of product: heat or energy – tCO ₂ e
	Impact on the environment	Impact of raw material extraction and production processes on the environment (air, water, soil, living organisms). Disturbance of hydrobionts – sound, vibration (Hz), pollution (g/hour) emissions of VOC (g/hour), land use (ha).
	Impact on human health	Impact of the product on human health. Effect on respiratory and immune system as substances evaporate from the product.
Technological	Interchangeability	Possibility to replace another biomass with reed biomass, which so far has been used to produce the product
	Consumption of reed	Used amount of reed resources (%) in final product
	Stage of manufacture	Stage of manufacture of the product – technological readiness level (TRL1 – TRL9)
	Complexity	Complexity of the technological process – structural complexity of material, spatial scale, technology size, computational intensity

Economical	Market and investments for launching	Product outlet market (internal or external); necessary investments for launching the product (R&D, facility, licencing, launching investments EUR)
	Product value	Product added value (EUR/kg), green value

2.2.4. Analysis of small psychrophilic plug flow digester with assisted solar heat

Technology and design analysis for the plug flow biogas reactor with solar support was performed prior to pilot scale construction and economic analysis. Based on literature, main technological requirements, size, output of structure suggested, were clarified. Several assumptions about the state of the system were made, and system components and their functions were based on previous scientific work in this field [60], [61]. Biogas yield is assumed to be determined only by digester temperature and feedstock. Heat produced by solar collectors is sufficient to heat digester; heat exchangers are adiabatic – heat loss with the environment can be avoided. Volume of the reactor was chosen to be adapted with the calculated degradation rate of the feedstock. The amount of biodegradable waste is equivalent to 130 kg of food waste per day. To achieve the right balance for reactor volume, two parameters were used to calculate the volume of the digester – organic loading rate (OLR) and hydraulic retention time (HRT). OLR describes the amount of feed processed per unit of the reactor volume per day, expressed in kilograms of total volatile solids (TVS) per day and per cubic meter of the digester (kg TVS/m³day). OLR was calculated by Eq. (2.2.4.1). To calculate the organic loading rate, TS and TVS values were adapted from [62]. The higher the OLR, the more sensitive the system becomes, and monitoring system is required to ensure the process efficiency. Plug-flow digesters function with a higher OLR than traditional digesters, up to 10 kg VS/m³day [63].

$$OLR = \frac{SI \times TS \times TVS}{DV}, \quad (2.2.4.1)$$

where *SI* – substrate input, kg/day; *TS* – total solids, %; *TVS* – total volatile solids, %, *DV* – digester volume, m³.

HRT is the theoretical time period that the substrate stays in the digester [63]. The *HRT* was calculated by Eq. 2.2.4.2:

$$HRT = \frac{NDV}{SI} \quad (2.2.4.2)$$

where *NDV* – net digester volume, m³; *SI* – substrate input, m³.

HRT must be chosen to allow adequate degradation of substrates without increasing the digester volume. To evaluate the potential energy produced from the biogas system, the energy production in this study was observed. Biogas is directly used for heating as a substitute for natural gas; according to [64] one cubic meter of biogas with 60 % methane is equivalent to 4713 kcal or 4.698 kWh electricity. The amount of energy from those aggregates was calculated by Eq. 2.2.4.3. The calorific value of 1 m³ of the biogas (KJ) is:

$$T_E = E_b \times T_b \times E_V, \quad (2.2.4.3)$$

where

T_E – total heat energy per year, kJ;

E_b – calorific value of 1 m³ of biogas with 60 % CH₄;

T_b – total biogas volume in m³ annually;

E_v – energetic value of 1 kcal, kJ.

Solar collector yield, or the useful thermal output of the collectors, depends on the total irradiation onto collector area and the collector efficiency. For estimating required solar collector area, Zijdemans [65] provides a simple calculation method:

$$A_{\text{abs}} = \frac{Q_{\text{demand}} \times SF}{Q_{\text{sol}}} \quad (2.2.4.4)$$

where

A_{abs} – collector absorber area;

Q_{demand} – total heat demand;

SF – desired solar fraction;

Q_{sol} – collector yield [66].

3. RESULTS AND DISCUSSION

3.1. Empirical studies carried out in RTU Biosystems Laboratory

3.1.1. Extraction of fish oil from round goby

Analysis of round goby composition showed that the average length of specimen is $19.53 \text{ cm} \pm 0.5 \text{ cm}$, 25% of that is fish head. Carcass was $77.46 \text{ g} \pm 2.00 \text{ g}$ and head $20.83 \text{ g} \pm 2.00 \text{ g}$. Centrifugation of thermally pre-treated samples showed no visually observable oil recovery. Microwave pre-treatment method also yielded no visible oil fraction. The total lipid content determination with Bligh/Dyer method showed that the highest oil content is in round goby's head $1.00\% \pm 0.13 \%$, oil content in carcass is lower – $0.67\% \pm 0.07\%$. Nutritional composition analysis showed that round goby protein content is $16 \text{ g}/100 \text{ g}$ fish (Table 3.1).

Table 3.1

Nutritional Composition of Round Goby

Part of fish	Water	Protein	Fat	Ash	Carbohydrates
Body	$83.68 \% \pm 12.86 \%$	$16.60 \% \pm 0.40 \%$	$0.67 \% \pm 0.07 \%$	$3.75 \% \pm 0.01 \%$	$0 \% \pm 1.00 \%$
Head	$81.18 \% \pm 1.10 \%$	$16.60 \% \pm 0.40 \%$	$1.00 \% \pm 0.13 \%$	$4.24 \% \pm 0.10 \%$	$0 \% \pm 1.00 \%$

Further fatty acid analysis was not performed due to low lipid concentrations. Free fatty acid content (%) and acid value indicate that properly stored fish is edible. Acid value from the head ($2 \text{ mg KOH/g} \pm 0.47 \text{ mg KOH/g}$) and the body ($1.90 \text{ mg KOH/g} \pm 0.06 \text{ mg KOH/g}$) in extracted fish oil is in accordance with the fish oil quality standards ($<3 \text{ mg KOH/g}$). Free fatty acid content (FFA %) in the oil of the round goby head is $1.03 \% \pm 0.24 \%$ and in the body $0.96 \% \pm 0.03 \%$. Examination of results show that the oil contains large molecular weight fatty acids, saponification value of oil is $233.4 \pm 15.84 \text{ mg KOH/g}$ (head) and $244.65 \pm 54.94 \text{ mg KOH/g}$ (body) (Table 3.2). Environment, seasonality, and feeding conditions show the effect on total lipid content of round goby. In warmer seasons a slightly higher lipid concentration is possible, but not a significant increase in lipid content. This fact does not make round goby suitable for fish oil extraction. For the same species in the Black sea, the lipid content was from $1.60 \% - 2.65 \%$ [67]. Goby has significantly less lipid content than herring and salmon [68]–[69]. Production of fish feed only from this species is also not possible, as a higher lipid content is required for the product to meet the quality criteria. In that case, mixing of fish with higher lipid content with round goby processing waste is required.

3.1.2. Biomethane potential of round goby fish waste

Fish waste fractions show slight differences in chemical composition. Based on the chemical composition, intestines show a promising theoretical BMP potential due to higher carbon and hydrogen percentage of TS and lower ash content than other substrates. Furthermore, high lipid concentration of viscera [70] is affecting the BMP test results, showing

the highest methane yield for the samples with intestines both for high and low temperature conditions. Similar effect was observed by Nges et al. in 2012 [71]. The VS content for round goby's intestines was similar for both biomass sources reaching 82.6% of TS. Regarding total accumulated biomethane volume per test vial, significant difference can be seen between the low temperature and high temperature batch samples. Overall, for the samples that were incubated at 23 °C, an average 23% reduction can be observed in total accumulated biomethane volumes (Fig. 3.1 A). This matches with the trends reported in literature stating that by lowering temperature by 10 °C, biogas productions decrease approximately two times [72].

