

Performance evaluation and efficiency enhancement of water purification system in a semi-recirculated aquaculture system (RAS)

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ABSTRACT

In this study, the status and efficiency of the wastewater treatment system of a semi-recirculated aquaculture system (RAS) in Firoozkoo County were investigated. Information regarding aquaculture systems in the region was obtained with the guidance of the experts at the Agriculture Organization in Firoozkoo County. During visits to the recommended aquaculture systems, Namrud Aquaculture was selected. To evaluate the effect of water pollutants on fish, National Standards 8726 and 7961 were employed to determine water quality parameters and to guide sampling and sample protection. Water quality parameters were assessed at four stations to determine the extent of pollution. The results indicated that ammonia (NH₃), nitrite (NO₂), and nitrate (NO₃) were the main issues in this aquaculture system. Rainbow trout (*Oncorhynchus mykiss*) exposed to ammonia levels exceeding the limits set by National Standard 8726 displayed behavioral changes, including altered swimming speed, total distance traveled, average direction change, and increased average distance from a central point. The second aeration tower was found to be ineffective in releasing dissolved gases due to its inappropriate design, including the spacing between floors and lack of proper atmospheric connection. It is recommended to redesign the second aeration tower, elevate the second entrance pond, and adjust the return flow to the grit chamber to reduce suspended particles and improve the water quality parameters of the effluent.

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1. Introduction

The constraints on freshwater resources and the forecast of a future global water crisis highlight the urgent need for optimal usage, and prioritizing drinking water. Meanwhile, the demand for the aquaculture industry increased significantly in recent decades. Water resource limitations and social pressures regarding the environmental impacts of aquaculture activities will compel breeders to adopt environmentally friendly methods for producing aquatic animals. (Avnimelech and Kochba, 2009) Recirculating aquaculture system (RAS) is a relatively new method in the field of aquacultural food production. RAS is characterized as a booming

industry globally, with a recent annual growth rate estimated at 5.3% during 2001–2018 (Marking and Bills, 1982; Ahmed and Turchini, 2021). Such a high growth rate is attributed to the general rise in food demand associated with population increase and the growing interest in a seafood diet for its rich nutritional values (Campanati et al., 2022). The attractive features of RAS, such as a conservative use of water and land, a well-controlled growth environment, nutrient recirculation, and minimal environmental impact have motivated the further flourishing of the industry (Badiola et al., 2012).



The recirculating aquaculture system has been an important approach to controlling non-spot pollution from the aquatic industry. The accumulation of ammonium and nitrite, which is a highly toxic substance for fish, shrimp, and other aquatic creatures, will prevent the widespread use of recirculating aquaculture systems in the aquatic industry. Therefore, the effective elimination of nitrogen pollutants, especially ammonia and nitrite, is the key to the treatment of aquatic wastewater. However, the efficient removal of nitrogen pollutants is still a challenge due to low organic carbon concentration and high concentration of dissolved oxygen in aquatic systems (Lu et al., 2020). Nitrogenous compounds can be removed from fish production systems by processes that may be mechanical, physicochemical, or biological (Lu et al., 2020). Among these, biological processes are more reliable, sustainable (Lu et al., 2020), economical, and efficient methods of nitrogenous compound removal, following natural decomposition routes under controlled conditions. Ammonia, nitrite, and nitrate levels in recirculating aquaculture systems are mainly controlled by nitrification (Hagopian and Riley, 1998) and denitrification processes (Van Rijn, 2013). Nitrifying bacteria include the genera *Nitrosococcus*, *Nitrobacter* (Fu et al., 2010), *Nitrospira* (Patel et al., 2022), *Nitrococcus* (Langone et al., 2014), *Nitrospina*, *Nitrosomonas* (Patel et al., 2022), which oxidize ammonia to nitrate, through nitrite, under aerobic conditions. Recent studies have shown that *Nitrospira* can perform both nitrifying processes, oxidizing both ammonium and nitrite (Van Kessel et al., 2015). The end product nitrate can be reduced to free nitrogen (N₂) under anaerobic conditions (Rajta et al., 2020). Nitrifying bacteria are known to be highly sensitive and susceptible to their environment, therefore, biological filters should consist of noncorroding material such as fiberglass, plastic, rock, or ceramic that have large surface areas (DeLong and Losordo, 2012) where nitrifying bacteria can attach (DeLong and Losordo, 2012).

