

Effect of different hydraulic loading rates on growth of basil (*Ocimum basilicum* L. 'Genovese') in nutrient film technique aquaponics

Farklı hidrolik yükleme oranlarının nütrient film tekniği akuaponiklerde fesleğen bitkisi (*Ocimum basilicum* L. 'Genovese') üretimine etkisi

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Abstract: Aquaponics are promising and sustainable technologies consisting of fish-plant-bacteria consortia in the same system, thereby providing an environmentally friendly system by recycling water and nutrients. This study was planned to investigate the influence of varying hydraulic loading rates (HLR) on the growth of basil plant (*Ocimum basilicum* L. 'Genovese') in a low-cost of electricity nutrient film technique aquaponics (NFT) integrated with African catfish (*Clarias gariepinus* (Burchell)) under the Eastern Mediterranean climate conditions, Antalya, Türkiye. The hydraulic loading rates tested in plant-growing troughs 2, 4, 8, and 12 m³/m²/day. African catfish showed an excellent feed conversion ratio (0.695) over the experiment. There was no statistically significant difference in plant height, number of leaves, and stem diameter for basil plants, but a statistically significant difference was found in plant weight and leaf area. The best plant weight gain was observed in the 4 m³/m²/day group with 23.0±2.5 g mean weight. The optimum HLR for basil production was estimated as 4.41 m³/m²/day based on yield (kg/m²) and energy consumption (KWh/kg basil) in a basil-African catfish integrated NFT aquaponics. The optimum HLR can maximize production without further increase of energy expenditure. Higher HLRs of 4.41 increase energy cost per unit of basil production.

Keywords: Aquaponics, hydraulic loading rate, *Clarias gariepinus*, *Ocimum basilicum*

INTRODUCTION

The tremendous increase in the human population and migration from rural to urban areas causes depletion of food resources and increase of food prices because of limited phosphorus and nitrogen resources in the earth, a trend particularly the case in developing countries (Wunderlich and Martinez, 2018). According to the United Nations' estimation, the world population will reach 9.6 billion by around 2050 and 75% of this population will be going to live in cities (Gerland et al., 2014). The population growth and overcrowding in cities will make sustainable food production inevitable.

Aquaponics is considered a good alternative for environmentally-friendly (Loomis et al., 2014), self-sufficient (El-Essawy et al., 2019), healthy (Sommerville et al., 2014), and sustainable (Tyson et al., 2011) food production. This system has versatile advantages such as easy integration into urban (Kledal, 2012), unoccupied, and unfavorable (Pantanello, 2018) areas. However, it is still a field that requires the expertise of several academic disciplines. It should be well-designed and well-organized in order to be economically feasible (Li et al., 2018).

Being the most common element in the atmosphere of

earth (Camargo and Alonso, 2006), nitrogen is an essential nutrient to build the structure of nucleic acids, proteins, adenosine phosphates, pyridine nucleotides, and pigments in all living organisms (Hagopian and Riley, 1998). On the other hand, supplemental nitrogen fertilization in agricultural production poses one of the major environmental problems of the world (Spiertz, 2009). The main nitrogen-dependent pollutants in nature are ammonium, nitrite, and nitrate, generally. Ammonium (NH₄⁺) is the most common inorganic form and pollutant of nitrogen in aquaculture systems (Clarkson et al., 1986). Nitrite (NO₂⁻) is the intermediate oxidation product of ammonium, and nitrate (NO₃⁻) is the last product of the nitrification process (Endut et al., 2014). These nitrogenous pollutants, if discharged to nature uncontrollably, can cause eutrophication in freshwater resources, especially during seasonal changes (Harada and Hiramatsu, 2010). On the other hand, these macromolecules are highly available sources of nutrients for plant species. In aerobic conditions, ammonia-oxidizing bacteria (AOB) convert the ammonium to nitrite whereas nitrite-oxidizing bacteria (NOB) convert nitrite to nitrate (Hu et al., 2015), which are non-toxic fertilizers for plants.