In net biomethane volumes, the difference between low and high temperature samples occurs to be very low. After calculating the final BMP values (always based on the net biomethane volumes) per kg of VS, the overall average BMP results for low temperature samples are only 2% lower than for 37 °C (Fig. 3.1 B). In total, the BMP difference per 1 kg of VS among the two sets of temperature conditions was only 2 % (Fig. 3.1 A).

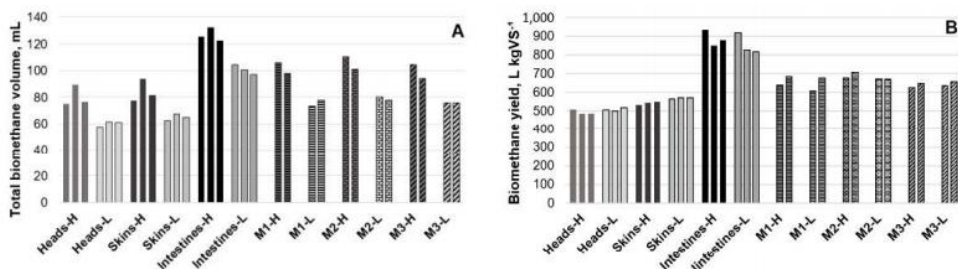


Fig. 3.1. Total accumulated biomethane amount (A) and BMP per 1 kg VS (B) during Experiments 1, 2, and 3. H – 37 °C, L – 23 °C.

During all three experiments, the highest BMP values were obtained in fish intestines, both in high and low temperature conditions. Average biomethane yield from all three experiments at 37 °C 887 L CH₄ kgVS⁻¹ and 853 L CH₄ kgVS⁻¹ at 23 °C. These high values were reached because of high lipid and protein content, especially in gonads – milt and roe that were present in round goby's abdomens. The theoretical BMP yield of lipids is about 1000 L CH₄ kgVS⁻¹, while the theoretical yield of protein is about 490 L CH₄ kgVS⁻¹ [71]. The BMP values of Experiment 1 are higher than those of Experiments 2 and 3, reaching 933 L CH₄ kgVS⁻¹ at 37 °C and 917 L CH₄ kgVS⁻¹ at 23 °C. In comparison, the results from Experiments 2 and 3 were only 850–878 L CH₄ kgVS⁻¹ for high and 816–826 L CH₄ kgVS⁻¹ for low temperature. In springtime, fish are ready for a new spawning season and have larger gonads and contain more mature fish eggs, thus increasing overall lipid and protein relative share in viscera. These results are slightly higher than reported 500 L CH₄ kgVS⁻¹ for perch (*Perca fluviatilis*) intestines [73]. Average BMPs acquired from three experiments for fish heads at high temperature and low temperature was 494 L CH₄ kgVS⁻¹ and 508 L CH₄ kgVS⁻¹, respectively. Skin and bone mix showed slightly higher results, therefore average BMP at 37 °C was 542 L CH₄ kgVS⁻¹ but at 23 °C – 570 L CH₄ kgVS⁻¹. At lower temperatures, average BMP values are slightly higher than at 37 °C, both for heads and skin/bone mixture. It is explained by the fact that for several high temperature samples, after 20 days, biomethane production was delayed, and a slight inhibition of methane production was observable, as blank reference samples on daily basis produced more gas than the samples containing fish waste, indicating the start of

inhibition, which is consequential after digestion of high organic content substrates and rapid VFA accumulation, as can be observed also during dairy product anaerobic digestion [74]. This also is in line with literature where it is suggested that anaerobic digestion under lower temperature conditions is more stable and less volatile fatty acids are accumulated [75]. However, no great change in pH was observed at the end of all experiments, only for few samples lowering from pH 8 to pH 7.7.

Three different fish waste fraction mixes were also prepared. The first mix (M1) contained all waste fractions in equal share based on TS. The second mix (M2) contained all waste fractions in equal share based on wet weight. The third mix (M3) contained all waste fractions in wet weight ratios: 2 parts of heads, 2 parts of skin/bone mixture, 1-part of intestines (based on practical fish processing approach). Average BMP of M1 at 37 °C was 662 L CH₄ kgVS⁻¹ and 642 L CH₄ kgVS⁻¹, respectively. Average BMP of M2 at high temperature was 693 L CH₄ kgVS⁻¹ and at low temperature 670 L CH₄ kgVS⁻¹. Average BMP of M3 at high temperature was 638 L CH₄ kgVS⁻¹ and at 23 °C – 647 L CH₄ kgVS⁻¹. As expected, average BMP was around 660 L CH₄ kgVS⁻¹, which is similar to mathematical average of BMP of heads, skins, and intestines. Other authors report similar results for the Pacific saury, Nile perch, mackerel and cuttlefish wastes, ranging between 562–77 L CH₄ kgVS [76], [77]. BMP for cod meat and intestine mix was reported to be 503–533 L CH₄ kgVS, after 14 days long incubation period [78]. For the 37 °C samples, the main production was observed during the first 7–9 days, accounting for 95% of the total BMP. In turn, for low temperature conditions, the main biomethane production was observed during the first 14–6 days, accounting for 94% of the total BMP. Similar pattern regarding the fish waste highest production rate time shift was reported by [79], where the highest biogas production rate under thermophilic conditions (50 °C) was achieved on day 10, in comparison to 17 days at mesophilic (35 °C) conditions. In respect to this research results, it would be more reasonable to use the HRT of 15 days instead of 30 days for low temperature fish waste anaerobic digestion, as more than 94% of BMP is achieved during this short time.

3.1.3. Evaluation of common reed use for manufacturing products

Reed is an undervalued bioresource that could be used to manufacture bioproducts and get added economic value. There are several inconsistencies between the two sides in terms of availability and quality of resources. Therefore, it is best to use reed as a substitute to other bioresources to produce products. To identify the most promising products from reed, 11 products were studied using multi-criteria analysis:

- 1) thermal insulation panel of reed,
- 2) sound insulation panel of reed,
- 3) reed roofing,
- 4) fuel from reed for direct combustion,
- 5) reed-clay composite,
- 6) reed-fossil composite material,
- 7) biogas,
- 8) extract,
- 9) bioethanol,
- 10) activated carbon,
- 11) paper and cardboard.

The results of the multi-criteria analysis are summarized in Fig. 3.2. For the construction industry, five products were analysed from which sound or thermal insulation panels of reed were equally well and promising and the most ancient and most used type of reed – the roofing

product. The production of reed composite material with binder of fossil origin is definitely not supported because the production of this product does not match the requirements of environmental protection.

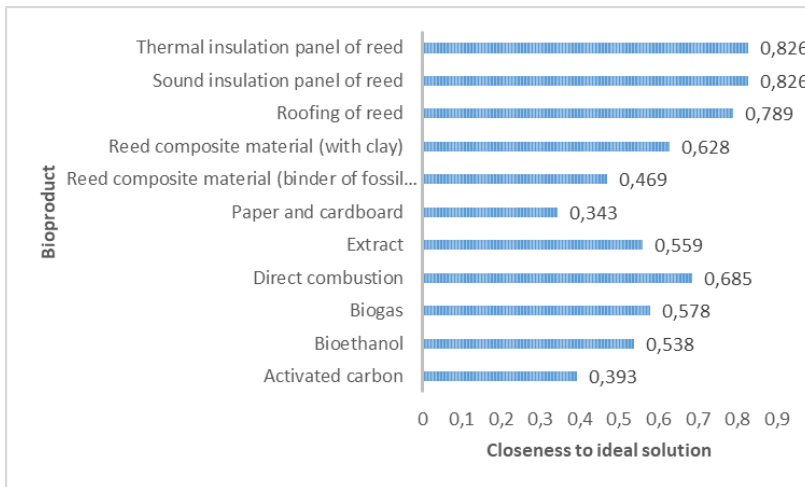


Fig. 3.2. Results of evaluation of products from reed using multi-criteria analysis.

