Growth Performance and Biochemical Composition of Nile Tilapia (Oreochromis niloticus) Reared in Membrane Bioreactor Treated Wastewater

Rachel Mwendwa¹, Michael Wawire¹, Peter Kahenya¹ & Edwin Oyoo²

¹ Department of Food Science and Technology, Jomo Kenyatta University of Agriculture and Technology, Nairobi, Kenya
² Department of Agriculture, livestock and Fisheries, Directorate of Fisheries, Kisumu County, Kisumu, Kenya

Correspondence: Rachel Mwendwa, Department of Food Science and Technology, Jomo Kenyatta University of Agriculture and Technology, P.O. Box 62000-00200, Nairobi, Kenya. E-mail: ramwa.24@gmail.com

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Abstract

The aquaculture sector in Africa has great potential for growth; however, it faces several challenges, one of them being the scarcity of clean water. This prompts the need for water recycling. The present study was conducted to investigate the effects of rearing Nile tilapia (Oreochromis niloticus) using municipal wastewater treated with membrane bioreactor (MBR) technology. A total of 270 Nile tilapia fingerlings (0.15±0.05 g) were reared in three treatment groups in triplicate. There were 2 treatments, including; MBR treated wastewater and stabilization pond treated wastewater (maturation pond), while the municipal tap water was used as the control. The growth performance (weight and length) of the fish was monitored over a 24-week period. After the experimental period, the biochemical composition of the fish muscle was analysed using standard AOAC methods. The results showed that the highest weight gain, length gain, survival rate, and specific growth were obtained in the fish in the control followed by the MBR treatment. Additionally, the crude protein, as well as the crude fiber and dry matter, were significantly higher in the fish in the maturation ponds at 23.10%, 0.29%, and 25.35%, respectively, while the crude ash was highest in the MBR at 1.22%. Results also showed that the MBR and maturation pond treatments meet the permissible levels for BOD, COD, NH₄, and NO₃ for water to be used in aquaculture. The bioaccumulation of heavy metals in the fish was mainly from the feed, with copper being the highest contaminant at 1.75 mg/100 g. In conclusion, both the MBR and maturation pond treated wastewater are viable for use in the rearing of Nile tilapia without adverse effect on the growth. However, MBR treatment showed better growth performance, suggesting that it could be used to increase productivity in fish farming.

Keywords: membrane bioreactor, Nile tilapia, physiochemical, heavy metals, maturation pond

1. Introduction

With the global population estimated to rise to 9.7 billion by 2050 from 7.7 billion currently, food demand will also increase proportionally (United Nations, 2019). To improve food security, eradicate hunger and malnutrition presently and in years to come, we need sustainable food production systems. Fish and aquatic plants are major contributors to a healthy and nutritious human diet. During the last five decades, global fish consumption has increased at a rate almost double that of global population growth in the same period (FAO, 2020). This, in turn, has fueled the demand for fish. Fish is an important part of the diet because it is a source of high-quality protein, rich in essential amino acids and long-chain polyunsaturated fatty acids, and when consumed whole with skin, head, and bones, it provides essential micronutrients such as selenium, calcium, iron, zinc, vitamins D and A (Khalili & Sabine, 2018; Tacon & Metian, 2013).

The world supply of fish is from wild catch and aquaculture, with aquaculture accounting for almost half of the total global fish production at 82.1 million tonnes in 2018 (FAO, 2020). There has been an overexploitation of capture fisheries through illegal, unregulated, and unreported fishing, leading to a decline in the wild fish stock and hence shifting the fish production system to aquaculture. Aquaculture has great potential to meet the demand for fish and fish products for the growing world population (Cao et al., 2013). In addition, it reduces overreliance on fisheries and improves the preservation of natural aquatic resources.
In developing countries, the growth of inland aquaculture is faced with the challenge of water shortage due to competition from other uses (FAO, 2014). Wastewater reuse is therefore a great alternative and valuable resource in sustainable aquaculture. Membrane technology is an effective wastewater treatment technology, in particular, the membrane bioreactor (MBR) technology (Bouhadjar et al., 2016; Stephenson et al., 2000). MBR technology employs a combined conventional activated sludge process with microfiltration or ultrafiltration process (Judd, 2010). The bioreactor is involved in the biodegradation of the organic waste while the membrane separates the treated water and the mixed liquor (Hoinkis et al., 2012).