With the entry of suspended solids into the aquaculture system, all suspended particles are placed at the bottom of the fish breeding pools,

and sediments cannot easily be removed from the breeding pools. The amount of suspended solids and the increase of turbidity in the wastewater are also mainly composed of fine particles of organic matter that can quickly reduce the dissolved oxygen in the system. Suspended solids are primarily removed with mechanical filters. Two common filters include "drum" and "sand" filters (Summerfelt, 2006). The rapid removal of suspended solid particles from nutrition and feces is one of the most important processes in a recirculating aquaculture system. Decreasing suspended solids as the first strategy for reducing loading from aquatic fields during wastewater treatment is mainly based on the removal of solids from sewage, various methods are used to remove the total suspended solids. Micro screens are one of the tools used for the initial treatment in recirculating aquaculture systems due to the minimum requirements for the use of labor and workspace. The drum filters are more likely to be preferred in the recirculating aquaculture systems. However; a drum filter's efficiency can show a lot of changes due to many factors, including pool management practices, feed features, particle size distribution, filter mesh size, hydraulic/solid loading speed, and total suspended solids concentration (Ali, 2012).

There is limited data on the efficiency of the purification system of recirculating aquaculture systems in fields with high pollution rates. Therefore; more efforts should be put into evaluating the efficiency of wastewater purification systems in recirculating aquaculture systems. The purpose of this study was to investigate the performance and efficiency enhancements of a wastewater purification system in the semi-recirculated aquaculture system of Namrud. This research stands out due to its innovative approach to optimizing wastewater treatment specifically within aquaculture systems, which are often challenged by water quality issues. By employing advanced purification technologies, the study explores the integration of novel filtration methods and bio-treatment processes that could significantly reduce pollutant levels and improve overall water quality. Additionally, the study examines the environmental benefits of these enhancements,

showcasing how improved wastewater management can lead to sustainable aquaculture practices. The findings aim to contribute actionable insights for aquaculture operations seeking to balance production efficiency with environmental responsibility.

2. Material and Methods

2.1. Study location and experimental timeline

The experiment was conducted from November to the end of January 2023 at the Namrud semi-recirculated aquaculture system. The unit is located at 135°45'08" with a height of 1980 meters above sea level.

2.2. Aquaculture process description

The semi-recirculated aquaculture system consists of seven breeding pools, each equipped with seven splash aeration units to enhance dissolved oxygen levels. Additionally, two aeration towers are utilized to release dissolved gases from the water. To ensure a reliable water supply for this aquaculture unit, three wells are employed, maintaining a relatively stable temperature and flow throughout the year. The inflow rate of water entering the system is 10 liters per second, while the outflow rate of discharge water is also 10 liters per second. The flow of recirculated water to the fish-breeding pools is 40 liters per second, indicating a semi-recirculated water system. The wastewater treatment process implemented in this study includes solid removal via a grit chamber unit, followed by biofilters, aeration, and oxygenation through aeration towers.

2.2.1. Fish stocking and management practices

During the study period in this unit, 25,000 pieces of Rainbow trout (*Oncorhynchus mykiss*), with an average weight of 50 grams, were purchased from the breeding and fish-selling center recommended by the Iran Fisheries Organization in Tehran province. In November, at the beginning of the breeding period, the trout were placed in fish-growing pools. Throughout the study, the trout were fed twice daily at 8:00 AM and 4:30 PM, using the diet recommended by the product of Behparvar Company. Feeding rates were determined by

observing the trout's feeding behavior and total biomass and were adjusted after weighing a sample of fish.

2.2.2. Experiment design and water quality parameters

The assessment of water quality (physicochemical properties) in fish breeding pools for both warm and cold-water species adheres to National Standard 8726. Additionally, National Standard 7961 is employed for sampling and protecting the samples. To understand fluctuations in pollution, 14 water quality parameters were examined across four different sampling stations. Sample containers were fully sealed and protected from excessive light and heat, as sample quality may fluctuate rapidly due to gas outflow, chemical reactions, and the metabolism of microorganisms. Therefore, to preserve the samples for a short period, the temperature of the icebox was maintained at 4 Celsius degrees according to National Standard 7961. Sampling was conducted using glass bottles. The sampling containers were first rinsed with deionized water and then with distilled water. Before transferring the samples to the laboratory, preliminary tasks were completed, including determining the temperature and preparing labels that contained station specifications, sampling time, atmospheric conditions, and sample names. The samples were then transported to Abram Lab at the Abbaspour Campus of Shahid Beheshti University (SBU) under suitable temperature conditions and within the shortest possible time. They were kept in the icebox before testing. Several physical and chemical parameters were measured on-site using portable devices available in the aquaculture system. On-site tests included measurements of temperature, dissolved oxygen, and pH. The results of these measurements, along with corresponding descriptions of the water samples, were prepared to monitor and control the accuracy of the devices in the aquaculture unit before being sent to Abram Lab. Parameters such as nitrite, nitrate, ammonia, total hardness, total alkalinity, iron, total dissolved solids (TDS), total suspended solids

(TSS), biochemical oxygen demand (BOD), and chemical oxygen demand (COD) were analyzed at Abram Lab. The test methods for analysis of water and wastewater samples adhered to standard protocols, including: 1. Standard Methods for the Examination of Water and Wastewater, 2. ASTM standards.