For the energy sector, 3 products were analysed of which direct combustion had the best results. This is mainly because this product requires relatively low investment as its production process is simpler. In the “other products” category were included only 3 products, and extract from reed showed the greatest potential. By comparing all of the eleven analysed products from reed, the most promising products, in compliance with environmental protection requirements, are reed panels for thermal insulation and sound insulation and roofs from reed. The first three products with the highest ratings in the multi-criteria analysis are the products for the construction industry. These are not products with the highest added value, but in any case, from the environmental and climate point of view, they are better than the products for energy sector, as they can replace the products which are made from fossil fuels and temporarily store carbon so that it does not enter the environment and does not contribute to climate change.

To assess the compliance of the most promising products more fully with the requirements of environmental protection, it would be necessary to make and compare their life cycle analysis to determine their long-term impact on climate and environment. From a business perspective, for the most promising products, detailed economic and market analysis is also required. The results show that, in view of environmental protection requirements, the most promising products are those whose production requires dry, winter-mown reed. Which, in turn, does not coincide with the interests of managers of reed areas who want to reduce these areas and therefore mowing is done in summer during the growing season. Planned and well considered management of reed area is needed to find a solution. It would include those areas where it is necessary to eliminate reed stands, mow in summer, and the rest in winter, to ensure availability of the resource in the long term. The use of multiple criterion analysis is a time-saving strategy for selecting the optimal bioproduct for analysis. Better data yields more accurate results, however, when evaluating the calibre of this data, an expert's opinion is crucial.

3.2. Analysis of researched technologies

3.2.1. Extraction of lipids from fish using green extraction methods

Although the green extraction methods can ensure the same quality or product, the green methods like traditional ones also have drawbacks (Table 3.4). Most famous green extraction method is supercritical fluid extraction (SFE) mostly using CO₂ as a solvent. CO₂ is the most traditional SCF solvent because it is easily available at a low price, it is not burning and has low toxicity and high diffusivity with tuneable solvent power. Relative to other solvents CO₂ has mild critical conditions ($T_c = 303.9 \text{ K}$; $P_c = 7.38 \text{ MPa}$) [80]. The 4 major factors that affect the SCF-CO₂ extraction is pressure, temperature, time, and CO₂ extraction flow rate [81–[83] as well as the extraction type: continuous, co-solvent, soaking, and pressure swing [84]. Main limitation of the SCF-CO₂ extraction is its low polarity. CO₂ is a good solvent for non-polar (lipophilic) compounds. Processing scraps of a hake (*Merluccius Merluccius* – *Merluccius paradoxus*) can provide around 10 g of oil/100 g of dry raw material, but the fatty fish species, salmon *Salmo Salar* and orange roughly *Hoplostethus atlanticus* offcut provide greater quantities of 40 g and 50 g of oil and 100 g of dry raw material [80]. Biomass of fish requires pre-treatment – moisture content reduction below 20 %. A freeze-drying method in temperature below $-40 \text{ }^\circ\text{C}$ is used to reduce the moisture, although the particle size reduction does not make a marked difference in the extraction yield [82].

Microwave-assisted extraction (MAE) uses microwaves to warm the solvents in contact with the solid matrix to extract the contents from the sample solution [85]. Microwave extraction is based on the principle that microwave heating system is very selective and it loses very little heat into the surrounding environment. Direct heating affects polar solvents and/or materials. If it is used for biomass samples, the moisture is reduced and it results in a considerable pressure generation, which breaks the cell membranes of the animal or plant cell walls freeing up material in cells [86]. A study that analysed the fat content of frozen fish found that fish oil extraction using MAE gives a similar or even greater yield than traditional extraction methods. Ramalhosa et al. in 2012 [85] used the CEM MARS-X 1500 W extraction unit to extract oil from chub mackerel, sardine, and horse mackerel using petroleum ether : acetone (2 : 1, v/v) as a solvent; extraction yield (raw material) ranged from 4.5 % for sardine to 9% for chub mackerel [87].

More recent studies have shown that ultrasonic assisted extraction using acoustic cavitation and mechanical impact can improve the efficiency of extraction. Acoustic cavitation can disrupt the cell wall facilitating the solvent penetration into plant material and allowing the cell to release the product. Ultrasonic mechanical impact offers greater penetration of solvents in the sample matrix because it increases the surface area of contact between the solvent and the extractable compounds [88]. Ultrasound is in frequencies above the human's hearing levels ranging from 20 kHz to 10 MHz. Ultrasound is classified by several criteria: the amount of energy generated characterized by the sound power (W), sound intensity (W/m^2), or sound power density (W/m^3). The use of ultrasound can be divided into two types: high intensity and low intensity. Low-intensity ultrasound has a high frequency (100 kHz to 1 MHz), and low-power $< 1 \text{ W}/\text{cm}^2$. While high-intensity ultrasound has a low frequency (100 kHz –16 kHz) and

high power (10–1000 W/cm²), it is effective. [89]. Several studies have critically assessed a variety of ultrasonic applications in the industrial extraction of bioactive materials [89], [90].

Table. 3.2.

Overview of Green Extraction Methods for Fish Oil Extraction

Extraction method	Advantages (A) and drawbacks (D)	Main influencing parameters (P) and conditions (C) for extraction
Supercritical fluid extraction (SCF-CO ₂) [91], [92]	(A) Fast. No need for organic solvent and hence extract is very pure. Free of heavy metals and inorganic salts. No chance of polar substances forming polymers. High yield. Lipids can be used for further analysis immediately. Low operating temperatures (40–80 °C). (D) Very pricey and complex equipment operating at elevated pressures. CO ₂ is highly selective – no polar substances are extracted. Supply of clean CO ₂ needed. High power consumption.	(P) Water content, temperature, pressure. Flow of CO ₂ . Extraction type: continuous, co-solvent, soaking, pressure swing. (C) Pressure 25–40 MPa, temperature 40–80 °C, >2 mL CO ₂ /min, soaking time 45 min – 6 h.
Microwave assisted extraction (MAE) [82]–[84]	(A) Decreased extraction time and solvent consumption; higher penetration of chosen solvent into cellular material and enhanced release of cell content in medium. Loses insufficient heat into the surrounding environment. Higher extraction rates, lower temperatures. (D) High power consumption. Heating affects only polar solvents and/or materials. Difficult to scale up. Heat generation, which can lead to unsaturated fatty acid oxidation; low efficiency when using volatile solvents.	(P) Particle size, the used solvent, time, capacity, and frequency of microwaves. (C) 110–2450 W, medium – water or organic solvent.
Ultrasound assisted extraction (UAE) [93], [94]	(A) Decreased extraction time and solvent consumption, higher penetration of chosen solvent into cellular material and enhanced release of cell content in medium. (D) High power consumption. Difficult to scale up.	(P) Ultrasonic frequency, power, time and medium. (C) 25 kHz, 200–2450 W, 30–60 min sonication time. Medium – ethanol, cyclohexane, other organic solvents.
Enzymatic hydrolysis [95], [96]	(A) No need for organic solvent. Using commercial low-cost protease provides an attractive alternative. (D) Expensive/difficult to scale up.	(P) Type, activity and amount of protease. pH. Endogenous enzymes absence. (C) 1–4 h at temperature 40–60 °C, E/S ratio ~ 0.5–5 %.