MBR has gained great interest over the years as it provides high-quality effluent, a small environmental footprint, and good disinfection capabilities (Assayie et al., 2017; Mutamim et al., 2013). MBR technology has been widely applied in recirculating aquaculture systems as biological filters for wastewater produced in the system. This technology creates a new alternative to produce fish to promote food and nutrition security, but it also poses the question of fish safety for human consumption. Due to increasing interest in food quality and safety as a result of stringent food standards at national and international levels (FAO, 2016), there is a need to establish the quality of fish reared in treated wastewater.

Fish normally interact involuntarily with its culture environment (Ibrahim, 2015), this therefore mean they can draw and accumulate components in the water. For instance, they can bioaccumulate heavy metals. In fish farming the quality of water used is important in determining yields and survival of the fish. In this study, the effect of using MBR-treated municipal wastewater to rear Nile tilapia (Oreochromis niloticus) was investigated and compared with conventional pond-treated wastewater at the maturation pond. In stabilization ponds, the maturation pond is the point at which the treated wastewater is released into the environment and can be used in agriculture (Van Sperling & Chernicharo, 2005). The specific focus was on the growth performance of the tilapia fish, the biochemical composition of the muscle and the concentration of heavy metals.

2. Materials and Methods

2.1 Experimental Set-Up

Nile tilapia fingerlings were obtained from the hatcheries of the study site (ViclnAqua project). After acclimatization (temperature at 25 °C and pH at 7.8) for two weeks, male Nile tilapia fingerlings of average weight 0.15±0.05 g (mean±SE) and 1.32±0.11 cm in length were randomly distributed in nine circular cylindrical tanks of 200 L at a stocking density of 30 fingerlings per tank. Three tanks were assigned to each of the treatments; MBR-treated wastewater, maturation pond water, and control (tap water). All tanks were provided with aeration to maintain dissolved oxygen levels above 3 mg/L with a photoperiod of 12:12 h light: dark. Throughout the experiment period, the quality of the water (dissolved oxygen, pH, and temperature) was monitored daily with an automatic multiparameter analyzer (OxyGuard, probes). The water quality was maintained by changing the water regularly (twice a week).

The specifications of the membranes used in the membrane bioreactor are as follows: ultrafiltration membranes with a polymer of polyethersulfone (PES), molecular weight cut-off (MWCO) of 150 kDalton and pore size of nominal 35 nm (MARTIN Systems).

2.2 Experimental Diets

The fingerlings were initially fed an isonitrogenous diet (mash) at a daily rate of 40 % of their body weight with frequencies of four times a day (8:00 h, 11:00 h, 14:00 h, and 17:00 h) for the four weeks. After four weeks, the diet was changed to an isonitrogenous and isoenergetic diet (pellets) and feeding was carried out three times a day (08:00 h, 12:00 h, and 17:00 h) to apparent satiation, for 20 weeks. The proximate composition of the feed (% dry weight basis (dwb)) is presented in Table 1 below. During the entire experimental period, the fish (10%) were randomly sampled at intervals of 14 days, and the weight and length were taken, then released back to their respective tanks.