2.2.3. Water Treatment and Restoration Method

In the semi-recirculated environment of Namrod fish farming, water treatment, and circulation are achieved through several processes. First, suspended solids are removed using a grit chamber. Next, ammonia is converted to nitrite and finally to nitrate through biofilters. Aeration is facilitated by air towers and splash devices. Finally, the water is disinfected by adding rock salt. The transfer of treated water occurs in stages; it first enters the second aeration tower (AB2) before being directed to the fish breeding pools. This transfer is carried out using Heli-fax or polyethylene pipes, and water transfer pumps with capacities of 8 and 5.5 KVA. Additionally, return effluent pumps operate in two 12-hour cycles.

2.2.4. Selection of stations and sampling periods

Sampling was carried out in the autumn and winter seasons in a trout breeding course in the Namrud fish farming in cooperation with the Abram lab at Shahid Beheshti University (SBU) at Abbaspour Campus of Shahid Beheshti University (SBU). Sampling was measured in 3 periods and 14 parameters in four stations. In general, the criteria involved in selecting sampling stations are 1. Quality control of incoming water from the well before it is combined with recirculated water from fish-breeding pools (input flow to the system) 2. Points that were likely to be the parameter value There is a significant increase, for example, fish-breeding pools 3. Points likely to be seen as a large reduction in the number of parameters were selected to control the function of biofilters. Since the main goal of monitoring water quality parameters is to understand the ecological quality of the return effluent at the site of the fish-growing pools and the changes made to it, therefore; tried to select water quality parameters, sampling stations, and

periods in a way to show the points that emit the highest amount of pollution.

Selection of Sampling Stations and Periods
Sampling was conducted during the autumn and winter seasons at a trout breeding facility within the Namrud fish farming project, in collaboration with the Abram Lab at the Abbaspour Campus of Shahid Beheshti University (SBU) (Fig. 1). The sampling was carried out over three periods, measuring 14 parameters across four stations. The criteria for selecting sampling stations included: **Quality Control of Incoming Water:** Monitoring the quality of water sourced from wells before it is mixed with recirculated water from the fish breeding pools (input flow to the system). **Potential High Parameter Values:** Identifying areas, such as fish breeding pools, where significant increases in parameter values were likely to occur. **Biofilter Performance Monitoring:** Selecting points where substantial reductions in water quality parameters were anticipated to evaluate the effectiveness of biofilters. The primary goal of monitoring water quality parameters is to assess the ecological quality of the return effluent from the fish breeding pools and to identify any changes. Therefore, efforts were made to select water quality parameters, sampling stations, and periods that would effectively highlight the locations with the highest levels of pollution.

3. Results and discussion

In this study, we focused on key factors such as water quality parameters, sampling points, and laboratory methods to identify sources of pollutants and assess the qualitative status of a semi-recirculated aquaculture system. Aquatic environments are complex ecosystems with numerous water quality variables, several of which play a fundamental role in aquaculture. The most critical parameters affecting fish growth performance include dissolved oxygen (DO), temperature, pH, suspended solids, ammonia, nitrite, and carbon dioxide (CO₂), while alkalinity is also important for nitrifying processes (Ebeling and Timmons, 2012).

3.1. Water quality parameters

3.1.1. Sample Collection in November

A review of water quality parameters in November indicates that the highest pH value was 7.2 at a station (AB1), which subsequently declined to 6.8 in the outlet channel of the breeding pools (S1). Nitrification is highly sensitive to pH, and this process significantly declines at pH levels below 6.8 (Tomaszewski et al., 2017). The total dissolved solids (TDS) in the aeration tower of incoming water to the system (AB1) increased slightly from 70 mg/L to 160 mg/L (Malone and Pfeiffer, 2006). Some biofilter materials, applied as fixed bed filters, can eliminate TDS from the system water (Malone and Pfeiffer, 2006). The results of the water quality parameters from the November experiment are shown in Figs. 2 and 3 (Malone and Pfeiffer, 2006). The biochemical oxygen demand (BOD) at the inlet flow aeration tower (AB1) was recorded at 0.3 mg/L, indicating a low level of organic load in the inlet water to the fish farm. However, in other areas, this

value increased. After passing through the grit chamber, the (BOD) remained high, and in the aeration tower of recirculated water from the breeding pools (AB2), it decreased to 4 mg/L. The total suspended solids (TSS) gradually increased, as shown in Fig. 2, rising from 70 mg/L to 190 mg/L in the outlet channel of the breeding pools (S1), before decreasing to 170 mg/L at station S2. Despite this decrease, the recorded TSS values remained high, with station S2 showing a value of 170 mg/L after the biofilters. The lowest concentration of nitrate (NO₃) recorded in the aeration tower of incoming water (AB1) was 1 mg/L, while the highest concentration, 1.3 mg/L, was found in the outlet channel of the breeding pools (S1). This nitrate level at the outlet of the nurturing pools (S1) exceeds the standard value. However, after passing through the biofilters, the nitrate concentration decreased to 1 mg/L at station S2.



Fig.1. Sampling stations.