Enzymatic hydrolysis in comparison with the other methods discussed here is much more widely studied. Adding exogenous enzymes makes digestion process better controllable and reproducible. Thus, enzymatic hydrolysis is an ideal way to recover oil and protein from fish and fishery processing waste. The enzymes and the fish that are used in the process have one thing in common – they must be of food quality, and if the enzymes are of microbial origin, they must not be pathogens. In most cases, alkaline/neutral proteases are used for the hydrolysis because they produce better results than the acidic proteases. Before the extraction, it is necessary to deactivate the exogenous enzymes by heating in about 80–90 °C temperature and adjusting the pH. Oil regain yield depends on the used protease, its activity, concentration, pH,

temperature, and particle size. It is reported that compared with the traditional thermal extraction enzymatic hydrolysis is better in oil regaining and it competes with the solvent extraction [88], [90], [95], [96]. Green extraction techniques are a great replacement for conventional ones. Quantity and quality of fish oil produced are comparable or perhaps superior.

3.2.2. Extraction technologies of valuable compounds from macroalgae

The following criteria should be considered when selecting the appropriate algae for food, feed, and fuel production – they are constantly and steadily growing (open pond/sea), produce large amount of biomass, produce high quality and relatively constant ingredients of desirable nutritional value, survive and grow seasonally and with daily climate change, exhibit high photosynthesis efficiency and energy conversion rate, provide minimal dirt from attachment to environment, it is easy to collect and extract substances [97]. According to HELCOM, the following seaweed species are available for biomass extraction in the Baltic Sea: *Furcellaria lumbricalis*, *Fucus vesiculosus*, *Cladophora aegagrophila*, *Laminaria digitata*, *Chorda filum*, *Fucus serratus*, *Chorda tomentosa*, *Fucus spiralis*, *Laminaria sacchari* [98]. There are several steps to increase the efficiency of seaweed extraction to get the highest quality product. Novel extraction techniques and methods that reduce the cost of extraction, reduce the number of extraction steps and increase the yield of biomolecules. Novel techniques are a significant improvement of existing technologies and are based on the use of physical phenomena (pressure, electric field, ultrasound, microwaves) and biological (enzymes) effects on the matrix [99]. Just before the extraction of the bioactive substances it is necessary to process the biomass in order to obtain maximum yield. Solvent used in the extraction process should be cheap and non-toxic [100]. Several types of extraction methods have been used based on the literature on extraction of bioactive compounds from various matrices. Existing conventional extraction methods include: (1) hydrodistillation; (2) Soxhlet extraction; (3) maceration; (4) percolation; (5) infusion; (6) decoction, and (7) hot continuous extraction [101]. Effectiveness of these methods depends on various influencing parameters, such as solvent properties (polarity, toxicity, volatility, viscosity, purity), sample size and concentration, particle size, time, and polarity of extractant [102], [103]. Drawbacks of conventional techniques are the long extraction time, need for very high purity solvents, energy consumption associated with evaporation of a large amount of solvent, relatively low extraction yield, and selective and thermolabile degradation of the components used [104].

Extraction of compounds from macroalgae can be accomplished by novel methods. These methods also include improved selectivity for isolation of the desired compounds while avoiding the formation of by-products during extraction and adverse reactions [105]. Most of the extraction methods listed below are considered "green" because they meet the standards that have crystallized in green extraction [106], [107]. Compared to conventional extraction methods, the main advantages of innovative extraction methods are higher efficiency, use of water, renewable raw materials, more environmentally friendly treatment conditions, significantly reduced use of hazardous chemicals, safer co-solvents, energy efficiency, and reduced derivatives [101]. Based on reviewed papers [100], [101], [103]–[105], [108]–[111] and others there are 7 novel techniques for biomolecule extraction from seaweed:

- 1) supercritical fluid extraction (SFE) – SC-CO₂;
- 2) microwave-assisted extraction (MAE);
- 3) ultrasound-assisted extraction (UAE);
- 4) high-pressure methods (HPM);
- 5) ionic liquids extraction (ILE);
- 6) enzymes-assisted extraction (EAE);
- 7) pulsed electric field extraction (PEF).

3.2.3. Approach for modelling anaerobic digestion processes of fish processing waste

Fish waste is a mixture of solid and liquid wastes [112]. Studies show that digestion and co-digestion of fish waste has a very good potential for producing biomethane. Anaerobic digestion studies of fish waste shows potential from 0.2 to 0.9 CH₄ m₃/kg VS added (Table 3.6). Fish processing waste poses a distinct technological problem – fish waste releases high levels of ammonia when digested, which then inhibits the digestion of substrates [113]. High concentrations of ammonia can result in the accumulation of VFAs (acetic acid as the main type in the batch tests) and depending on reactor type and organic loading rate can inhibit process especially if the substrate is very high in oils [114]. Co-digestion of two different substrates is a technological solution or at least has a mitigating effect for this problem. Agricultural waste streams have immense potential for energy production both by using dry residues in direct incineration and using dry or wet residues in anaerobic digestion for biomethane production. Global production of agricultural residues from barley, bread, rice, soybean, sugar cane, and wheat are estimated to a total of $3.7_{-1.0}^{+1.3}$ Pg dry matter yr⁻¹ [115].

Overall need for model development was determined by the fact that anaerobic digestion is an intricate group of processes and there is no universal model for predicting/analyzing anaerobic digestion of different substrates. The closest to a universal model is the anaerobic digestion model No 1 (ADM1) developed by the International Water Association (IWA). This model has been widely applied, modified and validated in simulating the digestion of various organic waste. The model includes several phases describing physicochemical and biochemical processes. It consists of a complex reaction kinetics and many concurrent and sequential reactions, which are primarily classed as physicochemical or biochemical. The complexity of such a model necessitates many input parameters, which ultimately results in a large number of stoichiometric and kinetic equations, identification and manipulation of which may prove challenging. Due to the fact that the models set out in ADM1 and other kinetic models described in [125] require a large amount of specialized data, they are not available to farmers and other interested parties with limited scientific knowledge of anaerobic digestion [125].

The first step in designing an anaerobic digestion model of fish waste is to analyze and evaluate the existing literature on theoretical models. The first stage is the mathematical description of relatively simple degradation reactions. The potential biogas yield of anaerobic digestion of a particular type of substrate and the produced gas composition can be determined theoretically by the chemical composition of the used substrates. The production of methane depends on the nutrient content of mainly organic substrates (crude fiber, crude protein, crude protein, N-free extracts), which can be degraded to CH₄ and CO₂. Nutrient content determines the degradability and hence the methane yield that can be obtained by anaerobic digestion.

There is a difference between these nutrients in specific methane yield – crude fat (850 l kg VS), crude protein (490 l kg VS), and carbohydrates (crude fiber and N-free extracts, 395 l kg VS) [126].