At the end of the experiment, the fish were fasted for 24 h, prior to sampling. Then they were individually weighed. Subsequently, the fish were slaughtered (asphyxiate on ice) and degutted, and the muscle (fillet) was collected for further analysis.
Table 1. Composition of experimental diets

<table>
<thead>
<tr>
<th>Ingredients (g/kg)</th>
<th>Experimental diets</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Diet 1 (mash) (g/kg)</td>
</tr>
<tr>
<td>Fish meal</td>
<td>-</td>
</tr>
<tr>
<td>Shrimp (Caridina nilotica)</td>
<td>1000</td>
</tr>
<tr>
<td>Wheat bran</td>
<td>-</td>
</tr>
<tr>
<td>Wheat pollard</td>
<td>-</td>
</tr>
<tr>
<td>Sunflower oil</td>
<td>-</td>
</tr>
<tr>
<td>Cassava flour (binder)</td>
<td>-</td>
</tr>
<tr>
<td>Mineral and vitamin premixes*</td>
<td>-</td>
</tr>
</tbody>
</table>

*Proximate composition (%)b

<table>
<thead>
<tr>
<th></th>
<th>Diet 1</th>
<th>Diet 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dry matter</td>
<td>91.00±0.10</td>
<td>94.15±0.06</td>
</tr>
<tr>
<td>Crude protein</td>
<td>62.16±1.26</td>
<td>33.09±0.34</td>
</tr>
<tr>
<td>Crude lipids</td>
<td>6.23±0.05</td>
<td>5.47±0.19</td>
</tr>
<tr>
<td>Crude ash</td>
<td>20.27±0.21</td>
<td>8.91±0.05</td>
</tr>
<tr>
<td>Crude fibre</td>
<td>2.32±0.48</td>
<td>28.36±1.17</td>
</tr>
<tr>
<td>NFEc</td>
<td>0.02±0.10</td>
<td>18.32±0.06</td>
</tr>
</tbody>
</table>

Note. a Vitamin and mineral premix composition per kg of feed: vitamin A, 600 I.U.; vitamin D3, 100 I.U.; vitamin E, 3 I.U.; vitamin K (menadione), 0.42 mg; vitamin B1, 0.25 mg; vitamin B2, 0.6 mg; vitamin B6, 0.5 mg; vitamin B12, 0.001 mg; niacin acid, 2.5 mg; pentothenic acid, 2.2 mg; folie, 0.15 mg; biotin, 0.001 mg; vitamin C, 1 mg; copper, 0.5 mg; manganese, 15 mg; zinc, 4.5 mg; iodide, 0.14 mg; selenium, 0.012 mg; cobalt, 0.02 mg; choline chloride, 15 mg; iron 4 mg.

(-) means that the component was not added.

b Proximate values are the means±S.E.

c NFE; Nitrogen free extract = 100 - (moisture content + crude protein + crude lipids + crude ash + fibre).

2.3 Analytical Methods

2.3.1 Physicochemical Analysis of Water

The pH, dissolved oxygen, and conductivity of the water samples were monitored using portable devices; pH meter (HANNA model, HI 2211), Oxygen Handy Polaris probe (OxyGuard model, Hv 312 Eu), and digital conductivity meter (HANNA DisT3 model, HI 98303) respectively. The nitrates and ammonia (NH3) levels were determined using rapid colorimetric methods. Chemical oxygen demand (COD) was analyzed using the potassium permanganate method, while biological oxygen demand was determined with the five-day incubation method (Li et al., 2018; Jouanneau et al., 2014). All the parameters were analyzed fortnightly.

2.3.2 Analysis of Growth Performance

The growth parameters (weight gain, length gain), survival rate, and food index parameters (specific growth rate, condition factor, and feed conversion ratio) were calculated using equations from Tekinay and Davies, 2001:

\[
\text{Survival rate (SR,\%)} = \frac{\text{Number of live tilapia at end of experiment}}{\text{Initial number of tilapia}} \times 100
\]

\[
\text{Weight gain (WG, g)} = \text{Average final weight} - \text{Average initial weight}
\]

\[
\text{Length gain (LG, cm)} = \text{Average final length} - \text{Average initial length}
\]

\[
\text{Specific growth rate (SGR, \%)} = \frac{\log \text{final weight (g)} - \log \text{initial weight (g)}}{\text{Experiment period}} \times 100
\]

\[
\text{Condition factor (CF)} = \frac{\text{Final weight (g)}}{\text{Final length (cm)}^3} \times 100
\]
Feed conversion ratio (FCR) = \frac{\text{Total feed intake (g)}}{\text{Total wet weight gain (g)}} \quad (6)