African catfish (*Clarias gariepinus* (Burchell)) is an indigenous fish for Türkiye (Geldiay and Balık, 2007) and is considered a good candidate for aquaponics in subtropical and tropical regions (Van der Waal, 1998; Baßmann et al., 2017) due to its high tolerance for suboptimal water conditions. Basil (*Ocimum basilicum*), which has glossy and oval-shaped leaves, is an annual and commercially important plant which have medicinal (Ahmed et al., 2014), ornamental (Kaurinovic et al., 2011) and cosmetic properties (Nguyen et al., 2010). Interestingly, basil appears to be an advantageous herb to grow in aquaponics due to its better growth performance in aquaponics and hydroponic systems compared with conventional production systems (Rakocy et al., 2003; Roosta, 2014). Basil requires a high amount of water for optimal growth (Meyers, 2003), which may be a reason why it is one of the most preferred plants for growing in aquaponic systems (Love et al., 2015).

Fish feed represents one of the major operating costs (Quagrainie et al., 2018) and the feed is the ultimate source of nutrients required by fish and plants in aquaponic systems (Roosta, 2014). The feeding level of fish is important not only for water quality and fish growth but also for plant growth (Liang and Chien, 2013). The water flow rate, which is important for nutrient distribution in aquaponics, is directly related to the hydraulic loading rate (HLR) in the system. HLR has a significant impact on the effectiveness of oxygen uptake by the plant roots by supporting nitrogenase activity that is produced by nitrogen-fixing bacteria in the root environment (Wittenberg et al., 1974). Low HLR can promote denitrification in an aquaponics environment, which is an unwanted situation for ammonium-oxidizing bacteria (Shete et al., 2016). Optimum HLR in plant beds, on the other hand, promotes nitrate production, which can improve the performance and yield of plants in aquaponics (Yang, 2019; Yep and Zheng, 2019) whereas excessive loading rates can decrease the nutrient uptake by the plants due to less contact time of the root with the nutrient (Shete et al., 2016).

To prevent chlorosis that caused loss of the normal green coloration of leaves, ferrous chelating agents are used in aquaponics (Kotzen and Appelbaum, 2010). FeDTPA, Fe-EDDHA, and Fe-HBED are the most effective and commonly used ferrous chelating agents commonly used in aquaponics (Kasozi et al., 2019; Tetreault et al., 2023).

Research on the determination of optimum HLR for fish and plant growth in aquaponics has attracted significant attention. Although various flow rates showed similar growth of fish (*Cyprinus carpio*) and in an aquaponic system, an HLR of 1.8 m³/m²/day was reported as the most effective in terms of growth of water spinach (*Ipomea aquatica*) (Endut et al., 2009). Higher HLRs carry much more macro and micronutrients to the roots in comparison to lower water flow rates (Caron et al., 2002). An HLR of 3.3 m³/m²/day significantly reduced the negative effects of ammonia for fish and had a positive effect on nutrient flux for plant production in aquaponics (Yang and Kim, 2020a). On the other hand, it is reported that increasing HLR could have caused lower lettuce production and worse FCR values in fish (Dediu et al., 2012). Increasing flow rate

from 4 L/min to 6 L/min resulted in higher nutrient uptake of nitrogen, phosphorus, potassium, and magnesium and better plant shoot, root, biomass, and fruit performance in tomatoes maintained in a hydroponic system fed with aquaculture effluent (Khater et al., 2015).

It appears from the literature that there is no one optimum HLR for all plant species maintained in the aquaponic systems. There is a lack of information about the impacts of HLR on basil growth in aquaponic systems. Therefore, the present study was planned to investigate the impacts of different HLRs, without aeration, in NFT aquaponics integrating African catfish with basil on plant growth (g) and yield (kg/m²).