Description of the Nemerud semi- recirculated Aquaculture System:

AB1: Aeration tower of entering water to the system.

AB2: Aeration Tower of recirculated water of breeding Pools.

S1: Outlet channel of breeding pools.

S2: Wastewater flow after passing through the drum filter and biofilters.

Sampling Period: Sampling was conducted in November and December of 2022, and January of 2023.

Average Weight of Aquatic Species During Sampling Periods: November: 50 to 100 grams December: 200 to 300 grams January: 500 grams 500 grams.

Inflow Water Rate to the System: 10 liters per second.

Outflow Water Rate from the System: 10 liters per second Recirculated Water Flow Rate to Fish Ponds: 40 liters per second.

Total Number of Aquatic Species at the Start of the Cultivation Period: 25,000 individuals.

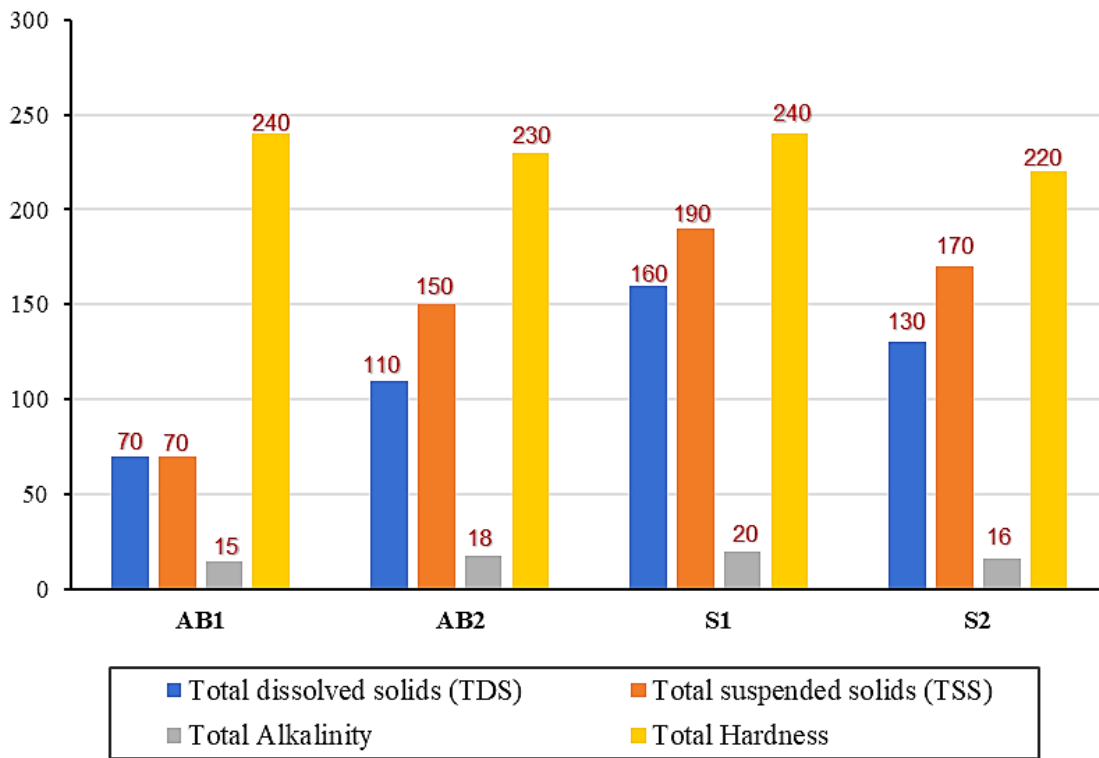


Fig. 2. Water quality parameters in November.