2.3.3 Proximate Analysis of Muscle

The proximate analysis of the fish muscle was performed using standard AOAC methods (AOAC, 1995). The moisture content was determined by drying the samples in a hot air oven at 105°C for 4 h. Crude protein (N x 6.25) was determined by the semi-micro Kjeldahl method after acid digestion using a Kjeldahl System (Velp Scientifica model fitted with DK6 heating digestor, SMS scrubber, and JP recirculating water aspirator). Crude lipid was extracted and determined using the Soxhlet system (Geobraeut model). Ash content was calculated from weight loss after incineration of the samples in an Advantec KL-420 electric muffle furnace at 550°C for 1 h, and then cooling.

2.3.4 Heavy Metal and Mineral Analysis

Mineral concentrations in the fish samples were determined using the atomic absorption spectrophotometry (Shimadzu AAS AA/AE) as described by Alvin and Gardner, 1986. Similarly, the water samples were analyzed for metals and minerals using the AAS after digestion with nitric acid. The detection wavelengths of the minerals were as follows: Mg, 285.42 nm; Na, 589.79 nm; Ca, 422.40 nm; K, 766.12 nm; Fe, 510.11 nm; Zn, 214.15 nm; Mn, 279.48 nm; Cd, 228.70 nm; Cr, 357.87 nm; Cu, 324.80 nm; and Pb, 283.52 nm.

2.4 Statistical Analysis

Triplicate samples were used in all the experimental analyses. The experimental data were expressed as the mean±standard error (SE). Shapiro-Wilk Test was used to determine the normality of the data while the F-test was used to determine the homogeneity of the variance. The analysis of variance was performed by applying a one-way analysis of variance (ANOVA) followed by the Duncan multiple-range test to compare differences between treatments. Statistically significant differences between the means were considered when P < 0.05. Statistical analysis was performed using R, version 4.0.2 Software.

3. Results

3.1 Physiochemical Properties of Water

The results in Table 2 show that the water quality of the three treatments were significantly different (p < 0.05) except for the dissolved oxygen. The values for the dissolved oxygen ranged from 7.39 to 7.47. Conductivity was highest in the MBR-treated water at 692.67 µS/cm while the maturation pond recorded the highest values in BOD and COD at 8.52 and 20.42 mgO₂/l respectively. The concentration of NH₄ was lowest in the control at 0.06 mg/l.

<table>
<thead>
<tr>
<th>Water parameters</th>
<th>MBR water</th>
<th>Maturation pond water</th>
<th>Tap water</th>
<th>Permissible level for aquaculture (FAO, 1992)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dissolved oxygen (mg/l)</td>
<td>7.39±0.04*</td>
<td>7.39±0.00*</td>
<td>7.47±0.04*</td>
<td>Min 3</td>
</tr>
<tr>
<td>pH</td>
<td>7.31±0.03*</td>
<td>7.98±0.04*</td>
<td>6.47±0.14*</td>
<td>6.5-8.5</td>
</tr>
<tr>
<td>Conductivity (µS/cm)</td>
<td>692.67±0.88*</td>
<td>578.67±1.56*</td>
<td>157.33±0.88*</td>
<td>Max 2500</td>
</tr>
<tr>
<td>BOD, days at 20 °C (mgO₂/l)</td>
<td>4.14±0.19*</td>
<td>8.52±0.26*</td>
<td>1.05±0.02*</td>
<td>Max 15</td>
</tr>
<tr>
<td>COD (mgO₂/l)</td>
<td>16.03±0.03*</td>
<td>20.41±0.19*</td>
<td>0.02±0.00*</td>
<td>Max 30</td>
</tr>
<tr>
<td>NO₃ (mg/l)</td>
<td>19.33±0.36*</td>
<td>25.17±0.60*</td>
<td>3.60±0.52*</td>
<td>Max 44</td>
</tr>
<tr>
<td>NH₄ (mg/l)</td>
<td>0.09±0.02*</td>
<td>2.07±0.08*</td>
<td>0.06±0.01*</td>
<td>Max 0.2</td>
</tr>
</tbody>
</table>