MATERIALS AND METHODS

System setup

The NFT aquaponics with a total volume of 3.40 m³ was established at Mediterranean Fisheries Research, Production, and Training Institute (MEDFRI), Antalya - Türkiye. The details of the system setup were given by Yeşiltaş et al. (2021). Briefly, the system consisted of a fish rearing tank (2.5 m³), two pieces of radial flow separators (80 L apiece), two pieces of bio-filter tanks (150 L apiece), twelve troughs (21.6 L each) and a sump tank (150 L). All tanks in aquaponics including fish tank were made of polyvinyl plastic materials. This study was conducted outdoor under the climate conditions of the Eastern Mediterranean for 42 days during summer between June and July. In this study, glass wool material was preferred as a substrate in troughs, unlike the previous study. Air temperature, humidity and wind speed during the study period were recorded, daily. The water used in the aquaponic system was taken from the main water supply of the MEDFRI aquaculture facility. The water that is lost through evaporation from the system and about 10 L daily loss through the removal of settled solids concentrated in the bottom of the fish rearing tank is reinforced by using an automatic float valve installed at the sump tank supported by the aquarium unit. Water flow rates into the treatment troughs (Figure 1) were controlled by manual valves at each inlet. HLR values were tested in this experiment and HLR of the troughs were calculated using the formula of water flow rate of trough (m³/day) / surface area of trough m². 2, 4, 8, and 12 m³/m²/day HLRs hydroponic rafts were set up for comparison with each other.

The previous study's biofilter tanks that had already included nitrification bacteria on the surface of bio-balls were reused in this experiment to utilize for the nitrification process (Yeşiltaş et al., 2021). However, the nitrification process did not work well as desired due to the lack of aeration in the system. High but not toxic to African catfish and plants ammonium concentrations were obtained in different aquaponics units. The wastewater from the fish-rearing tank flowed by gravity into the sequential radial flow separators and then to the bio-filter tanks filled with about 100 L of volume. Nitrified water in the bio-filter tanks was flowed by gravity into the hydroponics unit and then the sump/pump tank. The water was then pumped back to the fish-rearing tank with the aid of a 0.25 kWh water pump all day and night long (Pedrollo, Model Top 1, Italy) (Figure 1).

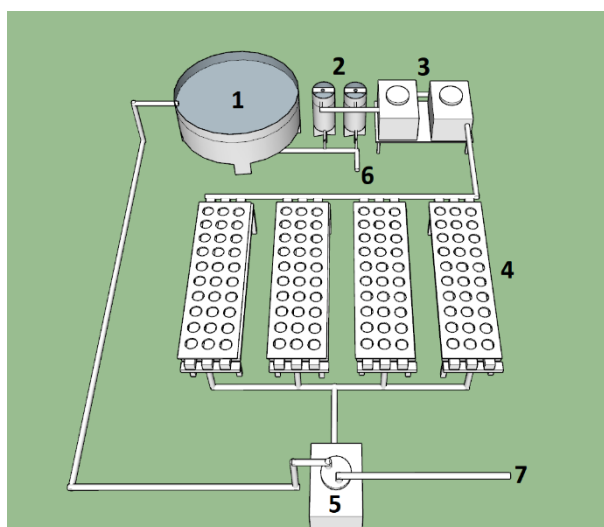


Figure 1. Schematic representation of the NFT aquaponics system used in study 1: Fish tank, 2: Separation tank, 3: Bio-filter tank, 4: Hydroponic troughs, 5: Sump/Pump tank, 6: Solid waste discharge pipe, 7: Freshwater inlet

Fish growth and rearing

African catfish used in the study were obtained from the hatchery of MEDFRI. A total of 128 fish with an average weight of $195.43 \text{ g} \pm 62.91 \text{ g}$ were stocked into the fish-rearing tank at the beginning of the experiment. The initial stocking density was set as 10 kg/m^3 . Fish were fed with a commercial juvenile common carp diet at predetermined levels. High protein juvenile commercial carp diet (50% crude protein, 8% crude oil) was used during the experiment (Özpekler Su Ürünleri, Denizli, Türkiye). Feed conversion ratio (FCR), specific growth rate (SGR), survival rate (SR), growth rate (GR), and fish biomass increase were calculated at the end of the experiment. FCR was determined using of "total weight of dry feed given/total wet weight gain" formula. SGR was calculated using of "(log final weight - log initial weight) \times 100 / culture period (days)" formula. SR was determined by use of "(final fish number/initial fish number) \times 100" formula. GR was measured using of "(final total weight - initial total weight) \times 100/initial total weight" formula.