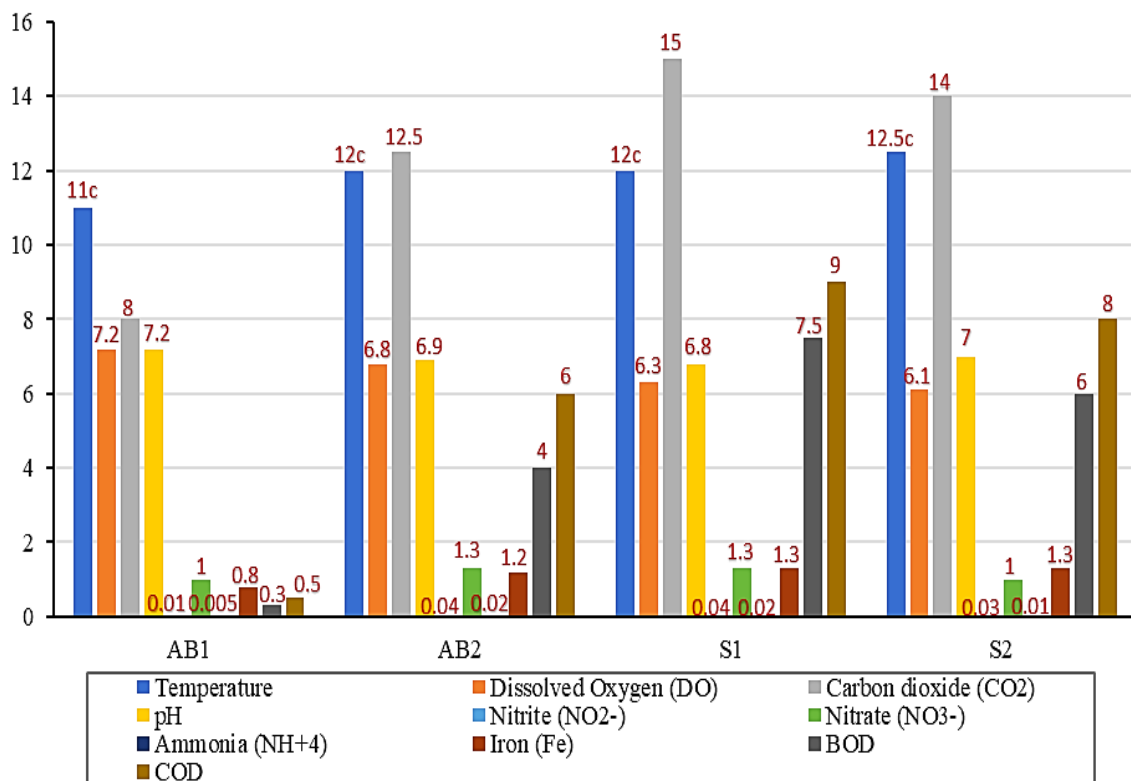


Fig. 3. Water quality parameters in November.

3.1.2. Sample collection in December

The highest concentration of (BOD) is 8.5 mg/Lit at the Outlet channel of breeding pools (S1) which indicates the high level of organic materials due to the high density of fishes (trout) In the fish breeding pools another reason is due to the average weight of fishes that increased from 50 grams to 300 grams in December that caused an increase in feces of fishes. Results for this water quality parameter in this experiment are shown in Fig. 4. The

nitrate level in the sampling performed in the outlet channel of breeding pools (S1) witnessed an increase from 2.5 mg/Lit in December compared to 1.3 mg/Lit in November, indicating an increase in the consumption of food by fishes. The value of total dissolved solids (TDS) in December was 190 mg/Lit at the Outlet channel of breeding pools (S1) which increased compared to the previous month due to the high consumption of food and the increase in the weight of fishes.

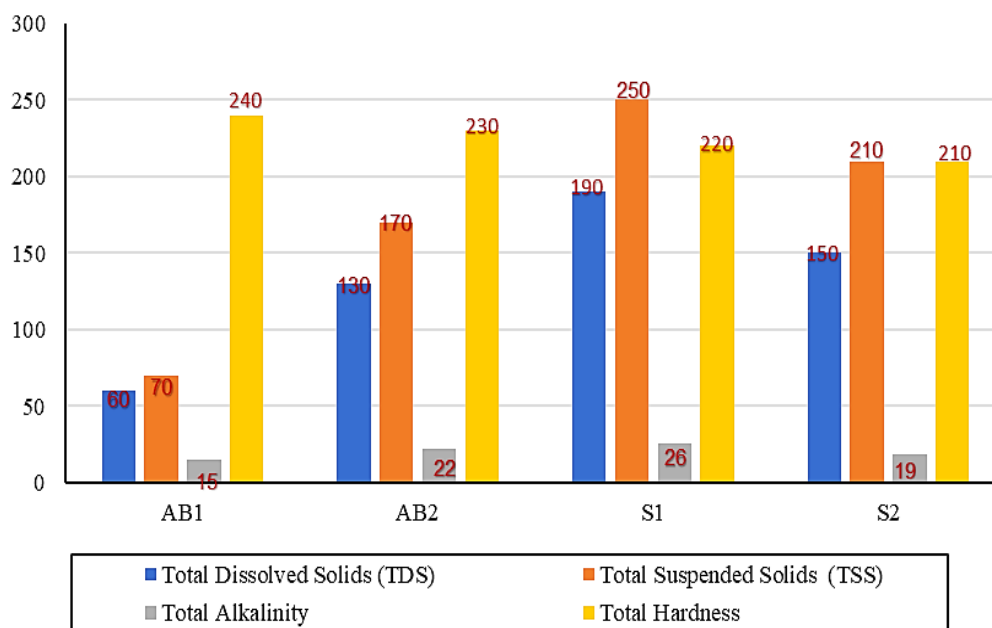


Fig. 4. Water quality parameters in December.