Table 3.3

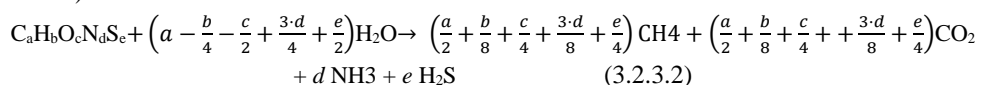
Anaerobic Digestion of Fish Waste

Type of substrate	Incubation days	BMP	Reference
Salmon heads	33	0.828 ± 0.15 CH ₄ m ³ /kg VS	[71]
FW	36	F/M ratio 0.2 with a total maximum methane yield 0.165 CH ₄ m ³ /kg VS COD _{Mn}	[116]
FW	25	0.39 CH ₄ m ³ /kg VS added	[62]
Nile perch waste	42	0.50–61 CH ₄ m ³ /kg VS	[77]
FW	15	180 mL/kg of waste	[117]
Jellyfish <i>Aurelia aurita</i>	-	121.35 mL/g and 870.12 mL/g	[118]
Tuna, sardine, mackerel waste	67	0.470.59 g COD-CH ₄ /g COD added	[119]
FW	67	0.453–0.554CH ₄ m ³ /kg VS	[120]
FW	-	0.380–0.920 CH ₄ m ³ /kg VS	[78]
Round goby waste	-	0.520–0.922 CH ₄ m ³ /kg VS	[121]
Co-digestion of fish waste			
Type of substrate	BMP		Reference
FWS : JA 1 : 1	0.531 CH ₄ m ³ /kg VS added		[71]
SE : FCIW 94 : 6	0.205 CH ₄ m ³ /kg VS added		[122]
FW : SP 33 : 67	0.62 CH ₄ m ³ /kg VS added		[62]
FW : CM 1 : 1.2	1950 mL CH ₄ /kg of waste (biogas)		[117]
FW : WH 1 : 2	0.408 CH ₄ m ³ /kg VS added		[123]
FW : BWS 20 : 80 %, TS	0.482 CH ₄ m ³ /kg VS added		[76]
CM : CI : FS 45 : 22 : 33	0.533 CH ₄ m ³ /kg VS added		[78]
FWS : CM2 16 : 86	0.400 CH ₄ m ³ /kg VS added		[124]
FW – fish waste, FWS – fish waste silage, CM – cod meat, CI – cod intestine, WH – water hyacinth, SP – sisal pulp, CD – cow dung, SE – strawberry extrudate, JA – Jerusalem artichoke, FCIW – fish canning industry waste, CM2 – cow manure, BWS – bread waste silage.			

According to Buswell and Mueller [127], methane and carbon dioxide yield can be calculated with uncertainty of about 5% using Relation (1), contemplating that the chemical composition of used organic matter is known. Relation (1) does not take into account bacterial metabolism – the synthesis of cell biomass and energy for growth and alimentation. According to Eq. 3.2.3.1, the methane fraction of fully degraded glucose is 50%. $C_6H_{12}O_6 \rightarrow 3CH_4 + 3CO_2$.



Organic matter does not consist only from carbon, hydrogen and oxygen. So 25 years later Boyle [128] presented a relation modified from Relation (1), which included nitrogen and sulphur in the composition of organic matter. This allowed the calculation of the ammonia and hydrogen sulfide fraction in the produced biogas, which should be evaluated by ratio (Eq. 3.2.3.2).



Amon et al. [129] offer a model that was developed by carrying out a multifunctional analysis of full regression models, which assessed methane yield from the substrate

composition of energy crops in mono-fermentation via regression models. Basically, it considers the impact of the content of crude fibre, crude protein, crude fat, and N-free extracts on the methane formation by the following equation:

$$\begin{aligned}
 & \text{MEV (I}_N \text{ CH}_4 \text{ kg}^{-1} \text{ VS)} \\
 & =x1 \times \text{crude protein (XP) (content in \% DM)} \\
 & +x2 \times \text{crude fat (XL) (content in \% DM)} \\
 & +x3 \times \text{crude crude fibre (XF) (content in \% DM)} \\
 & +x4 \times \text{crude N-free extracts (XX) (content in \% DM) [30].}
 \end{aligned}
 \tag{3.2.3.3}$$

The next stage in the development of the model would be to analyze the anaerobic digestion kinetics considering the growth of microorganisms, substrate degradation, and product formation. The process set can be divided into continuous and discontinuous, depending on the supply of substrate. In continuous processes, the substrate continuously flows and exits from the system, resulting in a process with constant substrate flow and gas production (equilibrium). Therefore, the growth requirements of microorganisms over time are unchanged. The process of molecular degradation is controlled by bacterial growth kinetics and to a large extent depends on the growth medium. Discontinuous processes are fed only once. Consequently, therefore gas production and substrate degradation changes over retention time, by which growth requirements for microorganisms change permanently. The substrate balance of a continuous or a discontinuous process can be expressed as

$$\begin{aligned}
 dS/dt = D \times S_0 - D \times S + (dS/dt)_r,
 \end{aligned}
 \tag{3.2.3.4}$$

accumulation input output reaction

where dS/dt is the accumulation rate (change of substrate concentration over change in time), D is the dilution rate (flow per reactor volume, in 1/h), S is the substrate concentration, S_0 is the initial substrate concentration, and $(dS/dt)_r$ is the reaction rate [125].

3.2.4. Small psychrophilic plug flow digester with assisted solar heat

Psychrophilic anaerobic digestion with assisted solar heat is a way how to maximize methane content and decrease organics in digestate. The technology is intended for non-profit and autarky. In this work, biogas production in the mini to small-scale as the main renewable energy resource is combined with solar collector as assisted heat. This is offered as a more efficient and faster alternative for composting of waste and better management of biodegradable residues. The system comprises five major components: biomass – pre-treatment and feedstock; digestate; psychrophilic plug flow digester; solar collector unit; and use of gas (Fig. 3.3). The solar collector heat will heat the reactor, if unnecessary, for the heating of accumulator. If it is necessary, firewood boiler can be used for heating the bioreactor.

Biogas producers and users are in a multi-local system. The author used term multi-local (multilocality) to denote a variety of technologies, solutions, applications, and scales of technology in a certain area or region. Development of biorefinery concepts will contribute to integration of biogas – the expansion of the scope, increase in a number of actors and feedstocks. The studies that determine the potential of gas production, technological, and economic conditions are considered but are vaguely related to the social conditions. Thus, these studies can be very subjective in scientific sense and cannot be used as a basis for

political decision-making. Researchers should reckon with many technological styles to develop industry policies, research into biogas systems [130].

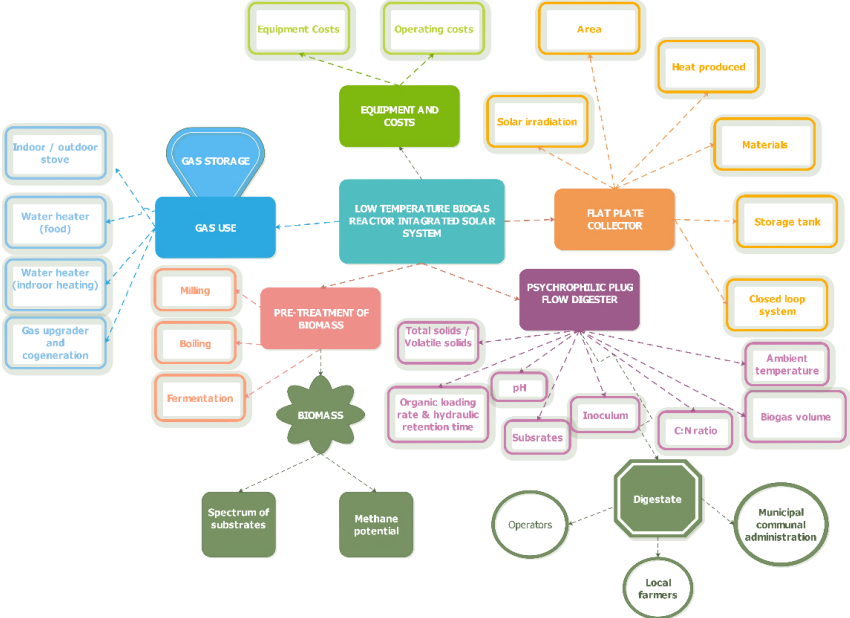


Fig. 3.3. Main components of low-temperature biogas production system with integrated solar heat.