Plant growth

Each HLR was tested in triplicated troughs for 42 days of the experiment. 120 basil (*O. basilicum* L. 'Genovese') seeds were sown in seedling trays and germinated within 12-15 days. Basil seedlings were irrigated daily by hand in a greenhouse and transferred from the trays to aquaponics troughs after 22 days. Basils with approximately a mean weight of 0.1 g were planted in twelve troughs with four groups. 60 x 240 cm length styrofoam boards and 13 cm height, 7.5 cm top diameter, and 4.5 cm bottom diameter foam cups were used as plant carriers for the plantation to the hydroponic units which means 20.83 plants per square meter. The water height was settled as nearly 7 cm in the troughs at different HLRs. Thus, an air gap of 6 centimeters was created on the shelves of the hydroponic unit of NFT aquaponics. Synthetic fiber was used to fix the plant roots. At the end of the cultivation period, all plants were

removed and measured for length and weight. From each trough, 5 individuals were separated for the determination of leaf areas. The samples were photographed from a vertical view to make measurements of leaf areas using the Image J software program (US National Institutes of Health) (Modarelli et al., 2023). Atmospheric parameters that are directly related to plant growth are obtained from the closest weather station.

Physicochemical parameters in water

Changes in physicochemical parameters of water (temperature, salinity, dissolved oxygen, oxygen saturation, and conductivity (EC)) over the study were measured twice a day, at 09:00 and 17:00, in situ by YSI Pro DSS model handheld multi-parameter device from fish and all plant units. The total suspended solid concentration was determined using the gravimetric method of APHA (1985). Ammonium, nitrite, nitrate, phosphate, sulfate, sodium, potassium, calcium, magnesium, and chloride analyses of water from outlets of all units were performed once a week using ion chromatography (Dionex 3000, Sunnyvale, CA). Chelated iron for plants was maintained at about 2 ppm in all tanks by adding Fe-EDDHA (Doctor Tarsa, Antalya, Türkiye) when needed (Yeşiltaş et al. 2021; Wallace-Springer et al., 2022). pH values were followed by a benchtop instrument in the laboratory (Orion 4 Star, Thermo Scientific, USA).

Statistical analysis

The normality of data was tested using the Shapiro-Wilk test. One-way ANOVA followed by a post-hoc Tukey HSD test was performed for data collected from the troughs for the plant measurements except for final length and leaf number, ion compositions (with some exceptions that are tested with nonparametric analysis), and physicochemical parameters. The final plant length and leaf number of treatments were compared with ANCOVA using their initial values as covariates, followed by the Tukey HSD test for detection of significant treatments. However, the Kruskal-Wallis test was performed for ammonium, nitrite, nitrate, phosphate, and sulfate ions (followed by Dunn's multiple comparison test if required) using JMP 13 Software (SAS Institute Inc., Cary, N. C.). A significance level of $P < 0.05$ was used and the values were given as mean \pm standard deviation. Optimum HLR was estimated based on total yield (kg/m^2) and electricity consumption values (kWh/kg basil) using a three order polynomial regression and a nonlinear broken regression analysis in GraphPad Prism 7 (GraphPad Software, San Diego, CA, USA).

RESULTS

Air quality parameters

Air quality parameters were monitored throughout the study. The average air temperature, average relative humidity, and average wind speed were measured as 27.42 ± 2.82 °C, $46.95 \pm 12.35\%$, and 12.48 ± 3.11 km/h, respectively. Figure 2 shows the general trends of these parameters throughout the experiment. These atmospheric parameters were used to monitor the basil plant needs.