A study by (Emparanza, 2009) showed factors affecting nitrification in commercial RAS with fixed-bed biofilters and revealed that high oxygen consumption in the biofilters leads to oxygen depletion in the system hence affecting nitrification and consequently production in the RAS. The concentration of total dissolved solids (TDS) in the sampling after biofilters decreased significantly, indicating the efficiency of the filters. Ammonia (NH_3) concentration at the Aeration tower of entering water to the system (AB1) was constant in the sampling performed in December compared to the previous month but the value of this parameter increased from 0.02 mg/Lit to 0.1 mg/Lit at Outlet channel of breeding pools (S1) compared to the previous month. In the nitrogen cycle, nitrifying bacteria utilize oxygen and alkalinity in the process of converting ammonia and nitrite into nitrate

which is less toxic to fish (Francis-Floyd, 2020).

3.1.3. Sample collection in January

During the last sampling in January, some water quality parameters slightly increased due to the accumulation of fish feces in the breeding pools. The highest concentration of nitrate was 3 mg/L at the outlet channel of the breeding pools (S1), which exceeds the recommended limits of National Standard 8726. The nitrate (NO_3) concentration did not decrease after passing through the biofilters, which is attributed to the function of microorganisms on the surface of the biofilters, as well as factors such as light, pH, and water temperature that significantly affect microbial activity. The lowest recorded value of dissolved oxygen (DO) at the sampling station after the biofilters (S2) was 5.2 mg/L, a slight decline compared to

the concentration of dissolved oxygen entering the fish breeding pools. This decrease is primarily due to the high consumption of Behparvar's formulated food products and the weight of the trout in the breeding pools. Researchers have observed a significant effect of dissolved oxygen (DO) concentration on ammonia-oxidizing bacteria (AOB), with *Nitrosomonas europaea* dominating the microbial community when DO levels fall below 0.24 mg/L (Xu et al., 2022). In contrast,

Nitrosomonas oligotropha were found to be optimally predominant at a dissolved oxygen concentration of 8.5 mg/L (Langone et al., 2014). The changes in total hardness concentration across different sampling stations are fluctuating, as shown in Fig. 5. The value of this parameter was 240 mg/L at the aeration tower where water enters the system (AB1) and then declined to 200 mg/L at the outlet channel of the breeding pools (S1).

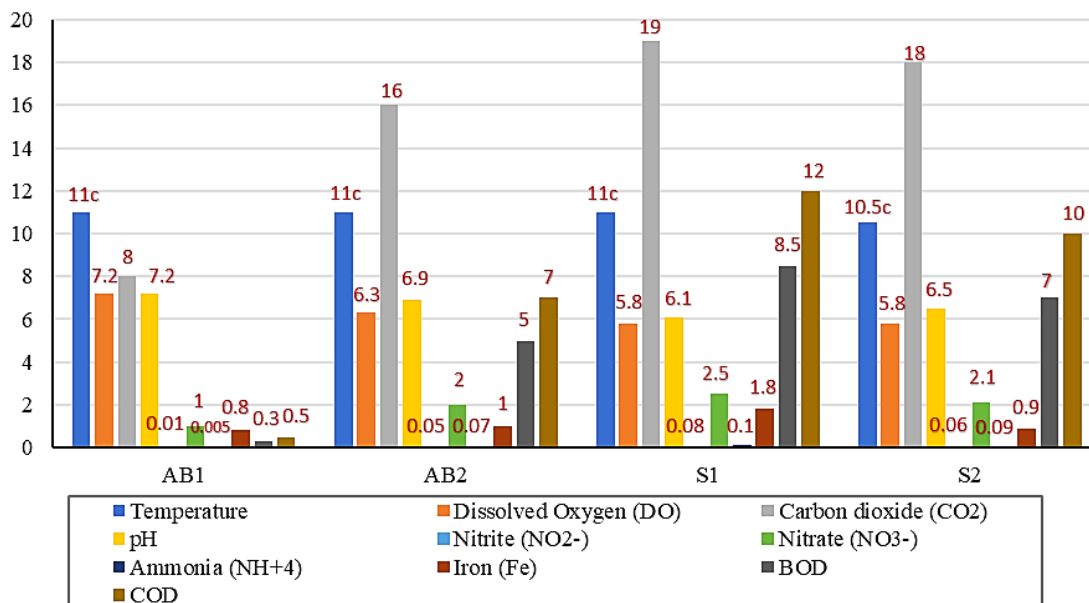


Fig. 5. Water quality parameters in December.

3.2. Qualitative comparison of three sampling periods

As illustrated in the figures from the three rounds of sampling, the values of BOD, COD, total dissolved solids (TDS), total suspended solids (TSS), alkalinity, total hardness, nitrite (NO₂), nitrate (NO₃), and ammonia (NH₃) all declined after passing through the grit chamber, aeration towers, and biofilters. One notable aspect of the Namrud semi-recirculated aquaculture system, compared to other aquaculture systems visited during the study, is the design of the grit chamber. With its appropriate slope and carefully considered height and width, the chamber effectively reduces organic materials after wastewater passes through it. The wastewater treatment process remained consistent throughout the research period. In the November sampling, the average weight of the fish was 50 grams, and

