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Review Article

Recent Developments in Recirculating Aquaculture Systems: A Review

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This article presents the recent advancements in recirculating aquaculture systems (RAS). The review explores new developments and potential future breakthroughs in RAS systems across leading countries. It highlights technical and technological advancement in plant management aimed at improving water quality, production efficiency, and animal welfare. A significant aspect of recent progress is the integration of artificial intelligence (AI), which is being used to optimize system performance, enhance monitoring, and support more precise and predictive management strategies. The review also addresses advancements in pathogen control and the prevention of disease outbreaks. Specific case studies of cutting-edge RAS systems from different parts of the world are discussed. The review also investigates how the improvements in RAS technology can help mitigate environmental impact. Finally, the paper focuses on advancements in the production of six fish species farmed in Europe, namely Atlantic salmon (*Salmo salar*), European seabass (*Dicentrarchus labrax*), gilthead seabream (*Sparus aurata*), yellowtail kingfish (*Seriola lalandi*), arctic charr (*Salvelinus alpinus*), and rainbow trout (*Oncorhynchus mykiss*). This review is part of the ERA-NET BlueBio cofound-funded project titled "Optimizing land-based fish production in next generation digital recirculating aquaculture systems," which is focusing on the above-mentioned fish species.

Keywords: aquaculture; innovative aquaculture tools; microbiota; RAS

1. Introduction

Oceans have always been a significant source of food for humans. Today, however, the biological productivity and stability of the oceans are constantly challenged by climate change, pollution, and overexploitation [1–3], leading to social and economic losses. However, high seafood traceability has helped reduce these losses and promoted marine conservation and better fishing practices [4].

The aquaculture sector may serve to mitigate the severe challenges imposed on the oceans.

This sector is growing faster than the global population and plays a vital role in uplifting food production, security, and human nutrition [5, 6]. Scientific and technological advancements in aquaculture have led to much safer, as well as environmentally and economically more sustainable seafood production. Such advancements include innovations targeting the produced species (e.g., genome selection/editing

[7, 8], alternative feed ingredients to replace fish meal and oil [9], and reducing carbon footprint by the use of solar energy [10]. With the advent of land-based, next-generation recirculating aquaculture systems (RAS), commercial production of seafood products has experienced a paradigm shift. These systems have evolved into technologically advanced land-based fish farming facilities for different marine and freshwater species, providing solutions to profitable seafood production and concomitantly reducing the environmental impact of aquaculture [11]. RAS has emerged as an adaptation strategy to overcome hurdles associated with climate change, biosecurity, and other environmental variables associated with fish production. Furthermore, this technology and its advancements have been integrated over the last decade into traditional land-based aquaculture practices to improve the sustainability and profitability of the aquaculture sector.

Fish farming pioneers are now blazing the trail by building controlled indoor fish production facilities worldwide. Over the past few decades, there has been an increase in aquaculture facilities based on RAS. This is primarily due to its reduced environmental impacts over traditional fish farming systems, providing better water management, eco-friendly farming, environmental biosecurity, reduced cost and labor requirements, and improved feed conversion rates [11, 12]. In this regard, RAS provides better control of the production environment, leading to enhanced growth, better hygiene, and disease management of farmed fish by reducing water consumption and improving water recycling [13–15]. Depending on the farm design, water recycling can be between 30% and 100%. Farming fish in land-based indoor facilities reduces the effluents with a negative impact on the environment and, at the same time, helps to conserve wild fish populations, for example, by minimizing escapees. However, considerable amounts of sludge may be produced in commercial RAS, and several strategies are used to manage and utilize this side-stream, including, for example, anaerobic digestion [16], nutrient recovery [17], and composting and soil amendment [18]. In addition, recent research revealed the potential of sludge from RAS as a resource for the production of low-trophic species, including microalgae, polychaetes [19], and insects [20]. By adopting various methods to convert sludge into valuable resources, the sustainability of RAS operations can be enhanced, promoting a more circular production model and minimizing environmental impact. However, to fully realize the potential of sludge valorization, further research is necessary, and legislative barriers must be addressed.

The main environmental advantages of indoor farming in RAS can be quantified as follows: (1) it requires far less water than conventional systems as they reprocess the water, making it particularly suitable for areas with limited water resources; (2) it requires far less surface area due to high stocking density; (3) it can control the water quality by maintaining water parameters such as temperature, dissolved oxygen (DO), carbon dioxide (CO_2), total ammonia nitrogen (TAN) and nitrate levels; and (4) it ensures increased biosecurity, as inflow water is in far more controlled quantities compared with a flow-through system. Further environmental advantages include improved opportunities for waste management, nutrient recycling [21], controlling biological pollution by preventing alternative escapees [22], reduced power use for heating or cooling water, and reducing CO_2 emissions associated with food transport. The minimal environmental footprint is another potential advantage that makes RAS a sustainable alternative. However, the establishment of the main functional components of a RAS system requires a higher initial investment and skilled technical staff [12]. The main functional components of a RAS include (1) production tanks, (2) mechanical filtration, (3) nitrifying biofilter, (4) disinfection (ozone + UV), (5) CO_2 degassing, (6) skimmer, and (7) auxiliary systems which monitor and control physicochemical parameters such as oxygen, pH, temperature, salinity, etc. (Figure 1). Water usage and nutrient emissions can further be minimized by (8) denitrifying biofilters and (9) phosphate precipitation. It should be noted that the specific design of these functional components differs between the different technology providers.

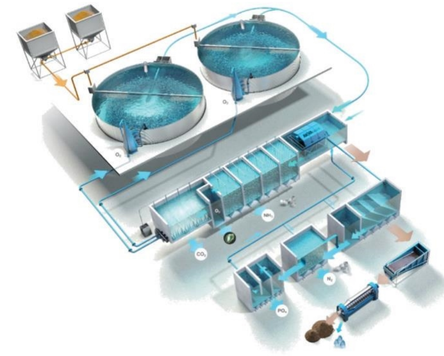

Figure 1: Schematic illustration of a state-of-the-art RAS. *Source:* Akva group 2022. As a first step of the water treatment loop, the water from the fish tank is filtered by drum filters to remove particles. The nitrifying biofilter removes ammonia and further particles (indicated by NH_3), followed by degassing to remove CO_2 from the water (indicated by CO_2). The water is then oxygenated and pumped back into the fish tank. To reduce water consumption, nitrate can also be removed microbiologically as N_2 in a denitrifying biofilter (indicated by N2). In the illustrated set-up, a split loop is generated, where the sludge from the drum filter is separated by a plate separator, followed by denitrification and phosphor-removal by chemical precipitation (indicated by PO4). The treated water is then added back to the main treatment loop. Oxygenation of tank water is indicated by O_2. RAS, recirculating aquaculture systems.

The concept of aquaculture dates as far back as 3000 BC when carp were bred in artificial lakes in China. As early as 1883, a growing interest in culturing salmon, trout, and oysters led to the first International Fisheries Exhibition that took place in London, England. Among the attendees, a Scottish landowner presented that pure water and rat-proof drains were necessities for success—a first attempt to address biosecurity [23]. The first scientific research on RAS was conducted in the 1950s in Japan, focusing on biofilter design for carp production [24]. Danish fish farmers pioneered the application of RAS for the commercial production of European eel in the 1980s. Since then, the land-based aquaculture