Development of renewable energy sector policies and support mechanisms requires implementation of diversified biogas production and interdisciplinary and applicable scientific research including comprehensive (social) and sectoral (economic) preconditions. The potential for production and uses of biogas globally is very high. Now a tiny part of the available resources is used. Diversifying the production of biogas with the solar collector support system is a way to promote and improve biogas production and, overall, renewable energies in the region (Fig. 3.4) [131].

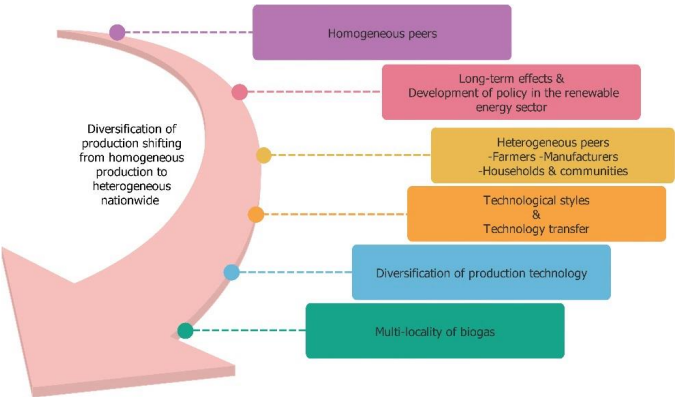


Fig. 3.4. Diffusion of innovation for diversification and increase of biogas production.

To build a solar heating system for Latvia, weather data for specific location must be collected. First, it is necessary to acquire data on the sun radiation and other environmental factors, such as the outside temperature, the relative humidity of the atmosphere, and the wind speed. Due to temperate meteorological conditions, reactor outages are possible during winter when external heating is required, most likely, the break could be from the beginning of January to March. Plug flow digesters gain attention because of ease of operation and portability [132]. What materials will be used for the construction of the biogas digester depends on the local conditions – geological, hydrological, and locally available materials [133]. For the construction of this type of digester, stones and bricks are used as a building material. With the advancement of technology, PVC and polyethylene are used because they are inexpensive [134]. As research in household biogas digesters shows, the psychrophilic biogas reactor in its simplest form may be a plastic or concrete tank. The decision on the reactor elements is determined by the availability of materials and price [132]. Characteristics of the bioreactor and solar components are shown in Table 3.4.

A small producer who generates a variety of food products generates 47 tons of biodegradables a year. Generating 47 tons of waste means that daily production is up to 130 kg of food waste. The results show that biomethane production in a low-temperature biogas reactor has a retention time of 53 days, in a co-digestion mode, with a maximum bioreactor size of 14 m³. Theoretically calculated OLR is 1.72 kg VS/m³ per day. Considering that plug flow digesters can withstand ORL up to 10 kg VS/m³ per day [63]. Therefore, the maximum size of the bioreactor is reduced three times to 4 m³, with OLR 6.88 kg VS /m³ per day.

Table 3.4

Characteristics of the Bioreactor and Solar Components

Component	Details
Digester type	Plug flow digester
Digester volume (for one household)	4 m ³ (2 m ³ to 15 m ³)
Length to width ratio	3.5 : 1
Process	Two-phase system
Gas collecting	The upper part of the digester or balloon
Portability	Portable
Operation	Semi-continuously
Hydraulic retention time	30–60 days
Solid content	7–14 %
Digester temperature range	15–35 °C
Inoculum source	Wastewater treatment plant or cow manure
Digestion unit	Plastic
Feed tank	Metal with pre-treatment unit
Mixing	No
Digestate storage tank	Metal/concrete
Tubes	Plastic, insulated metal
Digester unit heating jacket	Metal tubes/wiring
Insulation	Composite material, rock or glass wool, organic
Feedstock	
Water source	Rainwater tank/underground
Heating source	No heating or solar collector/heat accumulator

Table 3.4 continued

Pre-treatment	Milling, boiling, chemical, drying
Co-substrates	<u>Methane potential in volatile solids (VS) or total solids (TS)</u>
Food waste (FW)	Co-digestion with other substrates was 0.27–0.86 m ³ CH ₄ /kg VS [135]
Fish waste (FIW)	Biomethane production potential of 0.2 to 0.9 CH ₄ m ³ /kg VS [136], [137]
Garden waste (GW)	0.10 ± 0.02 biogas (m ³ /kg VS) [8], [138]
Cow manure (CM)	0.6–0.8 m ³ /kg TS CH ₄ /g TS [139]
Slurry storage, organics content	Digestate storage tank, organics content after digestion is variable depending on reactor temperature and specific activity of microorganisms and other complex factors
Solar collector type	Flat plate collector
Solar irradiation, annual	950–1050 kWh/m ²
Flat plate collector, model	Optional
Gross area of collectors	20 m ²
Inclination angle	34°
System type	Closed loop system
Oriental angle	0°, south
Storage tank	Cylindrical tank
Heat exchanger	Helical coil heat exchanger
Heat transfer fluid	Water + glycol (for freeze protection)
Collector interconnection	Parallel-connected collector array
Control systems	Pumps, controllers, temperature control
Portable	Yes
Solar heat application	Heating of water for different uses

The average yield of biomethane in the co-digestion of food waste and activated sludge, at low temperatures with substrate retention of 28 days, is from 90 to 200 m³ of CH₄/t of food residue, depending on the type and water content [79], [132], [140]. The production unit of this size theoretically could produce an equivalent of ~20 000 m³ of biogas a year if the biomass is digested with maximal efficiency. Depending on the feedstock used and its volatile solids, biomethane content is from 4230 m³ to 14 800 m³ a year. In best case scenario, a system of this size in the maximum effective mode would produce 27.5–96.2 MWh of heat per year (Table 3.5). The thermal energy of the hybrid-system can be used for heating living and production premises, drying wood or food, sprouting grains, growing vegetables and mushrooms, growing insects, earthworms, and similar solutions.

Table 3.5

Characteristics of the Studied Technology

Biomass quantity, annually	47 000 kg
Biomass volume, annually	~95 m ³
Biogas yield for food waste	0.4 m ³ /kg TS
Average FW feedstock density	510 kg/ m ³
Reactor temperature, average	20 °C
Biomethane concentration in biogas	60 %
Hydraulic retention time	~53 days
Reactor size, m ³	4–15 m ³

3.3. Managing aquatic biomass residue issue

Biorefining by green technology's most notable advantage over traditional methods is minimizing losses of functional properties of the bioactive compounds extracted from marine by-products. When developing any framework, it should be considered that the blocks are contextually and informatively different in terms of importance and can be mutually subordinated. Development of a detailed framework requires a great deal of involvement from both industry and related companies, as well as public and labour participation in the process. Although the marine processing sector is characterized by a large amount of data in the primary processing sector and traceability, the use of aquatic waste can be improved. Accurate and enough information in the planning process and operation of the biorefinery ensures successful and smooth operation of the system. Analysis carried out in this work shows that a peripheral but quintessential example of the main blocks of marine bioprocessing can be organized into the following groups:

1. Fishery.
2. Logistics.
3. Bioresources and descriptive.
4. Processing technology in food.
5. Processing technology in added value products.
6. The niche of products.
7. Residue processing technology between or in the final product, including methods of purification or improvement.
8. Product and by-product packaging.
9. Long-term storage of products.
10. Appliances.
11. Legislation and safety.
12. Feedback.
13. Driving forces behind industry – exogenous or endogenous.
14. Planning and information throughout the production of products.