Note. Data are expressed as the mean±SE. n = 3. Values in the same row with different superscript letters are significantly different (P < 0.05). Abbreviations: BOD, biological oxygen demand; COD, chemical oxygen demand; NO₃, nitrate; NH₄, ammonium; MBR, membrane bioreactor.

3.2 Growth Performance and Survival

The data on the growth performance and survival rate of the Nile tilapia in the three treatments are shown in Table 3. There was a significant difference (P < 0.05) in all growth parameters in the three treatments. After the
24 weeks feeding period, the mean weight differed significantly in the three treatments at 62.55 g, 57.84 g, and 50.48 g for tap water, MBR treated water, and maturation pond water respectively. The survival rate was significantly different with a high survival rate in the tap water (95.56%) followed by the MBR treated water (86.67%) and the lowest in the maturation pond water (76.67%). The maturation pond water showed the lowest performance, while the tap water had the best performance.

The weight growth curve showed a sigmoid pattern in all the treatments while the length gain over time was not significantly different among the treatments.

Table 3. Growth performance and survival rate of O. niloticus fed for 24 weeks.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Weight (g)</th>
<th>Length (cm)</th>
<th>SR (%)</th>
<th>SGR (% per day)</th>
<th>FCR</th>
<th>CF</th>
</tr>
</thead>
<tbody>
<tr>
<td>MBR</td>
<td>57.84±0.11</td>
<td>57.69±0.11</td>
<td>15.10±0.11</td>
<td>1.54±0.00</td>
<td>1.42±0.03</td>
<td>0.017±0.00</td>
</tr>
<tr>
<td>MP</td>
<td>50.48±0.22</td>
<td>50.33±0.22</td>
<td>14.70±0.08</td>
<td>1.50±0.00</td>
<td>1.26±0.06</td>
<td>0.016±0.00</td>
</tr>
<tr>
<td>TW</td>
<td>62.55±1.03</td>
<td>62.40±1.03</td>
<td>15.29±0.20</td>
<td>1.56±0.02</td>
<td>1.53±0.02</td>
<td>0.017±0.00</td>
</tr>
</tbody>
</table>

Note: Values are means±SE, n = 3. Values in the same column with different superscript letters are significantly different (P < 0.05). Abbreviations: SGR, Specific growth rate; SR, survival rate; FCR, feed conversion ratio; CF, condition factor; MBR, membrane bioreactor treated water; MP, maturation pond water; TW, tap water.

![Figure 1. Fish weight gain over a 24 weeks period in three different water qualities](image)

Note: MBR, membrane bioreactor treated water; MP, maturation pond water; TW, tap water.
Figure 2. Fish length gain over a period of 24 weeks in three different water qualities

Note. MBR, membrane bioreactor treated water; MP, maturation pond water; TW, tap water.

3.3 Muscle Biochemical Composition

Nile tilapia reared in the three treatments exhibited a biochemical composition similar to that shown in Table 4. The dry matter, protein, lipid, and fiber content in the three water qualities varied significantly (P < 0.05). However, there were no significant differences in the ash content in the rearing of fish in the three treatments. Tilapia fish reared in maturation pond water showed the highest fiber content of 0.29% and protein of 23.10%. The tap water-reared tilapia recorded the highest lipid content (1.04%) while the MBR water-reared tilapia had the lowest lipid content (0.74%).