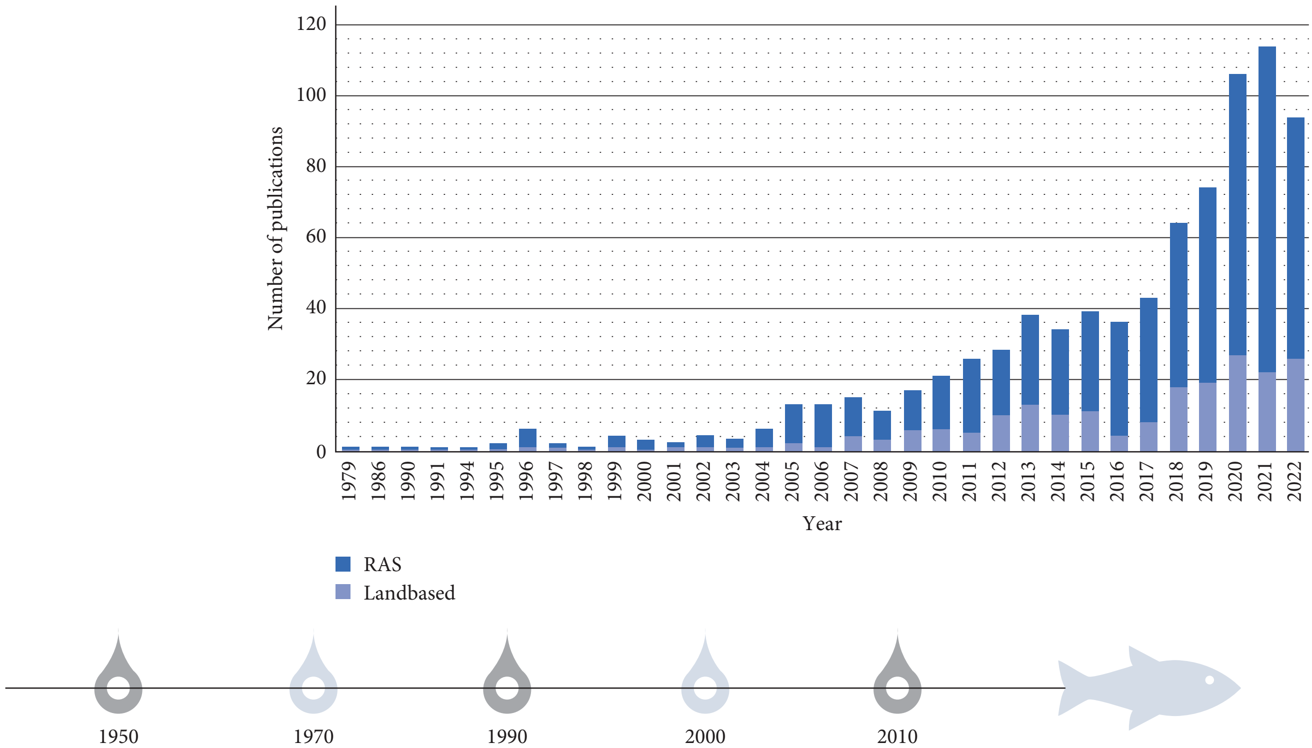

Figure 2: History and development in RAS: a timeline of publications on RAS and land-based aquaculture. RAS, recirculating aquaculture systems.

industry has received more attention due to its unique advantages, receiving support through policy, legislation, and financial backing in Europe and North America [11, 15, 25, 26], leading to further development.

In the early 1980s, it was difficult to sell RAS technology, as many fish farmers attempted to build small-scale systems themselves—some being more successful than others. In turn, the unsuccessful fish farmers sought advice and consulting companies selling their knowledge and experience, which emerged in the 1990s in Denmark, Netherlands, and Norway. Initial efforts for technical improvements were often hampered by reporting inconsistencies and a lack of common terminology, resulting in miscommunication between scientists, designers, construction personnel, and operators [24]. This organic growth within the land-based aquaculture sector contributed by trial and error in developing today's modern RAS. A brief timeline showing the succession of publications in the field of RAS as a measure for corresponding R&D activity is shown in Figure 2.

2. Recent Trends and Innovations in RAS

2.1. Machine Learning Technologies. The agriculture-associated industries have been assisted by computer vision, high-resolution proximal and satellite imagery, sensor networks, the Internet of Things (IoT), data mining, and artificial intelligence (AI) technologies. Similar technologies are adopted in the aquaculture sector to manage aquaculture facilities more effectively [27, 28]. The main advantage of AI is the ability to learn from historical data instead of using equations or assumptions. This results in a powerful tool for predictive modeling (The AI Revolution—RASTECH Magazine). Technological advances in high-performance computing and convolutional neural network technologies have enabled the scientific community to use such machine learning approaches to optimize various aquaculture management practices, such as fish behavior monitoring [29–31], feed optimization [32], disease detection [33], noninvasive size and biomass estimation [34], fish identification [35, 36], population counting [37], age-based fish classification, sex identification [38], water quality monitoring [39], and mortality [40]. Selected machine learning technologies that have drawn significant interest from aquaculture researchers are as follows.

2.1.1. Advancements in Monitoring Fish-Behavior. The use of computer vision to quantitatively analyze fish behavior was first seen in the mid-1990s [41]. At this time, only offline behavior analysis was possible due, at least in part, to limitations in computing performance. Recent computer vision and machine learning advances have enabled significant progress in self-driving cars [42] and uncrewed aerial vehicles [43]. This progress has been a function of increasingly powerful computing capabilities, specifically graphical processing units (GPUs) performance and their employment in AI, known as deep learning. The advances in computing power, computer vision, and machine learning approaches made it possible to track entities in real-time, creating numerous opportunities in aquaculture.

A continuously growing number of fish farms apply AI, edge computing, 5G, big data, IoT, cloud computing, machine vision, deep learning, robots, and other data-driven technologies, enabling for precision fish farming to improve productivity and fish welfare [44]. These technologies can be applied in aquaculture for various purposes, such as environmental monitoring, screening fish seeds, smart feeding, analyzing the behavior of aquatic species, diagnosing diseases, and managing logistic processes.

The recently reported smart aquaculture farm management system is an impressive example of how AI and IoT technologies (AIoT) have been employed to demonstrate the operation of a deep learning-based smart California Bass fish pond, with the major intention of reducing the manpower required for the maintenance of a corresponding aquaculture farm [45].

A recent study by Ranjan et al. [40] presents an imaging platform (RASense1.0) developed for underwater image acquisition. Here, the authors acquired data from the imaging sensors under two light conditions arranged in sets of 100 images and annotated as partial and whole fish and concluded that the developed underwater platform could effectively acquire images/videos at a depth of 0.5 m below the tank water level in both ambient and supplemental light conditions.

Machine vision has found application in correlating fish swimming behavior with the presence of hydrogen sulfide (H_2S) in Atlantic salmon RAS [46]. The study revealed that fish exhibit a concentration-dependent stress response to H_2S, characterized by a faster and erratic swimming pattern, as well as a loss of schooling behavior. The system successfully identified a threshold in the response at a concentration of 30–40 µg/L, which is below the toxicity threshold for Atlantic salmon, set at 60 µg/L [47]. This suggests that machine vision could serve as a potent tool for providing early warnings of suboptimal water conditions.

2.1.2. Advancements in Assessing Health and Welfare. As seafood production in RAS continues to grow and intensify, there is an increasing public concern over the health and welfare of fish produced in such systems. Various indicators have been recommended to monitor welfare and health during farming. However, detailed and reliable monitoring is still not standard in aquaculture operations [48]. In recent years, the field of AI has witnessed unprecedented advancements, encompassing machine learning, deep learning, and neural networks, demonstrating remarkable potential in analyzing large volumes of data collected from fish farms [49]. The authors suggest that employing AI algorithms has the potential to provide valuable insights into fish behavior, growth patterns, feeding behavior, and environmental factors affecting fish health. By exploiting AI's capabilities in data analysis, pattern recognition, and predictive modeling, precision farming tools can be developed, enabling better decision-making and targeted interventions based on reliable and objective data. The integration of such tools in RAS will help to identify deviations from optimal conditions and enforce targeted corrective measures, thereby reducing the risk of fish mortality and improving health and welfare.

Recent examples of such precision farming tools are, for example, "MyFishCheck," a fish welfare indexing model [50], and MortCam, an underwater mortality monitoring and alert system [51].

2.1.3. Advancements in Improving Feeding Systems and Water Quality. Machine learning models of fish behavior facilitate real-time fish health monitoring and optimized feeding according to fish appetite. Precise feeding in the RAS is a critical scientific problem that requires a solution. Chen et al. [52] developed an intelligent feeding technique in RAS for rearing shrimps by accurately predicting shrimp biomass. They introduced multiple linear regression, artificial neural networks and a support vector machine (SVM) to develop the shrimp biomass predicting model. The intelligent feeding machine can select the optimal model, calculate the biomass, and determine the appropriate feeding amount by reading the sensors in real-time. Video tracking, in combination with AI-analyzed fish behavior monitoring, has the potential to directly indicate RAS water quality and provide early warnings, as reported in a recent review [53]. One significant limitation of modern machine learning technologies is that they are tested only at smaller scales (small and shallow tanks ($<1\ m^3$) with fewer fish (<100)). Despite impressive results, such models cannot yet be employed in semi-commercial to commercial RAS systems scale tanks (100–1000 m^3).

2.2. Current and Emerging Technologies for Water Treatment and Minimizing Water Consumption

2.2.1. Challenges in RAS Water Treatment. Reducing water consumption in RAS has numerous benefits and brings economic and environmental gains. However, new problems arise from water recirculation in the system due to the formation and accumulation of compounds, which can harm fish health and RAS productivity. The main concerns are large quantities of particulate matter, that is, residual feed, fish feces, small suspended solid particles, nitrogenous compounds like ammonia, nitrite as well as bacteria and CO_2 [54]. These problems are currently addressed in RAS through the application of filtration, biological filtration, and disinfection.

However, these conventional water treatment methods do not remove all the harmful compounds that can be found in the inlet water or that are formed and accumulated in RAS, for example, hormones, veterinary drugs, pharmaceuticals, and taste and odor (T&O) compounds as well as H_2S. Waterborne hormones accumulating in RAS water can cause early maturation in fish, which is detrimental to farm profitability due to the associated decrease in growth and in feed conversion efficiency [55]. T&O compounds such as geosmin (GSM) and methylisoborneol (MIB) can taint fish flesh (at 1 ng/g in fish flesh) [56], causing consumer dissatisfaction and lead to economic loss [57]. H_2S, formed in the presence of carbon-based compounds and sulfate or methane-reducing bacteria, can lead to mass mortality of cultured fish within hours [56, 58]. Inlet water and the use of pharmaceuticals to control disease outbreaks can be a source of contaminants of emerging concern (CEC). It has been shown that pharmaceuticals, pesticides, stimulants, and polycyclic aromatic hydrocarbons (PAH) have been detected in aquaculture water [11]. A lower risk of detection of CEC is expected in RAS; however, CEC can still be introduced with inlet water or be added into the system, especially hormones, steroids, parasiticides, and antibiotics [59]. Additionally, a new class of micropollutants, antibiotic resistance genes (ARGs), were also reported in RAS [60]. Consumption of fish that bioaccumulated antibiotics or ARGs will pose a threat to human health [61].