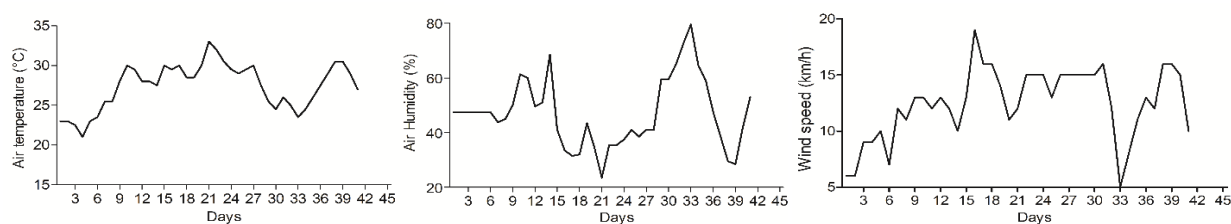


Figure 2. Air temperature, air humidity and wind speed parameters

Water quality parameters

Water temperatures were averaged as 27.19 ± 1.74 (\pm SD) °C in the fish tank, 27.24 ± 1.79 °C in the separation tank, 27.38 ± 1.70 °C in the bio-filter tank, and 26.84 ± 2.54 °C in the sump/pump tank. pH changed between 7.61 ± 0.17 in the fish tank and 7.82 ± 0.13 in the sump/pump tank. The EC and salinity varied between 1.15 ± 0.24 mS/cm and 0.55 ± 0.10 ppt in the sump/pump tank and 1.26 ± 0.24 mS/cm and 0.60 ± 0.11 ppt in the bio-filter tank. Mean dissolved oxygen concentrations were 1.67 ± 0.74 mg/L, 0.81 ± 0.77 mg/L, 0.27 ± 0.24 mg/L, and 6.14 ± 1.10 mg/L in the fish tank, separation tank, bio-filter tank, and sump/pump tank, respectively. The water physicochemical parameters including temperature, EC, salinity, pH, dissolved oxygen, oxygen saturation, and total suspended solids in fish rearing tank, radial flow separator, bio-filter tank, and sump/pump tank showed no significant differences ($p > 0.05$).

The water parameters can be considered favorable for African catfish in the fish tank. However, the dissolved oxygen concentration of the fish tank showed a decreasing trend during the first 9 days, and on the other days of the experiment

dissolved oxygen concentration showed a wavy trend to the end of the experiment (Figure 3). Therefore, large quantities of $\text{NH}_4\text{-N}$, EC, and salinity concentration changes occurred. Then 30 percent of the fish tank water changed with freshwater three times to sustain the remineralization process of the biofilter over the study. The addition of freshwater to the aquaponics, the reason why oxygen concentrations were decreased, caused limited satisfaction for the biofilter. At the first 3 weeks, extremely high levels of ammonia concentration in aquaponics because the system was not functioning optimally. In the 4th week of the experiment, the biofilter started to convert large amounts of ammonia to nitrite and nitrate. At this stage of the study nitrate levels of different units of aquaponics were at the highest concentrations (Figure 4). Total suspended solids were determined as 0.19 ± 0.08 mg/L in the fish tank, 0.10 ± 0.06 mg/L in the separation tank, 0.16 ± 0.20 mg/L in the bio-filter, and 0.06 ± 0.06 mg/L in sump/pump tanks (Figure 3). Total suspended solids showed a meaningful relationship with the dissolved oxygen concentrations of the aquaponic system. Average physicochemical parameters in different groups of HLRs have been shown in Table 1.

Table 1. Average physicochemical parameters in hydroponics in different HLRs

HLR	Temperature (°C)	EC (mS/cm)	Salinity (ppt)	Dissolved oxygen (%)	Oxygen concentration (mg/L)
2 ($\text{m}^3/\text{m}^2/\text{day}$)	27.49 ± 1.70^a	1.27 ± 0.24^a	0.60 ± 0.11^a	3.42 ± 2.89^a	0.29 ± 0.25^a
4 ($\text{m}^3/\text{m}^2/\text{day}$)	27.45 ± 1.69^a	1.27 ± 0.24^a	0.60 ± 0.11^a	3.42 ± 2.89^a	0.30 ± 0.25^a
8 $\text{m}^3/\text{m}^2/\text{day}$)	27.42 ± 1.71^a	1.27 ± 0.24^a	0.60 ± 0.11^a	3.42 ± 2.89^a	0.31 ± 0.25^a
12 ($\text{m}^3/\text{m}^2/\text{day}$)	27.39 ± 1.71^a	1.24 ± 0.23^a	0.60 ± 0.11^a	3.42 ± 2.89^a	0.31 ± 0.25^a

An initial fish of $10 \text{ kg}/\text{m}^3$ was almost quadrupled up to $37.3 \text{ kg}/\text{m}^3$ at the end of the study. The high stock density appears to have played a major role in low oxygen concentrations in the present study. However, even under these low oxygen concentrations fish performance was very well in terms of SGR and FCR.