Within the framework of the Thesis, the aspects of processing three aquatic waste biomass feedstocks, possible technologies, and obtainable products were studied. The results show that the sector of aquatic bioresources is given relatively little emphasis on raising the added value and creative use of residues, mainly due to the low quality of resources, the fragmentation of resource provision for economically based economic activity, the low level of investments and high initial costs in innovative processing methods. From these three substrates, it is possible to obtain very high value-added products, which are in demand in the global market, but in these latitudes there is a marked seasonality and there are months when the raw materials to produce the product are not available. Therefore, resource storage and recycling planning is necessary. Storing of resources increases the marginal cost of production. The main task of biorefinery in the processing of aquatic bioresources is, to reduce costs and the amount of low-value residues by ensuring the extraction of several products from several feedstocks in one place. For processing in the biorefinery to be possible, the continuity of electricity is very important, and in case of interruptions – additional backup energy sources, because in biotechnology, the manipulation of plant or animal biomass is carried out at a certain temperature and pre-treatment and extraction methods may require electricity to be used. The use of technologies and equipment and their specific solution in industrial processing requires electronic and mechanical engineering, chemical engineering, engineering teams, and research to ensure the error-free operation of the refinery. Human resource expertise and creativity in

technology solutions provide an opportunity for bioresource processing industries to develop, and development of biorefining is also linked to logistics and supply chains of biomass and additional resources, shortages of materials, and energy can render the production system ineffective.

Choice of feedstock is a significant part of biorefining. Literature suggests that the price of raw materials is the biggest contributor to the final price of product. Therefore, it is important that the raw material is inexpensive and available, with a high content of substances and sufficient yield and quality for the process to be economically competitive. This biorefinery description includes three researched groups of aquatic biomass: fish waste, algae waste, and reed waste. Regardless of other factors, aquatic biomass usually has different origins. Fish and algae come either from wild harvest or aquaculture, reed biomass from green biomass management in wetlands or from special wastewater processing reed growing stations. Pre-treatment, extraction, separation, and purification in one word is called processing, and diverse approaches are used, purification highly depends on further use of intermediate. The goal of pre-treatment is to make slurry suitable to be used as feed in batch or continuous system. Concentration of solids in slurry depends on further extraction process, but it is necessary to ensure fluidity so that the mass can be easily moved through the pipes. To obtain the desired solids loading, the dry matter content of the feedstock had to be determined first. Materials are homogenized to ease the formation of slurry. Animal and plant biomass have different pre-treatment options. Extraction of products can be done one batch at the time run-to-run or continuous process depending on feedstock availability. Seasonality factor plays important role in year-round processing in the eastern part of the Baltic Sea region. Co-treatment of mixed biomass is also possible if pre-treatment was done right. Yield of the product and defining characteristics are indicators of extraction procedures efficiency. Each feedstock has theoretical by-product biorefinery processing stages:

1. Sourcing of feedstock.
2. Pre-treatment.
3. Extraction and separation.
4. Refinement of extract.
5. Storage, packaging, distribution.
6. Product laboratory testing and process efficiency.
7. Treatment of residual biomass and effluents.
8. Evaluation of mass and energy inputs and outputs.
9. Costs and sustainability monitoring.
10. Retail price of extract and application.

Global fish processing waste is increasing, so effort to develop an effective environmentally friendly treatment technology still plays important role in sustainable biomass waste management. Economically and technologically justified sustainable zero residue process is needed for added value and mitigation of environmental impacts. Scientific research on environment and food shows that food-grade fish protein hydrolysate and fish oil recovery have the biggest economic benefit. Full use of waste streams includes two-stages. First, nutrient recovery operations, then, energy and fertilizer production. More likely this means that there is a value chain network of companies is associated with fisheries-biomass processing where intermediates are purchased at a certain price. Quality of waste streams should be defined as the main indicator when utilizing fish resources because it changes the final yield of target compound. Detailed design research and increase in data information can further elevate utility and aid decision-making process [141]. Nutrient recovery from food waste or biomass waste

streams in most cases is a straightforward process of extracting proteins from protein rich by-products. Technology for feed grade protein recovery from seafood wastewater is still being developed and membrane separation, adsorption, and microbe-assisted recovery are the methods that show promising results, but there is a delay in development of new technologies for large-scale manufacturing [142]. Production of energy and fertilizer takes place in one system – anaerobic digestion process of fish waste where digestible by-products are co-fermented into gaseous forms – methane, carbon dioxide, and digestate – liquid mineral and solid fertilizer, and water. Anaerobic digestion is a promising energy recycling technology for biorefinery system, as it may be used for decentralized conversion of large-volume fish waste. Research shows that pre-treatment, anaerobic digestion, and combustion of gas have TRL9 and overall fish waste biorefinery reaches minimum TRL7 because limitation of operational capacity in separate distinctive parts of biorefinery. It should be emphasized that for this well-known technology to be economically profitable, the system requires certain conditions in biomass prices, quality and product sales prices, as well as favorable local policy and legislative conditions [143].

The processing of macroalgae has also become more relevant for manufacturing of value-added products. *Furcellaria lumbricalis* are naturally harvested in the Baltic Sea and as a beach wrack for manufacturing of various products. Commercially viable aquaculture options have also been considered. Low salinity in the middle part of the Baltic sea is the main limiting factor for increased utilization [144]. Interesting and profitable compound extracted from red seaweed is furcellaran, which is naturally sulfated anionic polysaccharide that is used in edible films, food, and cosmetics [145]. Furcellaran is a promising new alternative to plastics in food packaging industry because of non-toxicity and biodegradability, and it is now researched for the production of new modified coatings in food industry [146]. Residue of furcellaran production is also used for methane production using co-digestion and shows profitable results [147].

Processing of reed biomass into ethanol is a promising option – ethanol concentration of 66.5 g/L is achieved [148]. For this technology to be cost-effective using the four-stage ethanol extraction technology, cheap sustainable electricity for pre-treatment and extraction are required. Better treatment operations of reed lignocellulose fraction in future can result in profitable industrial scale reed ethanol production [148]. Remaining fibres are used in the production of biofuels. Pyrolysis of common reed produces gases and volatile materials that are valuable for their energy content. Composition of the products and their energy value are largely influenced by the temperature of pyrolysis [149], [150].

Advanced biorefinery aims at valorisation of variety of biomass into products and energy. The concept has different stages of technological maturity, and biorefinery is subject to constant flux and change. This leads to challenges in assessment and standardization of concepts. Based on the overview of the Federal Government of Germany on technology readiness level (TRL), marine biorefineries have TRL of 5–6 for seaweed and 5–8 for green and lignocellulosic biorefineries. Implementation of biorefinery at a commercial scale necessitates dependable feedstock processing and presents technological, strategic, and sustainability concerns. Most technical hurdles are related to biomass supply and manufacturing costs. Because biomass heterogeneity necessitates distinct pre-treatment and extraction techniques, a multi-feedstock biorefinery with optional variable substrates and creative processing is advised. Biorefinery

biomass cascading demonstrates greater usage of primary biomass and may overcome feedstock rivalry for food and feed. Nevertheless, problems may arise when defining the functional unit, often the functional unit reflects material flows. Also, multifunctional biorefinery causes problems for allocating the environmental impacts to various outputs. Life cycle approach of biobased product makes premise for assisted decision-making for finding the best solution within several scenarios. Further research in marine and green biorefineries is needed because it shows the lowest TRL compared to other biorefineries. Regardless of TRL, technical, economic and environmental assessment of exact biorefinery are needed for better use of biomass [151]. Manufacturing of intermediates from aquatic biomass and value improvement of residues is a technology-intensive process. Techno-economic analysis assessing capital and operational cost factors lead to sustainable biomass utilization [152]. In regional context it is vital to investigate how the Baltic nations might overcome the "Valley of Death" of bioeconomy (TRL6) [154] in the manufacture of additional goods and energy from blue wastes and biomass, as well as the ideal scale of the biorefinery. Performing of extensive research and creating individual scale-up plants to make confident and fact-based decisions on future growth directions is also advised. In comparison, the traditional industries – textile, construction, and energy-intensive industries have higher TRLs both in processing and communication technologies because of the characteristics of circular technologies for different industrial ecosystems, coupled with the need to address the full life cycles of circular products in specific value chains [155].