RASs operate at various water quality parameters, for example, temperature, salinity, DO, and pH, due to the requirements of the farmed fish. Water quality parameters like total suspended solids (TSS) are dependent on the bred species, feeding frequency, and environmental factors [62]. Furthermore, fish show varying tolerance to contaminants, for example, nitrate. Although nitrate is considered harmless, high levels can have a negative effect on growth and feed conversion ratio (FCR). A safe nitrate upper limit for Atlantic salmon has been determined to be 100 mg/L in freshwater [54].

As a result, challenges for RAS are strongly related to (1) water quality monitoring and, if direct monitoring for contaminants is not available, (2) monitoring of fish behavior as a response to pollutants in the system, and (3) selection of appropriate water treatment methods and development of processes in which micropollutants are removed from RAS water, without negatively affecting fish or microbial community. Hence, in the next sections, we discuss developments and needs in water quality monitoring and water treatment technologies.

2.2.2. Advancements in Monitoring Chemical Water Quality and Composition. Water monitoring serves multiple purposes in RAS; first, it allows for the control of basic water parameters important for stable fish farming; second, it provides information on the efficiency of the water treatment system; and third, it enables fast response in case of system failure. Traditionally, handheld sensors have been used at certain time intervals [58]. Nowadays, real-time monitoring of basic water quality parameters (such as temperature, pH, DO, oxidation-reduction potential [ORP], and TSS) is manageable and does not require high investment costs, as reported in a recent review [58].

However, monitoring of micropollutants, T&O, and H_2S requires the development of simple methods with low detection limits, especially in the case of T&O compounds that can be present in concentrations as low as a couple of nanograms per liter. Despite this urgent need, no simple detection and monitoring methods are available yet, and quantifying these compounds still requires advanced equipment, that is, gas chromatography–mass spectrometry (GC–MS) and trained technical staff. Continuous monitoring of water quality, together with an automated control system, would allow for fast response in case of, for example, buildup of H_2S concentration, therefore preventing incidents that have led to acute fish mortalities (e.g., Atlantic salmon mortality in Norway and Denmark) [56].

Son et al. [63] reported on real-time monitoring of T&O compounds by means of a bioelectronic nose, which, as reported, was able to selectively detect GSM and MIB at concentrations as low as 10 ng/L. To the best of our knowledge this promising solution was not tested in RAS.

Recent advancements in RAS include the SeaRAS AquaSense, Aquaduct, and Blue Unit System [64]. The AquaSense utilizes real-time monitoring of H_2S in parallel with other water quality parameters, such as CO_2, O_2, pH, and temperature, through a set of autonomous wireless sensor units installed in multiple locations in RAS. SeaRAS Aquaduct is a unit that transports, degasses, and oxygenates the water and, at the same time, skims out small particles, leading to an improved water quality of existing RAS. Like the AquaSense system, the lab station from Blue Unit (https://blueunit.com/) allows for online monitoring of H_2S and other water quality parameters, including CO_2, DO, turbidity, carbonate, pH, redox, temperature, ammonia, dissolved solids, salinity, and conductivity. Additionally, the previously mentioned fish behavior monitoring can be used as an early warning system for increased H_2S concentration [47].

2.2.3. Conventional Water Treatment Technologies, Limitations, and Advancements. Numerous solutions exist for water and wastewater treatment; however, their direct application to RAS is not straightforward, as the treated water needs to be recirculated back into fish tanks. Hence, it cannot contain chemicals affecting fish health or growth. Furthermore, the preferred solutions have small floor space, high treatment efficiency, low power consumption, and reliable and easy operation. The selection of specific solutions is often also dependent on the hydraulic retention times (HRTs) in RAS [65]. At reduced water exchange rates, the water treatment strategy becomes crucial to prevent the accumulation of indigestible feed components and metabolites produced by the fish and the microbial community. Therefore, the currently available technologies mainly focus on removing this contamination from the system, that is, particulate organic matter (POC), nutrients, and microorganisms. As mentioned before, the RAS water treatment consists of several units: filtration, nitrification, disinfection, CO_2 degassing, denitrification, and phosphate precipitation. This section will describe the role and challenges in applying filtration, biological filtration, and disinfection methods to RAS (Table 1).

2.2.3.1. Filtration. Removal of POC in RAS is important for maintaining high water quality [66] as solids are its key component, holding 10%–30% of the total nitrogen (TN) and 30%–80% of the total phosphorus (TP) [59]. Particles in RAS water contribute to various possible problems, that is, gill damage and increased stress in fish, support bacterial proliferation, reduce light penetration, and facilitate pathogen infections [59, 67, 68]. A variety of particle sizes are found in RAS, both precipitable TSS (>100 µm) and non-precipitable TSS (<100 µm) [67]. A recent investigation by Becke et al. [69] has shown that RAS solid load is dominated by 30–100 µm (EEAW—equivalent elliptical area width) particles. Their findings contradicted previous research suggesting the importance of fine particles (<20 µm) in RAS. Generally, settleable solids are removed in fish tanks as they accumulate at the bottom and can be easily removed by, for example, double drain devices [70]. Many solid–liquid separation devices have been developed and used in RAS systems, like sand filtration, parabolic screen filters (PFS), and microscreen drum filters (MDF) [71], and can remove particles

Table 1: Water treatment technologies used in RAS.

Technology	Removed contaminants	Efficiency	Disadvantages/challenges	Comments
		Disinfection processes		
UV disinfection [53, 60]	Pathogens	Disinfection rates proportional to light intensity and size of pathogen	+Easy to install and operate +No harmful effect to cultured species −Clear RAS waters required as turbidity affects effectiveness −Requires routine replacement of UV lamps (typically every several months) −Can decrease UV transmittance	
Ozonation [53]	Pathogens, ammonia, nitrite, COD	56.3% and 55.8% removal of heterotrophs and total coliform at OPR = 375 mV 97.6% and 99.1% removal of heterotrophs and total coliform at OPR = 375 mV with UV	−In marine systems, the risk of bromate formation −Iodine consumption +Increases dissolved oxygen +Low ozone dose needed (0.33-0.39 mg/L) in comparison to traditional water treatment −High installation costs −Requires operation by technically trained staff	Combination with UV irradiation to eliminate residual ozone Off-gas should be vented outside to ensure the safety of operating staff
		Mechanical filtration		
Swirl separators	Large-particle suspended solids	90% >250 µm	−Not good for removal of small particles	
PFS	Suspended solids	80% >70 µm	+No power consumption +Simple structure −Low automation degree, −Manual cleaning	
Microscreen drum filters [53, 60]	Suspended solids	>60 µm	+Require very little floor space +Do not need daily washdowns −Limited ability to remove fine suspended particles +Large surface area available for filtration	
Floating bead filters [53]	Suspended solids	>30 µm single pass all suspended particles, after multiple passes	−Requires backwashing	Widely used in both freshwater and marine applications
Sand filtration	Suspended solids	30–75 µm	+Low cost +Simple structure −Needs regular backwash	
FF/protein skimmer	Surfactants, DOM, suspended particles, COD, BOD, TN, TP	80%–90%, <30 µm 7%–32% TN and 30%–84% TP	−Not effective in freshwater systems	Often combined with ozonation
		Biofilters		
Fluidized sand biofilters [60]	Ammonia (NH_4-N), BOD_5, COD, TP, TAN	86%–88% TAN, 66%–82% $cBOD_5$, and 1–2 $\log_{10}$ total coliform bacteria	+High nitrification capacity +Self-cleaning capacity +Compact +Low price of filter medium (sand) −High energy consumption	Second-generation biofilter three-phase FSB can also perform denitrification

Table 1: Continued.

Technology	Removed contaminants	Efficiency	Disadvantages/challenges	Comments
Moving-bed bioreactors [53]	Ammonia (NH_4-N), BOD_5	110.9 g TAN/m^3 per day	+Low cost +Continuous operation +Self-cleaning capacity +Easy to design and operate	Second-generation biofilter
Fixed-bed biofilm reactors	Ammonia (NH_4-N), BOD_5, COD, TP, TAN		−Easy to plug −Needs regular backwash	Has been found to be more robust and protect nitrifying bacteria from the effect of disinfectant
Trickling filters [60]	Ammonia (NH_4-N), CO_2	90 g TAN/m^3 per day removed	+Low cost +High durability −Large units −Expensive biofilter media	Older solutions
Rotating biological contactors [60]	Ammonia (NH_4-N), CO_2,	76 g TAN/m^3 per day removed	+Simplicity of operation +Self-cleaning capacity −High capital cost −Mechanical instability	Older solutions, widely used in domestic wastewater treatment
Bead biofilters	Ammonia (NH_4-N) solids	1.2 kg TAN/m^3 per day removed	+Not easily blocked −High energy consumption	Combine physical filtration with biofiltration

Abbreviations: FF, foam fractionators; PFS, parabolic screen filters; RAS, recirculating aquaculture systems; TAN, total ammonia nitrogen; TN, total nitrogen; TP, total phosphorus.

larger than 20 µm. MDF, due to their small footprint, simple operation, and strong processing capacity are most used in RAS [62].