There were no statistically significant differences found in mean ammonium concentrations, nitrite-nitrogen concentrations, nitrate-nitrogen concentrations, and phosphate-phosphorus concentrations between different units ($p > 0.05$), but some significant differences were detected among the weeks ($p < 0.05$) (Figure 4).

By the 4th week of the study, relatively low pH affected the ammonia-oxidized bacteria however, after the 4th week of the experiment when pH reached from 7.5 to 8 ammonia oxidation process accelerated (Figure 3, Figure 4). In the 6th week of the experiment, increasing ammonium nitrogen levels in the

system pointed out the poor performance of the biofilter tank again, and the experiment ended. Average nitrite concentrations were determined as 1.03 ± 0.33 mg/L in the fish tank, 1.41 ± 0.66 mg/L in the separation tank, 0.95 ± 0.33 mg/L in the bio-filter tank, and 1.36 ± 0.55 mg/L in the sump/pump tank. Average nitrate concentrations were determined as 13.04 ± 8.19 mg/L in the fish tank, 17.65 ± 12.79 mg/L in the separation tank, 7.37 ± 3.25 mg/L in the bio-filter tank, and 10.65 ± 5.68 mg/L in the sump/pump tank. In the 4th week of the experiment, the lowest ammonium concentrations we had in all units, and the highest nitrate concentrations were detected in the system, the typical indication of completion nitrification process with accompanying decrease in nitrite concentrations.

During the last week of the experiment, 500 L/day water exchange was applied to compensate for low oxygen level in aquaponics tanks which reflected in average ammonium and phosphate concentrations as an increase.

The lack of an aerator affected the biofilter efficiency in a negative way. However, nitrite and nitrate levels in aquaponics units decreased (Figure 4). A disappearance of a significant amount of nitrification bacteria in the system appeared to be the case as a result of the compulsory water changes on the days of the 18th, 27th, and 35th of the experiment. This may also be a reason for the increase in ammonia levels in different units over the last week of the study (Figure 4). The increase in water exchange also resulted in an elevation in calcium, potassium,

and phosphate concentrations.

Fish growth parameters

The final mean weight of the fish was determined as 784.26 ± 207.13 g. Over the study period, only 8 fish died resulting in only 6.25% mortality. The SGR of fish was 3.31 %/day and relative weight gain was 3.08% although some water quality problems were experienced during the study. The growth rate of fish was calculated as 273%.