Table 4. % Proximate composition of fish muscle

<table>
<thead>
<tr>
<th>% (wwb) Proximate composition/Treatment</th>
<th>Dry matter</th>
<th>Crude protein</th>
<th>Crude lipid</th>
<th>Crude ash</th>
<th>Crude fiber</th>
</tr>
</thead>
<tbody>
<tr>
<td>MBR</td>
<td>24.38±0.38a</td>
<td>22.35±0.37a</td>
<td>0.74±0.01a</td>
<td>1.22±0.08a</td>
<td>0.17±0.01b</td>
</tr>
<tr>
<td>MP</td>
<td>25.35±0.35a</td>
<td>23.10±0.30a</td>
<td>0.86±0.01a</td>
<td>1.10±0.05a</td>
<td>0.29±0.03a</td>
</tr>
<tr>
<td>TW</td>
<td>23.60±0.58b</td>
<td>21.36±0.51b</td>
<td>1.04±0.03a</td>
<td>1.01±0.07a</td>
<td>0.16±0.01b</td>
</tr>
<tr>
<td>P-Value</td>
<td>0.07</td>
<td>0.05</td>
<td>0.00</td>
<td>0.17</td>
<td>0.00</td>
</tr>
</tbody>
</table>

Note. Data are expressed as the mean±SE. n = 3. Different superscript letters in the same column are significantly different (p < 0.05). MBR, membrane bioreactor treated water; MP, maturation pond water; TW, tap water.

3.4 Minerals and Heavy Metals

The comparison of mineral and heavy metal concentrations in the tilapia fish muscle in the three treatments are shown in Figure 3. All the treatments exhibited high levels of Ca and low Mg levels in the macro minerals category. No significant difference (P < 0.05) was observed in macro and trace minerals between treatments except for Na, Mg, Zn, and Fe. The concentration of Mn, Ca, and K did not show a clear variation between the MBR and the maturation pond treatments. For the relative abundance of the minerals examined, the sequence of concentration in the fish muscle was Ca > K > Na > Mg > Fe > Zn > Mn. There was a significant difference in the heavy metal composition, with the Pb content being the highest. Analysis showed that fish samples from all three water treatments did not contain Cadmium metal.
Table 5. Comparative analysis of the heavy metal concentration in the feed, water and the fish muscle

<table>
<thead>
<tr>
<th></th>
<th>Treatment</th>
<th>Concentration (mg/100 g)</th>
<th>Pb</th>
<th>Cu</th>
<th>Cr</th>
<th>Cd</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>MBR</td>
<td>MP</td>
<td>TP</td>
<td>Feed</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Fish</td>
<td>Fish</td>
<td>Fish</td>
<td>Fish</td>
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<td>Water</td>
<td>Water</td>
<td>Water</td>
<td>Water</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>0.50±0.01</td>
<td>0.46±0.02</td>
<td>0.37±0.00</td>
<td>0.38±0.01</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>0.11±0.06</td>
<td>0.18±0.04</td>
<td>0.09±0.03</td>
<td>1.75±0.01</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>0.25±0.01</td>
<td>0.34±0.04</td>
<td>0.17±0.02</td>
<td>0.50±0.01</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
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<td></td>
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<td></td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
</tbody>
</table>

Note. Values are means±SE, n = 3. Abbreviations: MBR, membrane bioreactor treated water; MP, maturation pond water; TW, tap water; ND, not detected.