The mechanical drum filters play a crucial role in achieving optimized water treatment for the removal of solid waste in RAS. The selection should be based on the microscreen rating (µm), the particle size distribution of the suspended solids in the RAS water, and the required water quality. Also, the character and density of particulate matter are important but not well-defined [72]. Drum filters in RAS are typically installed enclosed, channel-mounted, and fully or partially submerged. All have in common that the influent water passes radially through a microscreen, typically a 60–200 micron-screen [73]. In principle, suspended solids larger than the mesh size are retained and accumulate inside the screen, thereby obstructing the water flow. The increase in resistance to the water flow leads to an increase in the level of influent water inside the drum until a maximum tolerable level is reached then, the filter must be backwashed [74].

Fossmark et al. [66] proposed the application of membrane filtration to 10%–15% of total water flow. This improved water quality by lowering turbidity, POC, and slightly TAN. However, even more significant effects were observed in the microbial community of RAS with membrane filtration when compared with conventional filtration. The microbial diversity has been mainly increased, and lower and shorter bacterial blooms and generally lower bacterial densities were observed.

Removing particles smaller than 20 µm requires using foam fractioning in protein skimmers, but those are not as effective in freshwater [66].

Another possibility for improved removal of is the application of chemicals or other methods leading to coagulation of smaller particles and their better removal by MDF. Xu et al. [62] showed that the application of electrocoagulation increased TSS removal efficiency by MDF by 24%. The positive effects on RAS water quality included also enhanced nitrification and COD removal. Combined ozonation with foam fractioning in freshwater significantly improved water quality in RAS and reached an 89% reduction in particle numbers [75].

2.2.3.2. Biological Filtration. The aim of biological filtration is the removal of nitrogenous compounds in the form of ammonia (NH_4–N), nitrite (NO_2–N), and nitrate (NO_3–N). While nitrifying bacteria oxidize the first two nitrogen forms in the biofilter, the latter must be actively removed in systems with low to zero water change to avoid accumulation over time.

From a biotechnological perspective, nitrate can be removed from the aqueous environment either by denitrification and anaerobic ammonium oxidation ($NO_2^- \rightarrow NO \rightarrow N_2$) or by assimilation in bacteria or plant biomass ($NO_3^- \rightarrow NO_2^- \rightarrow NH_3 \rightarrow$ atmospheric nitrogen). The following section addresses the challenges and opportunities associated with nitrate removal processes in intensive RAS. Despite the large-scale application within the field of wastewater treatment over the past decades, denitrification in closed RAS is still in its infancy, as recently reviewed by Preena, Kumar, and Singh [76]. The denitrification process refers to the sequential microbial reduction of nitrate to atmospheric nitrogen ($NO_3^- \rightarrow NO_2^- \rightarrow NO \rightarrow N_2O \rightarrow N_2$). Autotrophic and heterotrophic microbes can facilitate denitrification, whereas the latter group has been identified to support the most cost-effective method for treating nitrate pollution [24]. Thus, heterotrophic denitrification is commonly applied to control nitrate levels in intensive RAS. This microbial process utilizes nitrate as an electron acceptor and requires a carbon source as an electron donor to sequentially reduce nitrate to molecular dinitrogen. In RAS farms, methanol is often used as an external carbon source, even though it is highly flammable and toxic at higher concentrations. Alternatively, acetic acid may also be used, yielding a higher denitrification rate [77]. Recent investigations also explore the possibility of using organic fish waste as an internal carbon source for denitrification [78].

From a microbial perspective, the denitrifying biofilter is an important hub for heterotrophic microbes, but it has not received the same attention as the nitrifying biofilter. Besides affecting the removal rate, the external carbon source regulates the microbial community structure of denitrifying biofilters [79]. Previously, studies have identified the denitrifying biofilter as a potential hotspot for off-flavor-producing bacteria, releasing GSM during cleaning [80]. In contrast, another study suggested that denitrifying bacteria may play a role in GSM and/or MIB degradation under anoxic conditions [81]. Therefore, operating the denitrifying biofilter the right way is of utmost importance. Proper microbial management will control the off-flavor occurrence and prevent the microbial community from switching to an undesired pathway, such as the dissimilatory nitrate reduction to ammonia (DNRA). This nitrate reduction process competes directly with denitrification, resulting in ammonia instead of N_2 as an end-product. The environmental factors regulating the balance between denitrification, DNRA, and other pathways of nitrate uptake in aquatic systems are not very well understood [82]. However, recent work indicates that besides carbon concentration, carbon composition influences the product of nitrate respiration [83]. Besides the above-mentioned challenges, the employment of denitrification biofilters can result in the formation of toxic compounds like nitrite and nitric oxide due to incomplete denitrification or the excessive use of carbon sources. Therefore, it is essential to carefully control the amount of carbon source added to the denitrification biofilter. This ensures that nitrate and nitrite concentrations remain below toxic levels for fish, comply with local effluent regulations, and minimize costs associated with wasted organic carbon sources [84]. This illustrates that careful consideration of system design, HRT/flow rate, carbon source addition, maintenance, and integration with existing water treatment modules, as well as thorough monitoring and control of the process, is critical to ensure effective operation.

2.2.3.3. Disinfection. Disinfection methods are important to the health, growth, and quality of farmed fish in RAS. In contrast to the traditional water disinfection in water supply systems, there is no need for a lasting disinfection effect like in the case of chlorination. The challenging aspect of the disinfection method is eliminating pathogenic microorganisms in RAS water, with no generation of disinfection byproducts or residual oxidation potential, which are harmful to the cultivated fish. The primary selection in RAS is between two disinfection methods, ozonation and UV irradiation, and sometimes their combination. There are two locations in RAS where disinfection methods should be applied: water intake and within the system, usually before biofilters. In the former case, disinfection is used to kill or inactivate pathogens before entry into RAS, hence increasing the biosecurity of the system. The latter case aims to control the microbiota in the system [68].

Ozonation of rearing water (loop water) has both positive and negative effects on fish health; this strongly depends on the fish species, growth phase, and the HRT of the system [68]. Aside from biocidal action, ozonation can reduce organic content and enhance biofilter nitrification rates [65]. Application of ozone into RAS can, unfortunately, cause the formation of toxic and carcinogenic byproducts, that is, hypobromous acid and bromamines, especially in marine water where the concentration of bromide (Br^-) is on average 65 mg/L [85, 86]. Additionally, as ozone readily reacts with iodine, as a result, it lowers its bioavailability and causes goiter in fish [85]. Hence, there is a need to optimize ozone dose concerning bromide concentration in RAS water. In some cases, UV irradiation or activated carbon column is advised as the solution for residual ozone removal to ensure that biofilter and fish are not exposed to high ORP.

Disinfection of water performed with UVC light (wavelength 200–280 nm), usually ~254 nm, has the great advantage of killing or inactivating pathogenic microorganisms without influencing water chemistry. It has been proven that as little as 10 mJ/cm^2 is needed to eliminate 99.9% of *Yersinia ruckeri*, *Moritella viscosa*, *Tenacibaculum* spp., and infectious salmon anemia virus (ISAV) (NoFirm website 31.03.2022). Therefore, the operational costs can be decreased as less energy is consumed and fewer lamps are needed.

Each component (water treatment unit) of RAS must be designed to work in conjunction with other components. More knowledge is continuously gained around the effects that different water treatment technologies have on the fish health and microbial community in RAS [66, 68].

Unfortunately, conventional RAS treatment methods are not sufficient to remove low concentrations of GSM and MIB present in RAS water, and some might contribute to the formation of these compounds, as mentioned before [81]. Currently, off-flavors are removed by purging the fish with clean water, extending the production time by days or weeks, increasing water consumption, and leading to loss of quality and weight by fish [60]. In turn, this weight loss from purging leads to significant financial losses [57]. Even though GSM and MIB have been proven to be removed by activated carbon with 96% efficiency at the low initial levels in drinking water systems, such effects cannot be expected in RAS water due to relatively high DOC concentration (>10 mg/L) [81]. This challenge, together with the removal of micropollutants, is addressed in Section 2.2.4.