the amount of feces produced was low, resulting in water quality parameter values that were within standard limits. However, in December and January, the average weights of the fish increased to 300 grams and 500 to 550 grams, respectively. This increase in fish feces significantly impacted the water quality parameters. For instance, in the January sampling, ammonia (NH₃) levels at the outlet channel of the breeding pools (S1) exceeded the values of National standard 8726. The feeding behavior of fish is strongly influenced by environmental conditions such as water temperature, dissolved oxygen (DO), total ammonia nitrogen (TAN), and nitrite nitrogen (NO₂-N) (Buentello et al., 2000). Improvements in water quality within a recirculating aquaculture system (RAS) may enhance feed intake rates. The RAS provides optimal environmental conditions year-round, contributing to fish health and minimizing the

feed conversion ratio (FCR), thereby improving feeding efficiency (d'Orbcastel et al., 2009). Dissolved oxygen is one of the most critical abiotic factors affecting the growth and survival of fish, both in the wild and in aquaculture (Jobling, 1993; Taylor and Miller, 2001). Increased DO levels enhance feed consumption, feeding efficiency, metabolism, and growth in fish (Buentello et al., 2000). In this study, the removal of the drum filters significantly affected the performance of the filtration system, leading to increased values of water quality parameters. A comparison of water quality parameters across the three sampling periods indicates that the absence of the drum filter, which is essential for removing suspended solids, resulted in suboptimal wastewater treatment in the Namrud aquaculture system. Consequently, this led to higher levels of parameters such as TSS, BOD, COD, nitrate (NO_3), and ammonia (NH_3) than the limits set by National Standard 8726. As shown in Figs 6 and 7, high values of water quality parameters, including nitrite (NO_2), nitrate (NO_3), carbon dioxide (CO_2), BOD, and pH, along with low concentrations of dissolved oxygen (DO) in January compared to November and December, significantly impacted the fish's skin color and caused darkening of their internal organs, affecting the taste of the fish meat. In aquatic environments, ammonia nitrogen exists in two forms: un-ionized ammonia and ammonium ion. Un-ionized ammonia is toxic to fish, while

ammonium ion is generally harmless unless present at extremely high concentrations. Toxic levels of un-ionized ammonia for short-term exposure in pond fish typically range from 0.6 to 2.0 milligrams per liter, while sublethal effects can occur at 0.1 to 0.3 milligrams per liter. Regarding pH, its value is crucial concerning ammonia toxicity and carbon dioxide concentration. According to National Standard 8726, the optimal pH range is 6.9 to 7.2, with a pH of 7 resulting in reduced ammonia toxicity. Considering the low pH observed in January at the outlet channel of the breeding pools (S1) and after the biofilters (S2), it is recommended by the Iran Fisheries Organization to use sodium bicarbonate or calcium hydroxide to adjust and increase pH levels. Suspended solids are a primary source of various water quality issues, impacting nearly all other components of the RAS, as shown in this study. Therefore, effective management of suspended solids is fundamental for optimal system performance, as previously noted by Badiola et al. (2012). The design of air towers must account for the distance between floors to effectively release dissolved gases from the water. In the first air tower (AB1), the appropriate spacing between the floors ensures that the concentration of dissolved gases released before entering the fish breeding pools, as well as the concentration of dissolved oxygen after the air tower, aligns with the limits set by National Standard 8726.

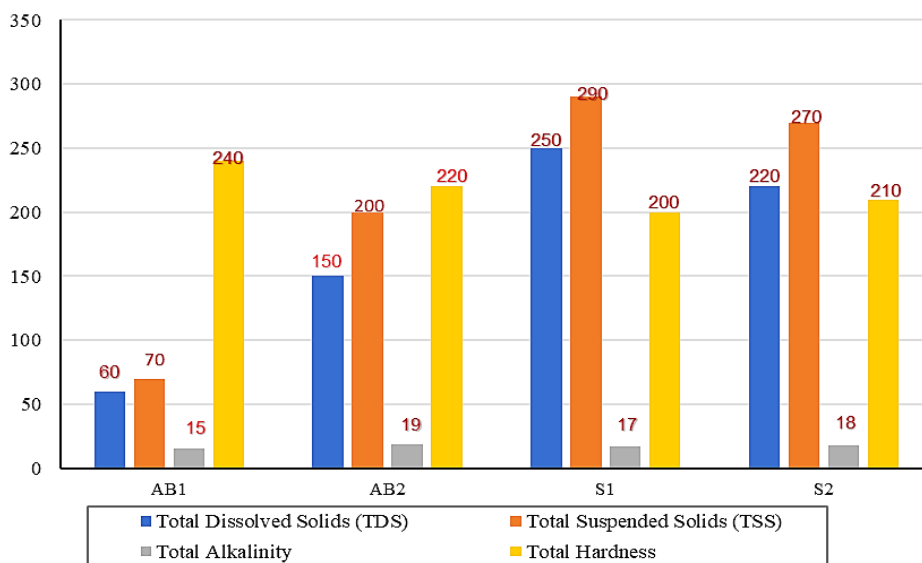


Fig. 6. Water quality parameters in January.