4. Discussion

In fish farming, the quality of water used is important in determining the yields and survival of the fish as the fish involuntarily interacts with its culture environment (Ibrahem, 2015). The quality of water for use in rearing fish needs to meet specific standards for the survival of the fish, to obtain a high yield, and even for the safety of the consumer. In a stressful environment (poor water quality), fish tend to expend more energy to cope with the stressor, and therefore the weight gain is less compared to fish in good quality water (Lugert et al., 2016; Tran et al., 2021). The present study shows that both treatments (MBR and maturation pond) meet the FAO recommended limits for water for use in aquaculture. However, both treatments were significantly different compared to the control. MBR treatment showed lower levels of BOD, COD, NO3, and NH4 than the maturation pond. The variation in quality could be attributed to differences in treatment techniques; the MBR employs both filtration and a bioreactor, while the maturation ponds are based on oxidation. In addition, the MBR system is equipped with ultraviolet (UV) treatment point to reduce the microbial load. Therefore, there appears to be room to further improve the maturation pond water by adding a UV treatment step. The findings of the study agree with Muralikrishna and Manickam (2017) who reported that the effluent at the outlet of the maturation ponds could reduce ammonia and BOD levels of approximately 10-15 and 5 mg/l respectively which is comparable to our findings. The ammonia levels in this experiment were even lower at 2.07 mg/l.

The Nile tilapia fish was used in this study because it is well-adapted and tolerant to a wide range of environmental conditions and has a high resistance to diseases and infections (Ndike et al., 2014; Ng & Romano, 2013). Low survival rates (76.67%) of the fish were experienced in the maturation pond due to deaths resulting from infections such as eye abnormalities (corneal cloudiness or opacity). Several studies have identified...
The nutritional profile of tilapia fish is influenced by its diet, its surrounding environment, and its ability to convert its feed into different nutrients (Shearer, 1994). The results of this study demonstrated that the water quality affected the biochemical composition of the tilapia, except for the ash content. The MBR-treated water performed better than the maturation pond water in terms of the biochemical composition. In a study to investigate whether the proximate composition of tilapia reared in pond water differs from that of the lake, the authors reported better performance in lake fish (Destá et al., 2019). Their findings on crude ash content (1.51-1.89%) are comparable to those in our study (1.01-1.22%).

As fish interact involuntarily with its environment it draws components in the water through bioaccumulation. However, the uptake of heavy metals is not exclusively by water exposure but depends also on the diet and feeding mode (Orata & Birgen, 2016). In the present study, the levels of heavy metals in the feed are included to substantiate how diet affects the overall composition of the fish. The high levels of these metals in the fish could be due to the inclusion of aquatic-based ingredients like fish meal and lake shrimp hence passing on the heavy metals through biomagnification along the food chain. Heavy metals in fish represent a threat not only to the fish themselves but also to the health of human beings (Vieira et al., 2011; Hill et al., 2005). From our results, copper was the major contaminant in the feed with 1.75 mg/100 g while lead bioaccumulation was highest in the fish muscle at 0.50 mg/100 g in the MBR treatment. The lead level in the fish muscle was above the safe levels recommended by the Codex Alimentarius Commission maximum of 0.03 mg/100 g in the fish, posing a potential risk to human health if consumed (CAC, 2019). Several studies have reported unsafe levels of lead in the muscle of fish caught in inland waters like rivers (Junejo et al., 2019; Jooste et al., 2015; Nevárez et al., 2015; Addo-Bediako et al., 2014). These findings suggest that fish caught in freshwaters may pose a risk of contamination with heavy metals as fish reared in treated wastewater. The heavy metal content in both the MBR-treated water and the maturation pond did not meet the permissible limits for discharge in the environment as established in the Kenyan legislation. The maximum limits for Cu, Cr, and Pb are 1.0, 2.0, and 0.01 mg/l (NEMA, 2006). The high levels of heavy metals in treated water could be attributed to industrial waste present in raw sewage and the persistent nature of heavy metals (Monier et al., 2022; Farouk et al., 2020; Duruibe et al., 2007). Furthermore, this shows that fish can be a good biomonitors of environmental pollution. A previous study by Orata and Birgen (2016) carried out on the same site as our study did not detect Cd as is our results. Furthermore, studies on some freshwater fish species did not also detect Cd (Korkmaz et al., 2017; Jooste et al., 2015) and indicated a lower bioaccumulation in the fish muscle (Ibrahim et al., 2018; El-Moselhy et al., 2014).