2.2.3.4. Heavy Metals. Heavy metals can have a significant impact on water quality in RAS, and with increasing recirculation of water, these contaminants can build up over time to potentially harmful levels if not properly managed. Metal in RAS originate typically from feed, pipe and fitting corrosion, or contaminated make-up water [87]. Trace amounts of elements like iron (Fe), zinc (Zn), copper (Cu), cobalt (Co), manganese (Mn), nickel (Ni), and selenium (Se) are added to fish feed because they are vital for normal fish metabolism. These elements can enter the RAS water through fish excretion, fecal leaching, or the release of uneaten feed [88]. The increasing accumulation of dissolved heavy metals poses potential risks to the growth and development of fish species, and certain metals, such as copper (Cu) and nickel (Ni), are known to be toxic to humans, while others, including arsenic (As), cadmium (Cd), and lead (Pb), are classified as carcinogenic [89]. Notably, the concentrations of these metals found in the muscle tissue of farmed fish—measured at 0.98 ± 0.95 µg/g dry weight for Cu, 0.014 ± 0.022 µg/g dry weight for Cd, and 0.120 ± 0.128 µg/g dry weight for Pb—are considerably below the established safety thresholds for human consumption, which range from 50 to 150 µg/g dry weight for Cu, 0.25 to 10 µg/g dry weight for Cd, and 2.5 to 30 µg/g dry weight for Pb [89, 90]. This indicates that while the presence of heavy metals in aquaculture systems warrants attention, the current levels detected in farmed fish do not exceed regulatory limits for safe human consumption. Methods for controlling heavy metals in RAS include processes such as adsorption, membrane, chemical, electrical, and photocatalytic treatments. The effectiveness of these methods relies on the specific characteristics of the wastewater source [91]. However, the complex biochemical composition of RAS wastewater, characterized by elevated levels of particulate matter, nitrogen, and phosphorus, makes it difficult to identify one superior method suitable for all pollutants and typically necessitates the implementation of integrated treatment methodologies to effectively remove heavy metals. These methodologies often involve a combination of processes, such as precipitation and membrane filtration, as well as denitrification coupled with slow sand filtration [88–92].

2.2.4. Emerging Technologies. The emerging technologies in the RAS industry are focused on providing better solutions to water quality problems, optimizing existing technologies for the specifics of RAS, and developing new solutions. One of the examples of sustainable thinking is the focus on converting waste to product, for example, nutrients into feed. Another focus of emerging technologies is the removal of micropollutants. In the case of RAS, one of the biggest challenges is the removal of T&O compounds. However, other CECs need to be considered regardless of their origin in the system. All the methods need to be tested in a pilot scale, under the operating conditions optimal for the cultivated fish species for an extended period to determine the long-term effects on fish health and growth as well as the operation of other water treatment units and microbial community both in RAS water and biofilters. For example, a surplus of ozone in the protein skimmer could change the microbial community of the biofilter, hence impairing its proper operation.

2.2.4.1. Advanced Oxidation Processes (AOPs) and Electrochemical Methods (EC). In this section, we focus on applying AOPs and EC in RAS to address one of the challenges RAS faces, that is, controlling T&O compounds and micropollutants. AOPs are processes in which hydroxyl radicals ($^{\bullet}OH$) are formed. The newer definition also includes sulfate radical ($SO_4^{\bullet-}$) [71]. Both are more potent oxidative agents ($^{\bullet}OH$ −2.8 V and $SO_4^{\bullet-}$−2.6 V) than ozone (2.08 V). Reactions of organic compounds with $^{\bullet}OH$ are nonselective, unlike their reactions with ozone. Numerous AOPs have been developed for water and wastewater treatment since the formation of reactive species (mainly $^{\bullet}OH$) can be obtained through various physical (UV irradiation, nonthermal plasma, etc.) and chemical (addition of, e.g., H_2O_2 and/or heterogeneous catalysts like TiO_2) processes. EC water treatment uses applied electrical current to remove contaminants through oxidation on the anode and/or reduction on the cathode.

The efficiency of removing T&O compounds, hormones, and pharmaceuticals by means of different AOPs has been demonstrated [93, 94].

Ozone, besides being useful as a disinfection agent, is also used in some AOPs, alone or in combination with H_2O_2, UV, ultrasounds, or a catalyst. Ozonation is included in AOPs when $^{\bullet}OH$ is formed (e.g., at pH >4) (so-called indirect ozonation). Direct ozonation has been proven less effective in removing various micropollutants [95] as ozone is very selective and attacks electron-rich groups [93]. When applied alone, direct ozonation is also not as effective toward T&O compounds removal as other AOPs, most likely due to their saturated cycled tertiary alcohol structure [96]. Hence, combined methods need to be used. Klausen and Grønborg [97] compared the effectiveness of MIB and GSM removal in RAS water with ozone in combination with UV and UV/H_2O_2 for increased $^{\bullet}OH$ formation. Higher rate constants were observed for the UV/H_2O_2 system (GSM 1.2 h^{-1}, MIB 1.5 h^{-1}). However, both combined methods showed increased electrical energy per order (EEO) required. Furthermore, the application of H_2O_2 can pose an additional threat to fish health if not fully consumed during the process. The required H_2O_2 dose can be lowered by utilizing pretreatment that would minimize the effects of the water matrix on process efficiency. Furthermore, similarly to ozone, residual H_2O_2 can be quenched with granular activated carbon [98].

Kye et al. [86] studied the removal of florfenicol (FF) and oxolinic acid (OA), two antibiotics used in the aquaculture industry, with the use of ozonation. FF was more resistant to ozone action than OA, of which 60% was removed within 15 s of ozonation. In RAS disinfection, ozonation is typically followed by UV irradiation to decompose residual ozone; in AOPs, ozonation and UV are applied in the same reactor.

When UVC irradiation is used as an AOP, the UV fluence is >200 mJ/cm^2; this exceeds the UV dose required for the 4-log inactivation of most pathogens [71]. Recently, it has been proven that a combination of UV/VUV (254 nm + 185 nm) is more effective than UVC alone in the elimination of PhACs [99] and GSM and MIB [100]. However, in RAS systems where nitrates are present even at low concentrations (~10 mg/L), the formation of 0.6 mg/L of toxic nitrite can be observed under UV/VUV irradiation [100].

Less research can be found concerning aquaculture water and even fewer on pilot scale dimensions, with the evaluation of AOPs application on fish health and growth. Davidson et al. [101] studied the effect of ozonation on the waterborne hormones in RAS, as steroid hormones were proven to accumulate in RAS. Their study showed that not only ozonation at the level of ORP of 300–320 mV was effective in the reduction of waterborne hormone levels but also water quality parameters like true color, total heterotrophic bacteria count (THBC), UV transmittance (UVT), copper, iron, and zinc were improved.

Pestana et al. [102] developed a continuous flow-packed bed photocatalytic reactor for the removal of GSM and MIB in a RAS system. 90% of MIB and GSM were removed by the TiO_2 pellets from the water of the fish farm with initial concentrations of 19 and 14 ng/L, respectively. In the face of the new legislation regarding the use of TiO_2 as a food additive (https://efsa.onlinelibrary.wiley.com/doi/epdf/10.2903/j.efsa.2022.7666), the application of TiO_2 in the RAS system should be limited to reactors with immobilized TiO_2 and water should be tested for the potential release of TiO_2 nanoparticles into RAS.

Ben-Asher et al. [103] showed efficient elimination of GSM and MIB in pilot-scale RAS from cold-water RAS-grown fish. The novel approach combined the effective elimination of GSM and MIB by electrooxidation with a new concept based on the depuration of the fish during the last stage of the normal growth period while maintaining regular feeding. The RAS system was detached from the biofilter during this time. Both the ammonium oxidation and off-flavors removal occurred simultaneously in electrolyzer. Off-flavors were removed from fish flesh, below the T&O threshold, already for 7 days, while TAN stayed below 12.1 mgN/L during the depuration experiment.

2.2.4.2. Application of Microalgae. In conventional RAS systems, usually, three main units are included for removing solid particles (fish feces and feed residual), for nitrification, and a reservoir for water conditioning (e.g., temperature adjustment, oxygenation, and disinfection) [104]. Due to the nitrifying biofilter of a conventional RAS, nitrate-nitrogen (NO_3-N) can accumulate up to 400–500 mg/L, having detrimental effects on the growth and health of farmed organisms. Therefore, a denitrification unit is embedded into modern RAS to avoid nitrate accumulation by transforming nitrate into N_2 gas [105] and to avoid the discharge of N components. Eutrophication poses a serious environmental problem, and stringent discharge regulations of N, P, COD, and solids have come into place in Europe. On the other hand, also the gaseous N loss as N_2O emission and NH_3 volatilization is another big problem due to the negative effect of global climate change. Despite technological progress toward improving the physicochemical quality of recirculated water in RAS, aquaculture wastewater treatment remains expensive, while valuable nutrients in wastewater are discarded. Therefore, in addition to removing nutrients, their recovery should also be considered in developing more sustainable and economically feasible technologies for RAS wastewater treatment. In this respect, bioremediation, in which live organisms are used to purify wastewater, is considered a promising alternative to traditional methods. Among tested organisms, microalgae appeared as an efficient biotechnological platform to produce quality biomass and high-value products in specific photo-bioreactors operated with RAS side streams (e.g., process water). In addition, microalgae are part of the natural diet of many farmed-raised species, such as fish, mollusks, crustaceans, and larvae [106]. Thus, microalgae could bring multiple advantages to RAS enterprises by integrating RAS wastewater purification, nutrient recovery, and aquafeed production. This section reviews the main advantages of using microalgae to treat RAS wastewater.