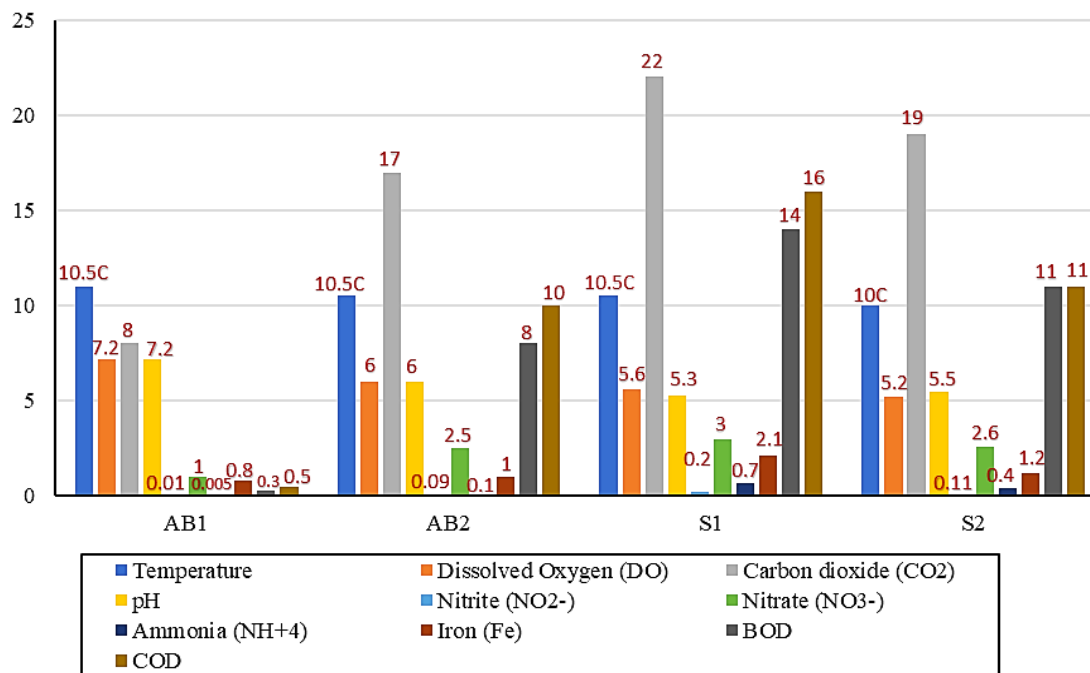


Fig. 7. Water quality parameters in January.

3.2. Qualitative comparison of three sampling periods

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Consequently, this led to higher levels of parameters such as TSS, BOD, COD, nitrate (NO_3), and ammonia (NH_3) than the limits set by National Standard 8726. As shown in Figs 6 and 7, high values of water quality parameters, including nitrite (NO_2), nitrate (NO_3), carbon dioxide (CO_2), BOD, and pH, along with low concentrations of dissolved oxygen (DO) in January compared to November and December, significantly impacted the fish's skin color and caused darkening of their internal organs, affecting the taste of the fish meat. In aquatic environments, ammonia nitrogen exists in two forms: un-ionized ammonia and ammonium ion. Un-ionized ammonia is toxic to fish, while ammonium ion is generally harmless unless present at extremely high concentrations.

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The design of air towers must account for the distance between floors to effectively release dissolved gases from the water. In the first air tower (AB1), the appropriate spacing between the floors ensures that the concentration of dissolved gases released before entering the fish breeding pools, as well as the concentration of dissolved oxygen after the air tower, aligns with the limits set by National Standard 8726.

4. Conclusion

The findings of this study confirm the significant impact of fish farming pools on surface water resources, underscoring the need for environmental measures to control pollutants from this industry through appropriate guidelines and regulations. Recirculating Aquaculture Systems (RAS) offer substantial benefits for the aquaculture industry, including high biosecurity, minimal effluent discharge, and flexibility in geographic location. The continued growth and diversification of the aquaculture industry necessitate an increasing number of governmental policies and recommendations. Results from performance assessments of the wastewater treatment systems in recirculating aquaculture systems, along with interviews conducted with aquaculture experts in Tehran Province and the Agriculture Organization in Firoozkooh County, reveal numerous problems within the aquaculture industry that contribute to its unsustainability. Selecting a suitable treatment method depends not only on a thorough cost/benefit analysis but also on factors that are directly or indirectly related to the location of the recirculating system. Climatic conditions, water availability, discharge regulations, and land availability are location-dependent factors that play a major role in determining the appropriate treatment methods to be utilized (Van Rijn, 2013). The most current policy, Hazard Analysis and Critical Control Points (HACCP), which has been adopted by the United States and the European Union, places the responsibility for the safe production and distribution of food within the aquaculture sector (Cole et al., 2009).

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