A blue feedstock biorefinery at plant level includes biomass treatment and pre-treatment units followed by main processing facilities and are based on thermochemical or biochemical conversion. Unwanted by-products are removed, and remaining components are made into the desired end-products. Operation of the biorefinery will depend on the equipment and the selected operating parameters that determine the biomass yield to product and the energy and mass balance of the plant. It is also important to be aware of the investment costs of the plant as well as the costs of integrating the plant into location. Techno-economic evaluations are needed to assess yield, energy efficiency, and production costs [153]. However, both traditional industrial and bioresource processing sectors can improve the use of residues, promoting more complete recycling and reducing volume of waste in landfills. Sustainable and multi-level development of the seafood processing sector is crucial to build economy with smaller carbon footprint. Diversification of production, not producers will strengthen the value chains and sustain enterprises. Clear terminology will aid communication through downscaling the messages from global scientific literature array and upscaling information and data for individual networks.

Recommendations and further research for the development of a biorefinery prototype

- Integrate a national decision-making support tool based on bioeconomy research data, economic and technological analysis in the development of national bioeconomy strategy.
- Establish national guidelines for the use of aquatic bioresources for energy generation.
- Define possible support mechanisms and the scope for expanding bioresource processing based on scientific study of bioresource availability and technological yield.
- Find out how and whether it is feasible to develop bioeconomy goods through social entrepreneurship, as well as operational and financing methods needed.

- Calculate the best site for the aquatic biomass biorefinery using mathematical modelling and geographical analysis.
- Define possible future marine and inland uses of aquatic bioresources through cross-sector and academic cooperation.
- Increase the disciplines of science engaged in the research of aquatic bioresources and promote how to cope with socio-economic problems linked to blue industries.

CONCLUSIONS

Sustainable use of resources in the long term is important – defining feedstock availability and condition, technological-economic justification for the specific situation, product market and retail price. Biorefinery, a processing plant where green principles and bioeconomy concepts are applied, will facilitate the use of financial, technological and land resources. Theoretical assessment of the processing suitability of local aquatic bioresources – fish waste, macroalgae, and reed – shows that these resources have reasonable potential as feedstock to produce bioproducts and energy by different technological approaches.

A review of the literature on green fish oil extraction methods shows that supercritical fluid extraction with carbon dioxide is an excellent way to obtain high-yield, high-purity fish oil at relatively low temperatures that does not contain polar compounds, but the equipment has increased production start-up and operating costs compared to traditional methods. A by-product of supercritical extraction is partially hydrolysed fish protein. The results of the laboratory research of the round goby, found in the coastal waters of Latvia, show that the species is not promising for use in fish oil extraction because the oil concentration in the fish biomass is only 1%, but the total protein concentration is 16%, therefore, in order to fully use the biomass, it is preferable to process it into hydrolysed protein, which can be used to produce food additives and animal feed. Liquid residues of hydrolysate production can be digested into biogas and the solid residues processed into fertilizer.

The results of round goby waste anaerobic digestion show that biogas production at low temperature (23 °C) takes twice the time, thus prolonging the hydraulic retention time, which means increased size of biodigester to produce same volume of biogas. Also ~ 23 % decrease in total produced biomethane was noticed. The best available technique for successful treatment is biomethane production in co-digestion regime with high carbon substrate, e.g., garden waste. Additional experimental data from the batch tests and continuous systems, and parallel, modelling of fish waste treatment process will assist reaching overall sustainability of fish waste digestion and favourable digester size in coastal rural areas. Undeniably technologic and economic analysis and supply chain strength should be assessed when optimizing energetic waste treatment options of seafood processing industry.

Multi-criteria analysis of reed biomass management options shows that production of value-added products is being implemented. From environmental and economic point of view the highest value products are construction materials – insulation panels and roofing, which have been harvested in winter. Literature suggests that manufacturing of ethanol on a small scale from reed could be possible using hot water sodium carbonate pre-treatment and semi-simultaneous saccharification and fermentation. Fibre residues from ethanol production are recommended to be used for pyrolysis fuel production.

Feasibility analysis of low-temperature biogas reactor with solar panel support as a management tool for household-to-small business biodegradable waste was performed. The analysis confirms that solar assistance to biogas increases the production of biogas, efficiency of production, and costs and decreases toxicity of digestate. There is socio-economical value of technology in two contexts – a renewable technology reduces waste and produces energy and serves as bottom-up integrator of renewable energy. Investigation showed that multilocality of biogas must be taken into consideration when the policy of the renewable energy sector is developed, particularly in rural areas. Implementation of a functioning system requires additional research for small-scale renewable energy hybrid systems – system modelling, techno-economic analysis, identification of specific technical parameters of the workable system in precise location, defining the boundaries of the hybrid system.

Blue bioeconomy vocabulary has become more precise when discussing the blue economy. Vocabulary used in scientific journals can help developed countries better comprehend the maritime sector conditions of less developed countries and help them have discussions about how to support their sustainability initiatives and protect natural resources. The use of terminology in research is recommended, since it will benefit both countries that launch the commercialization of research-based products as well as smaller less developed pelagic fisheries. In both science and politics, achieving the long-term strategic goals of sustainability and nature preservation necessitates making choices today and taking steps tomorrow to guarantee that there will be resources and a functioning society. It is also essential to develop action programs/development strategies in particular sub-sectors and have clearly defined national government goals for the blue bioeconomy business to advance. It calls for thorough understanding of and keen interest in particular crucial subjects in the growth of the aquatic bioresources technology industry from universities and research institutions. It is crucial to ensure international cooperation to undertake research, train young scientists, develop technologies with the potential for commercialization, and create new beneficial goods and services.

The author's research examined the use of Latvian water bioresources in the creation of products using various processing techniques. It also examined the resource composition in resources that have not previously been researched. The Thesis compiles information that is currently accessible regarding the primary categories and makeup of resources and residues, processing techniques, and products that can be obtained, as well as the processing of secondary biomass residues. Based on scientific data, a conceptual review of the integration of three distinct resources (fish, algae, and macrophytes) was performed. Because the technical readiness of these methods for extracting products from fish biomass varies, experiments in extraction using small-scale bioreactors are required to gather data about the factors that need to be optimized in the extraction process, costs, etc. to develop products on a larger scale and safeguard cross-over TRL 6. Whenever resource availability varies owing to natural factors or anthropogenic impacts, research into the processing of aquatic bioresources is crucial to ensure the viability of various future scenarios in the context of biomass management. In developed countries, the technical level for using macrospecies is currently very high, but there are chances to build integrated multi-trophic aquaculture, boost processing efficiency, and increase consumer acceptability of the products. Biotechnology offers more opportunities to produce specialized products, since it allows for the use of state of art modern techniques for studying

microorganisms and the ability to develop products in bioreactors that are tailored to specific needs. Although these microbial technologies typically do not operate on an industrial scale, funding and successful operation of such initiatives are nonetheless achievable.

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