2.2.4.2.1. Removal and Recovery of Nutrients. One of the prominent features of microalgae for the treatment of RAS effluents is their ability to remove multiple nutrients from wastewater. Compared to nitrification-denitrification-based technologies that focus on removing nitrogen compounds from RAS wastewater, microalgae can simultaneously remove several nutrients from the water. Indeed, the continuous growth of microalgae requires the supplementation of macronutrients (e.g., C, N, P, K, Na, O, H, S, Mg, and Ca) and micronutrients (e.g., Fe, Zn, Cl, Cu, Mn, V, Mo, B, Co, and Si) [107]. Therefore, sustainably, RAS effluents can supply nitrogen and phosphorous compounds for microalgal growth, and microalgal cells can purify wastewater. For example, Tejido-Nunez et al. [108] tested *Chlorella vulgaris* and *Tetradesmus obliquus* species to remove nitrate and phosphate from a fish tank with initial concentrations of 96.3 and 9.9 mg/L, respectively. They reported that microalgae could remove 98% of nitrate and 99% of phosphate from RAS wastewater at a laboratory scale. Also, a high nitrate-removal efficiency of 98.6% was observed in the pilot-scale cultivation of microalgae. The treated water can be recycled into the aquaculture system or safely discharged into the environment.

Another advantage of RAS wastewater bioremediation using microalgae is the simultaneous removal and recovery of nutrients from wastewater. As compared to conventional methods, microalgae not only remove nutrients from RAS wastewater but also recover them as microalgal biomass. For instance, in a study conducted by Li et al. [109], more than 87% of nitrate and 91% of ammonia were removed from the wastewater of a shrimp farm by *Scenedesmus acuminatus*, and 1.22 g/L dry microalgal biomass was also produced within 8 days cultivation.

2.2.4.2.2. Microalgae as Aquafeed. Low-cost aquafeed and good-quality water are two critical needs in an intensive aquaculture system. Supplying fish feed is important for farmers as feeding is a continuous process from a few days after the hatching of eggs until the day before selling their products. Nutrients found in aquaculture effluent are ideal substrates for the cultivation of microalgae and biomass production [110]. Microalgae can take up these nutrients for the biosynthesis of high-quality protein (essential amino acids), fatty acids (long-chain polyunsaturated), polysaccharides, bioactive compounds, minerals, vitamins, and pigments [111].

Additionally, microalgal biomass, produced in RAS wastewater, can be utilized as aquafeed directly (for feeding of larvae or as feed supplement) or indirectly (through intermediate). Cultivation of microalgae in aquaculture hatcheries of finfish and shellfish as live aquafeed has been practiced for a long time. In 2010, Allen and Nelson [112] published the first report about the application of microalgae as feed in aquaculture entitled. In traditional extensive aquaculture, microalgae bloom in cultivation tanks or ponds. By contrast, in hatcheries of advanced intensive aquaculture, usually single species of microalgae are produced in photobioreactors [113]. Moreover, microalgal cells could also be used by intermediate zooplankton like rotifers, which are used as live feed for larvae of many fish species. In addition to microalgae as a live feed, other microalgal-based products such as dried whole cells, defatted cells (after lipid extraction), pigments, and fatty acids have also been utilized as fish feed ingredients [114]. Therefore, as a green technology, microalgal cells can convert waste nutrients in RAS effluent into valuable biomass. Consequently, nutritionally enhanced microalgal-based aquafeed produced using aquaculture waste effectively closes the value chain loop, creating a more efficient, sustainable, and profitable modern RAS aquaculture.

3. Modern RAS and Microbial Management

When accurately designed, RAS support optimal conditions for fish growth and welfare. As described in detail in Sections 2.8 and 2.10, good system management, mechanical and biological filtration, and physical disinfection methods such as UV light and ozone reduce potential pathogen load in the water. On the other hand, suboptimal RAS design and failure in biosecurity systems might lead to the proliferation of opportunistic pathogens and disease outbreaks [115–117]. The following chapters will discuss common pathogens detected in RAS systems and the advancements in biosecurity and disease control.

3.1. Common Pathogens in RAS. RAS can harbor pathogens belonging to different groups, such as bacteria, parasites, fungi, and viruses [10, 118]. Over the years, several studies have identified common potential pathogens present in RAS farming different fish species.

Typical pathogens in rainbow trout (*Oncorhynchus mykiss*) and salmon (*Salmo salar*) RAS include bacteria (*Flavobacterium*, *Aeromonas*, *Renibacterium*), parasites (*Gymdactylus*, *Chilodonrlla*, *Trichodina*, *Epistylis*, *Trichophrya*, *Ichrhyoprhirius*, *Ichryobodo*, *Coleps*), fungi (*Saprolegnia*), and viruses (IPN, VHS, and IHN virus) [119]. Deep-sequencing of bioreactor's biofilm and water microbiota, during commercial-scale Atlantic salmon post-smolt production, allowed the detection of highly diverse microbial communities [120]. Despite no reported disease outbreak, such communities included potential pathogens from the general *Flavobacterium*, *Polaribacter*, *Pseudoalteromonas*, and *Photobacterium*. In European perch (*Perca fluviatilis*), *Aeromonas hydrophila* and *A. salmonicida* were the most common pathogens detected, associated with deep skin ulcerations and muscle fiber necrosis [121]. *A. salmonicida* caused systemic disease in European perch RAS at a temperature around 20°C with mortality up to 3% per week [122]. *Flavobacterium columnare* was also isolated from gills and skin in European perch RAS, but no systemic disease was detected [122]. Florida pompano (*Trachinotus carolinus*) cultured in RAS systems showed granulomatous lesions caused by *Mycobacterium marinum* [123].

Reduced microbial biodiversity could be an index of pathogenicity in RAS, as an increased abundance of a pathogen might impair the growth of other bacteria. Disease-free yellow grouper (*Epinephelus awoara*) RAS showed a more diverse bacterial community in water than disease-prone RAS [124]. The former was composed of different bacterial orders, including Vibrionales (35.8%), Alteromonadales (17.3%), Rhodobacterales (10.7%), Kordiimonadales (7.43%), and Oceanospirillales (6.26%), while the latter by mainly Vibrionales (50.5%) and Flavobacteriales (36.5%). The presence of potential pathogens such as *Vibrio harveyi* and *Vibrio rotiferanus* was significantly higher in diseased RAS.

Indeed, pathogens are commonly present in RAS and, if not properly controlled, can lead to disease outbreaks. These challenges have driven significant advancements in disease control strategies through a variety of approaches, including chemical, physical, and biological methods.

3.2. Advancements in Disease Control. Using therapeutics to control a disease outbreak is a challenge in RAS, as it might disturb the beneficial bacterial communities in the biofilter [10, 118] or on the fish [125]. Recent studies have focused on developing treatment protocols using minimal doses of drugs that are efficacious against pathogens while not damaging microbial communities [126–130].

Bacteriophage-based treatments are emerging in aquaculture as a valid alternative, or a complement, to therapeutics [131]. The mechanisms by which phages cause pathogenic bacteriolysis are well understood and follow a specific sequence of steps, beginning with phage infection. Typically, phages are highly specific and can target bacterial pathogens at the species or even strain level. However, in some cases, phages capable of infecting multiple bacterial species or genera have been identified, and these are referred to as polyvalent phages [132]. While phage therapy was first used in fish in 1981 [133, 134], it is only recently that specific applications for RAS have been explored. The applicability of phage therapy in RAS has been tested with Rainbow trout (*O. mykiss*) cultures using a *F. columnare* - infecting phage [135]. This study demonstrated the persistence of the phage for up to 3 weeks in the system, suggesting phages as a potential biosecurity strategy in RAS. The water recirculation process, typical of RAS, offers a significant advantage for phage delivery, as phages are small enough to pass through filters and other barriers, allowing them to remain in the system for extended periods. This continuous circulation in the water could provide prolonged protection by extending the exposure time to the phages. Additionally, it may promote phage evolution in response to bacteria that develop resistance, a phenomenon observed in open aquaculture systems [136]. Indeed, this needs further research and validation. Additionally, more investigation is also needed on the sustainability of a commercial application, formulation for layman's use, and protocol development for different diseases.

Probiotic treatments can be administrated in RAS systems via feed or water to strengthen the immune system of farmed fish, reduce the activity of fish pathogens, and improve water quality [118, 137]. For instance, waterborne probiotic delivery decreases mortality and ulcer development in Atlantic salmon RAS after just a single application [138]. Dietary probiotic supplementation from larvae to fry stage in Nile tilapia (*Oreochromis niloticus*) increased the relative abundance of beneficial Bacillus in the fish gut microbiome, potentially increasing fish health and welfare [139].

4. Advancements Around the Globe (Practical Applications and Selected Case Studies)

Recirculating systems are encountered throughout Europe mostly for fingerling production of both freshwater and marine species, as well as for on-growing of freshwater species such as eel, trout, catfish, and sturgeon, and marine species such as turbot, sole, and seabass. Hatchery production is optimized in a RAS system because it is cheaper to heat or cool the water to an optimal temperature and thereby produce fish around the year without being limited by the natural spawning season [140]. In France, for example, the use of RAS systems in pre-on growing units allows transfer to the sea of larger juveniles and thereby results in better exploitation of sea cages [115]. The two leading countries in terms of production volume in Europe are Denmark and the Netherlands. In Denmark, outdoor RAS systems are used to produce trout, whereas in the Netherlands, indoor RAS systems are used to produce eel and African catfish [26].