At low concentrations, certain minerals such as manganese, zinc, copper, iron, and magnesium are essential and have a recognized role in the metabolism of aquatic organisms (Shepperd, 2001). These minerals are actively regulated through the bile elimination process and their concentration in the fish muscle is maintained at certain levels depending on the need of the fish to maintain homeostasis (Murugan et al., 2008; Wdaminokoko, 2000). However, there exists international legislation on maximum permissible levels for Zn (4 mg/100 g) and Mn (0.25 mg/100 g) in fish (FAO, 1983). The legal limits for these minerals were not exceeded in our present study with Zn ranging from 1.05 to 1.35 mg/100 g and Mn from 0.13 to 1.15 mg/100 g. The concentration of the essential minerals Mg, Na, Zn, and Fe in the fish muscle showed variation in the maturation pond and MBR-treated water. Our study showed that the muscle of the fish from the maturation pond water contained relatively higher concentrations of the micronutrients Mn, Fe, and Zn, while the muscle of the fish from the MBR water had higher concentrations of the macronutrients Na and Ca. This accumulation pattern is likely related to their feeding, the presence of suspended green algae in the maturation water could have added to the diet of pellet feeds. A study by Santhakumaran et al. (2020), showed that algae are a valuable mineral source and some species contain Fe ranging from 0.08 to 10.21 mg/g while Zn content can range from 0.01 to 1.10 mg/g. The bioavailability of minerals from ingested food can be influenced by the ingestion rate, the nature of the food, and the effectiveness of food assimilation (El-Khatib et al., 2020; Marengo et al., 2018). The micronutrients concentration in the maturation pond and the MBR were listed in the order Fe > Zn > Mn. A similar trend was reported by Islam et al. (2012) in tilapia collected from the wild and cultured (pond, gher, and cage) in Bangladesh and in fish caught in the coastal waters of the southeast coast of India (Adam et al., 2022). Besides,
Jim et al. (2017) reported a similar range of concentration of Zn in Nile tilapia from three lakes in Zimbabwe. Our results for the macrominerals in the fish from the three water qualities were in the same order of magnitude (Ca > K > Na > Mg), compared to those reported for Oreochromis mossambicus (Ullah et al., 2022). However, these findings did not match those found in the literature for some freshwater species (Guerra-García et al., 2023; Islam et al., 2021; Jim et al., 2017), but in all studies K > Na. Our findings, therefore, suggest that MBR and maturation pond-treated water do not affect the capacity of fish to absorb and assimilate these trace minerals from water and diet.

5. Conclusions
From this study, it is clear that water quality plays an important role in the growth of fish altering how they utilize feed nutrients with the exception of trace minerals. The use of MBR was found to be performing satisfactorily in achieving the standards set for water for aquaculture, indicating the technology is viable for use in aquaculture. In addition, the study showed variation in the growth of tilapia in MBR-treated water and maturation pond, indicating better growth in MBR, hence suggesting that MBR technology can be used to promote productivity in fish farming. However, an additional polishing step has to be considered to remove the heavy metals in the MBR-treated water. A nanofiltration membrane is recommended to further upgrade the water quality by removing the divalent ions. Although fish can bioaccumulate heavy metals from the water, feeds also play a role in the final quality of the product and regular monitoring of feeds and fish habitat is recommended.

References


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Authors Contributions
RM was responsible for conceptualization, methodology, investigation, writing—original draft, writing—review & editing and funding acquisition. Dr. MW and Dr. PK were responsible for conceptualization, methodology, writing—reviewing, and supervision. EO was responsible for Conceptualization and methodology. All authors have read and agreed to the published version of the manuscript.

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There is no conflict of interest in connection with this study.

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