Innovative RAS facilities range from large-scale systems, for example, Atlantic Sapphire in Miami (USA) (https://atlanticsapphire.com/innovation/), to comparatively tiny distributed closed RAS, designed to fit in a parking spot, as by ARK Inc (Japan) (RAS in a box—RASTECH Magazine). Also, different designs are being developed, such as optiRAS in Norway, where the nitrifying biofilter is located along the side walls of the round fish tank (Første enhet av helt ny type RAS installert [landbasedaq.no]). As an alternative to biofiltration, both Eloxiras and Biofishency (ELX-NEW.pdf [biofishency.com]) have recently developed a water treatment module for brackish and marine RAS based on electrochemical oxidation [141]. This technology can remove ammonia, while simultaneous disinfection has been observed [142]. However, unlike classic biofiltration, the incoming water first needs to be pretreated by filtration steps and passing a foam fractionator to remove solids, oil and fats. Furthermore, this treatment technique efficiently decomposes the off-flavors MIB and GSM in fish while feeding and maintaining fish growth (https://aquaculturemag.com/2023/03/17/biofishency-hails-first-in-the-world-success-in-electro-chemical-removal-of-off-flavors-in-recirculated-aquaculture-systems/). In terms of sustainability, several projects are planning to integrate RAS and aquaponics in the near future. For example, EcuNor Aqua plans to combine aquaculture production with water-based plant production to minimize environmental footprint and resource utilization. Also located in Norway, Columbi Farms aims at circular food production by utilizing nutrient-rich water from fish tanks to produce 4000 tonnes of leafy greens annually, while the solid waste from the fish tanks will be converted into biogas for energy generation and fuel for transporting the fish to market. The company is planning sustainable vegetable production at a large scale and will become Norway's largest vertical farm by 2024. Located further in the south in Europe, the integration of RAS with microalgae and vegetable production has recently been demonstrated in Portugal by BGI, a business accelerator dedicated to promoting sustainable food production (www.bgi.pt/ampliaqua). AmpliAqua's multitrophic system minimizes greenhouse gas (GHG) emissions through algae and plant capture and is designed as a modular, replicable solution for global deployment, including resource-limited areas.

In the future, integrating innovative technologies will be pivotal in establishing a more sustainable and resilient food system. This involves developing symbiotic ecosystems that use less land and water while contributing to CO_2 mitigation.

5. How Can Advancements in RAS Help With Climate Change?

Aquaculture and climate change have intensifying effects on each other. Aquaculture activities directly contribute to releasing GHGs. Consequently, vulnerable effects of climate change-related phenomena affect aquaculture industries. Climate-induced vulnerability is different depending on the type of aquaculture system, farm-raised species, geographical areas, and climatic zones. In this respect, coastal areas are more exposed to risks of climatic hazards such as cyclones, salinity fluctuation, rainfall variation, and sea level rise than those located inland [143]. Among various types of coastal and inland aquaculture systems, RAS is minimally affected by phenomena related to global warming due to operation in an indoor and controlled environment. RAS systems with good adaptation strategies to climatic variables can be constructed in various climatic zones and geographical areas, from subpolar and temperate regions to tropical, desert, and arid regions [11].

Aquaculture is envisaged as a promising solution to the increasing global food demand. It carries a lower environmental impact than many other animal protein productions (5–6 tonnes CO_2-eq per vs. 27–34 tonnes CO_2-eq per ton of edible beef) [144]. Sustainable aquaculture, conversely, can adapt to and decrease the effect of climate change. Among many available aquaculture solutions, RAS has a high potential for sustainable seafood production [12]. RAS provides an adaptation of food production to climate change due to decreased water requirements, intense and resilient production, and more localized supply chains. Higher electricity consumption by RAS increases its carbon footprint and has more detrimental effects on climate change. This is relevant when fossil fuels are used to generate the required electricity. However, this can be minimized by implementing renewable energies, for example, produced by photovoltaic technology (https://www.leroyseafood.com/en/tasty-seafood/environment-and-society/solar-power-is-reducing-the-energy-footprint-of-fish/; Infinite Sea's power play—RASTECH Magazine RASTECH Magazine). For instance, supplying renewable energy (hydropower electricity) to a RAS system in the USA reduced carbon footprint from 7.01 to 3.39 kg CO_2eq/kg live-weight salmon [145]. Also, the construction of RAS systems near the target markets can significantly reduce GHG emissions. In addition to using renewable energy sources, enhancing building insulation, optimizing water treatment systems, and increasing byproduct utilization have been suggested as effective strategies for reducing the carbon footprint and enhancing sustainability in fish production within RAS [146]. The implementation of renewable energy sources has the potential to be utilized across various scales, including small-scale systems, for example using solar and wind-powered equipment [147].

When discussing the impact of climate change, another important factor to consider is the land use change, which can increase carbon footprint due to disruption of ecosystem carbon sinks, for example, the conversion of mangroves to shrimp farms led to the release of 190 tonnes CO_2-eq per ha [11]. RAS systems do not require vast land areas, and their location can be selected; therefore, they can minimize harm to ecosystem services. Conversion of mangroves and paddy fields to shrimp and fish farms decreases carbon capture and intensifies the effects of climate change.

In addition to direct power consumption and land use change, aquaculture is also associated with GHG emissions due to the production of aquafeeds. Transportation of feed ingredients to factories, processing of raw materials to compounded feed, delivery of produced aquafeeds to farms, and aquafeed storage consume a considerable amount of energy and contribute to GHG emissions [148]. Refrigerated transportation is responsible for the higher carbon footprint of transportation and it is increased by a high-temperature difference between the outside and inside of the insulated truck body and refrigerant leakage. Liu et al. [145] reported that the highest climate impact on aquaculture is associated with feed production and transportation. Feed carbon footprint was higher than 1 ton CO_2-eq per ton of salmon produced both for RAS and open-water Atlantic salmon farming [145]. However, in RAS, the obtained value was lower by 180 kg CO_2-eq per ton of produced fish due to higher feed efficiency and lower FCR. These issues need to be considered in the implementation of RAS systems and the production of seafood in a sustainable way.

6. Advancements in RAS for Different Fish Species in Europe

Selecting the right species for cultivation in RAS is very important as the technology requires high capital and operating expenditures. One important selection criterion is the required water temperature, as reaching and maintaining optimum temperature affects the OPEX but also results in the highest growth rate. Besides this, the suitability for rearing a certain species in RAS also includes many other factors, such as profitability, environmental concerns, and biological suitability [149]. The here presented finfish species are well-established and emerging aquaculture species and are commercially produced in RAS which have been part of the Blue-Bioeconomy project DIGIRAS (http://www.digiras.org/).

6.1. Atlantic Salmon. Atlantic salmon (*S. salar* L.), commonly referred to as salmon, is an esteemed and economically significant fish species worldwide. It holds a prominent position as a highly nutritious food source, extensively cultivated for human consumption. The aquaculture industry dedicated to salmon production generates an estimated annual revenue of ~8.5 billion GBP (equivalent to 9.7 billion Euro) (FAO 2017), making a substantial contribution to the food supply, economic stability, and employment opportunities of numerous nations. The farming of Atlantic salmon in sea cages was introduced in Norway in the early 1970s and quickly expanded to other countries in the North Atlantic, Pacific Canada, Chile, and Australia. In 2020, global production of farmed Atlantic salmon reached 2.7 million metric tonnes, making up 71.4% of total salmon production. This was a significant increase from 2000, when farmed Atlantic salmon made up 47.3% of total production. The largest producer of farmed Atlantic salmon is Norway, accounting for over half of the total production at 1,232,200 metric tonnes. Other significant producers include Chile, the United Kingdom, Canada, and the Faroe Islands, with these five countries making up 90% of the global production of farmed Atlantic salmon [150].

Today's commercial salmon production is dominated by the classical two-step process where the juvenile fish are produced in land-based aquaculture systems (LBAS) with freshwater or brackish water with low salinity from hatching to smoltification and then are transferred for grow-out to market-size salmon (MSS) in sea-based net pens, giving an environment that cannot be controlled. LBAS for smolt production are principally either conducted in RAS or flow-through systems (FTS). Despite intensive research in the past years, it is still controversially discussed which of those two technologies is more beneficial for producing robust and healthy smolt, optimally prepared to meet challenges in open sea cages after transfer to sea, and how the robustness of (post) smolt can be positively affected. In recent years, conducting the entire production cycle of Atlantic Salmon in RAS has become an attractive alternative for the aquaculture industry. For example, Atlantic Sapphire is aiming at an annual production of about 200,000 metric tonnes of Atlantic salmon in RAS in the USA. However, the company experienced massive setbacks due to technical, biological, and economic challenges, and in early 2024, only 1150 tonnes market size fish was harvested (https://atlanticsapphire.com/). Another example is Nordic Aqua Partners, a Norwegian company that is producing market size Atlantic salmon in a RAS facility in Ningbo, China. The company recently reported the first harvest of 190 metric tonnes of head-on-gutted fish after experiencing off-flavor challenges [151]. Despite advancements in RAS technology, these examples highlight the challenges of producing market-size fish exclusively in RAS on an industrial scale, underscoring the need for further improvements. However, these companies are also paving the way for potentially more sustainable production of fresh Atlantic salmon near consumer markets, thereby reducing carbon emissions and transportation costs.

6.2. European Sea Bass. The European Sea bass belongs to the family Moronidae of teleost fishes [152, 153]. This marine species is euryhaline (0–40 ppt salinity) and eurythermal (2–32°C) and plays an important role in Europe, both for cultural and economic reasons [154]. The coastal sea bass inhabits shallow waters from the north-eastern Atlantic Ocean to the Mediterranean and the Black Sea, where it is often found in estuaries and lagoons from spring to fall. During winter, the sea bass migrates from the coastline to deeper waters with more stable temperatures above 9−10°C. However, since 1992, aquaculture has been producing more Sea bass than fisheries, and in recent years, the production has seen significant growth. In 2021, European sea bass production reached 305,000 metric tonnes, and 98% of this is derived from aquaculture [155]. The EU Fish Market ranked seabass as the main commercial marine fish species in 2013 and 2014 [156].

Sea bass aquaculture is essentially located in the Mediterranean, as 94% of the production is in Turkey, Greece, Egypt, and Spain. Sea bass aquaculture production is a two-step process: the land-based hatchery-pregrowing phase produces 1–20 g in three to 8 months, followed by an on-growing phase to 250–450 g in 12–20 months [154].

Currently, intensive farming of this species is mainly conducted in FTS, followed by a transfer to sea cages. However, research on cultivating Sea Bass in RAS is ongoing. For example, Li et al. [157] identified that the ORP should be elevated but should not exceed 300 mV. Elevated ORP levels resulted in lower feed intake, slower growth rate and elevated stress, but a better resistance toward bacterial infections.

6.3. Seabream. Gilthead seabream is a euryhaline marine species belonging to the family of Sparidae. It is a protandrous hermaphrodite, so the fish are functional males during the first reproductive seasons (20–30 cm) and thereafter may turn into females in the next seasons (>33 cm). Small-scale production was first succeeded in 1978–1980 in France, Italy, and Israel [158]. Large-scale production took place a few years later, and in 2020, production reached 282,000 tonnes [159]. Production of juveniles is based on intensive marine hatcheries, which often apply partial recirculation of seawater for both reduction of pumping and heating costs, as well as for reasons related to the microbial balance of the water. Gilthead seabream grows best at a temperature range of 18–25°C, a salinity of 5–44 ppt, free ammonia of 0–0.2 ($mg\,N\,L^{-1}$), TAN 0–5 ($mg\,N\,L^{-1}$), and CO_2 0.5–12 $mg\,L^{-1}$) [160].

6.4. Yellowtail Kingfish (Seriola lalandi) (YTC). YTC is a marine pelagic warm water species in the southern hemisphere and the Northern Pacific and is primarily consumed fresh as sushi. Because of its high market value and exemplary performance in land-based RAS, it is an up-and-coming candidate for RAS. Aquaculture production of YTC is relatively recent, and production volumes are small. The production in RAS is established in Chile (est. production in 2018 = 100 t) and the EU (est. production in 2018 = 180 t), specifically in the Netherlands (The Kingfish Company, formerly Kingfish Zeeland), Denmark (Sashimi Royal, Maximus) and Germany (Fresh Völklingen). (https://www.hatcheryinternational.com/yellow-is-the-new-green-3447). While global production is still relatively small, it is growing. As of 2022, production estimates included volumes of ca. 7000 metric tonnes in Japan, Australia and Europe [161]. Recent data on the production in RAS also shows significant growth, particularly reported by the Kingfish Company. The standing biomass at the end of Q2 2024 was reported 1075 tonnes, which is more than doubling from the same quarter in 2023 (Kingfish company, financial report) (https://thekingfishcompany.com/investors/q2-and-h1%E2%88%922024-financial-results/). Yellowtail kingfish is currently considered an emerging species with high potential for production in RAS. Major arguments include the tolerance to high stocking densities (up to 80 kg/m^3), the fast growth rates (up to 2.5 kg within 1 year), and the high market value [161].

6.5. Arctic Charr. Arctic charr (*Salvelinus alpinus*) is the salmonid species with the northernmost geographic distribution [162]. Arctic charr has been farmed since the 1970s, mainly in northern Europe and production has increased slowly but steadily during the last 30 years to reach 6000–10,000 metric tonnes. However, the global production of Arctic charr in aquaculture is still limited, with Iceland as the largest producer (4900 tonnes in 2018) (https://www.statice.is/publications/news-archive/fisheries/aquaculture-in-iceland/). Arctic charr has several characteristics that make it an attractive species for commercial aquaculture production. Compared with other salmonid species, it has a higher cold-water tolerance, faster growth at lower temperatures ranging from 0.5 to 14°C, greater tolerance for higher stocking densities, high fillet yield, and flesh is perceived of high quality [163]. In Sweden, it is typically produced in farms located in large, semi-oligotrophic freshwater lakes in the northern part of the country [164]. In contrast, in Iceland, most of this fish is produced in coastal land-based farms with good access to brackish (15–25 ppt) water at stable temperatures [165]. Such LBAS are either FTS or RAS.

Arctic charr has a long track record of growing well in cold water aquaculture, and it is distributed in specific markets at fairly good prices [149]. There is growing interest in Arctic charr aquaculture due to its suitability for intensive farming and high market value. The species can be grown at higher densities than salmon or trout, making it well-suited for RAS production. Currently, Sapphire Springs Inc. (Canada) is building the largest land-based aquaculture facility of Arctic charr to produce 5000 metric tonnes using RAS technology [166].

6.6. Rainbow Trout. Rainbow trout (*O. mykiss*) has a relatively long history in Norway, as commercial fish farming started with rainbow trout in the 1950s. In 1977, Norway produced 1795 tonnes of trout, while in 2020, production reached 96,633 tonnes [167]. Global trout production reached roughly 700,000 tonnes in 2014 [168], with a steep increase to 940,000 tonnes in 2019 and rainbow trout as the main farmed species, accounting for 97% of total production [169]. Rainbow trout is easy to culture and widely reared in freshwater RAS, from fry up to plate-sized fish at optimum water temperatures of 16°C [149]. The *O. mykiss* species comprises a land-locked strain and an anadromous strain [170], also called steelhead salmon, due to its life history, which is like salmon [171]. Accordingly, the first stage of the production chain is often conducted in RAS before the fish is transferred for on growing to raceways or ponds with flowing water, or cages in freshwater and marine environments [172]. Thus, larger trout can be grown in fresh or saltwater. The transfer to the sea is practiced in many European countries, with Denmark being one of the largest steelhead producers. Recently, seawater farming of *O. mykiss* has attracted interest in Korea due to the larger harvest sizes in sea cages [173]. However, due to relatively tough competition in most markets, the products need to be diversified to become profitable [149].

7. Summary and Recommendations

Oceans have always been a significant source of food for humans, but climate change, pollution, and overexploitation are threatening the productivity and stability of marine ecosystems. However, advancements in RAS offer a solution to these challenges. RAS is a land-based fish farming method that uses advanced technology to create a controlled environment for fish production. It reduces the environmental impact of aquaculture by minimizing water usage, improving water quality, and reducing the risk of disease outbreaks. RAS also allows to produce seafood in areas with limited water resources and provides opportunities for waste management and nutrient recycling. The use of machine learning technologies, such as computer vision and AI, in RAS systems has also improved fish behavior monitoring, feed optimization, disease detection, and water quality monitoring. To date, there are several AI solutions available on the market, covering the fields of remote monitoring, feeding and growth statistics, monitoring fish behavior, optimizing harvesting time of shrimps, estimating the fish size and weight in real-time, and disease detection [174].

Future applications may also allow us to improve systems efficiency and reduce energy consumption [175].

Additionally, RAS systems have been successfully implemented to produce various fish species, including Atlantic salmon, Arctic charr, rainbow trout, yellowtail kingfish, European seabass, and gilthead seabream. These advancements in RAS systems have the potential to mitigate the challenges posed by climate change and promote sustainable seafood production. Extensive fish mortalities in a RAS system inflicted by bad water quality can cause significant direct economic losses. H_2S-poisoning in a RAS system can lead to significant health and economic damage to both fish and the farmer, respectively.

Several countries have taken political initiatives to promote land-based RAS as a solution for promoting aquaculture production while limiting environmental harm [176].

RAS is a fast-growing industry, expected to reach $700M in 2027 with a CAGR of 15%, and forecasted to represent 30% of all aquaculture production by 2030 (https://www.marketsandmarkets.com/PressReleases/precision-aquaculture.asp), with large growth due to the export of water treatment technology to countries in the Asia-Pacific region. For RAS technology to become sustainable, full control of water quality parameters and the optimization of rearing conditions with the lowest environmental impact are crucial [176]. Given the recent rapid advancements in real-time monitoring techniques and predictive modeling solutions, the future of RAS is happening now. Optimizing land-based fish production based on digital models promises to unlock the full potential of RAS to improve and maintain water quality, thus ensuring supreme fish welfare while minimizing resource usage.

Data Availability Statement

This is a review paper with no primary data, so the data availability statement is not applicable.

Conflicts of Interest

The authors declare no conflicts of interest.

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