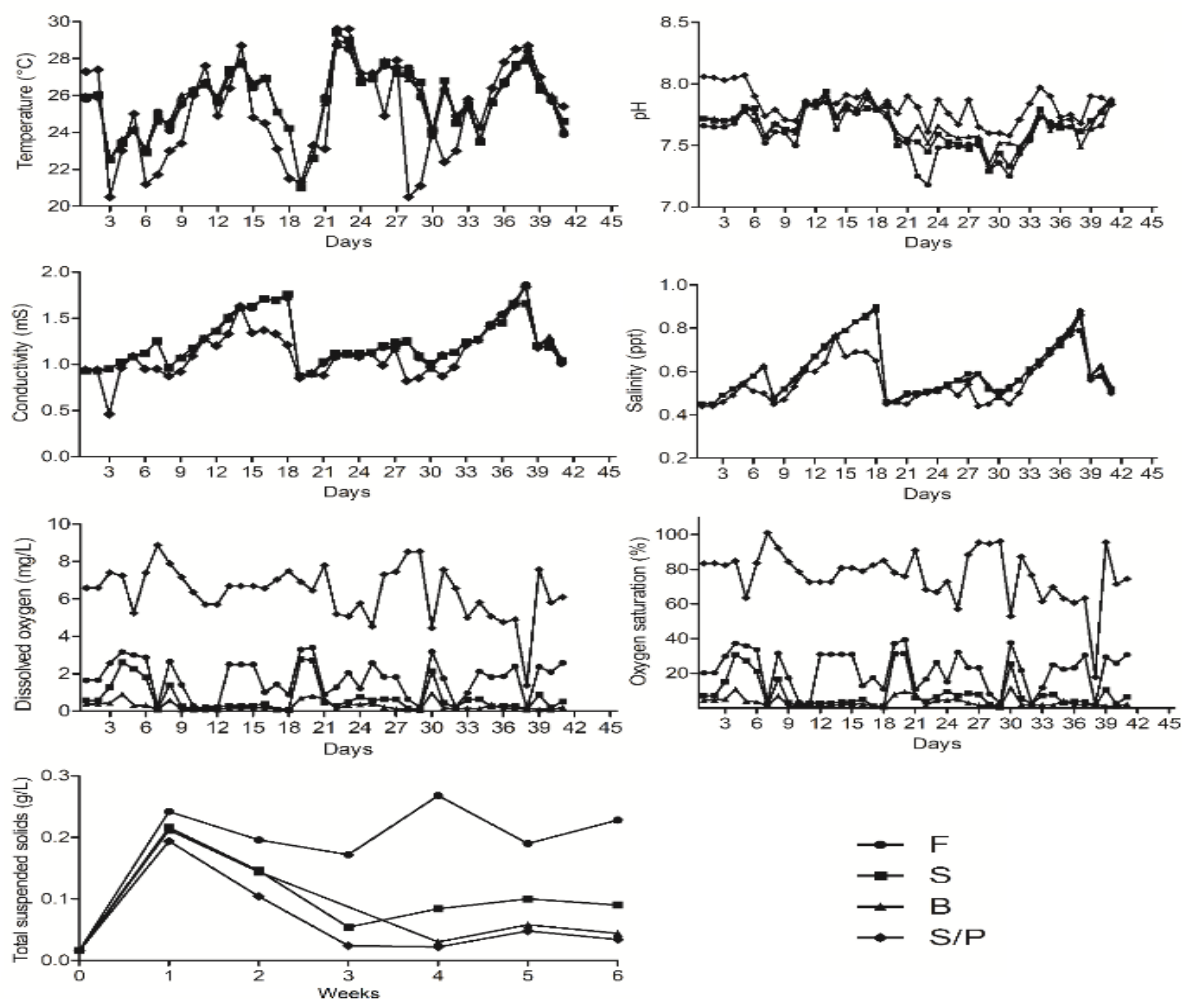


Figure 3. Physicochemical parameters in Aquaponics (F: Fish tank, S: Separation tank, B: Bio-filter tank, S/P: Sump/Pump tank)

Plant growth parameters

The tallest basil plants were in the group of $12 \text{ m}^3/\text{m}^2/\text{day}$ HLR with an average value of 196.0 ± 35.7 mm whereas the shortest plants were in the HLR group of $2 \text{ m}^3/\text{m}^2/\text{day}$ with an average value of 173.0 ± 48.0 mm. The growth difference between these treatments was statistically insignificant ($p > 0.05$). Similarly, the best and worst groups in terms of plant weight were 4 and $2 \text{ m}^3/\text{m}^2/\text{day}$ respectively, which were statistically significant with the values of 23.0 ± 2.5 g and 15.5 ± 1.7 g, respectively ($p < 0.05$). There were no statistically significant differences among all groups ($p > 0.05$) in terms of average leaf number with the highest value of 44.5 ± 24.0 in the HLR of $4 \text{ m}^3/\text{m}^2/\text{day}$ (Table 2).

Average stem diameters of basil plants insignificantly changed among the treatments ($p > 0.05$) with the maximum and minimum values in 4 and $2 \text{ m}^3/\text{m}^2/\text{day}$ HLR treatments, respectively. The highest average leaf area was detected as $1314 \pm 145 \text{ mm}^2$ in the HLR of $4 \text{ m}^3/\text{m}^2/\text{day}$ whereas the lowest in the HLR of $4 \text{ m}^3/\text{m}^2/\text{day}$ with $870 \pm 71 \text{ mm}^2$ leaf area ($p < 0.05$). The annual plant yield changed between 4.66 ± 0.33 and 7.06 ± 1.66 kg without significant differences in the 8 and $4 \text{ m}^3/\text{m}^2/\text{day}$ HLR groups, respectively ($p > 0.05$). The best performing HLR group in terms of plant growth in the present study was $4 \text{ m}^3/\text{m}^2/\text{day}$, which could be due to better uptake of the nutrients by the plants.

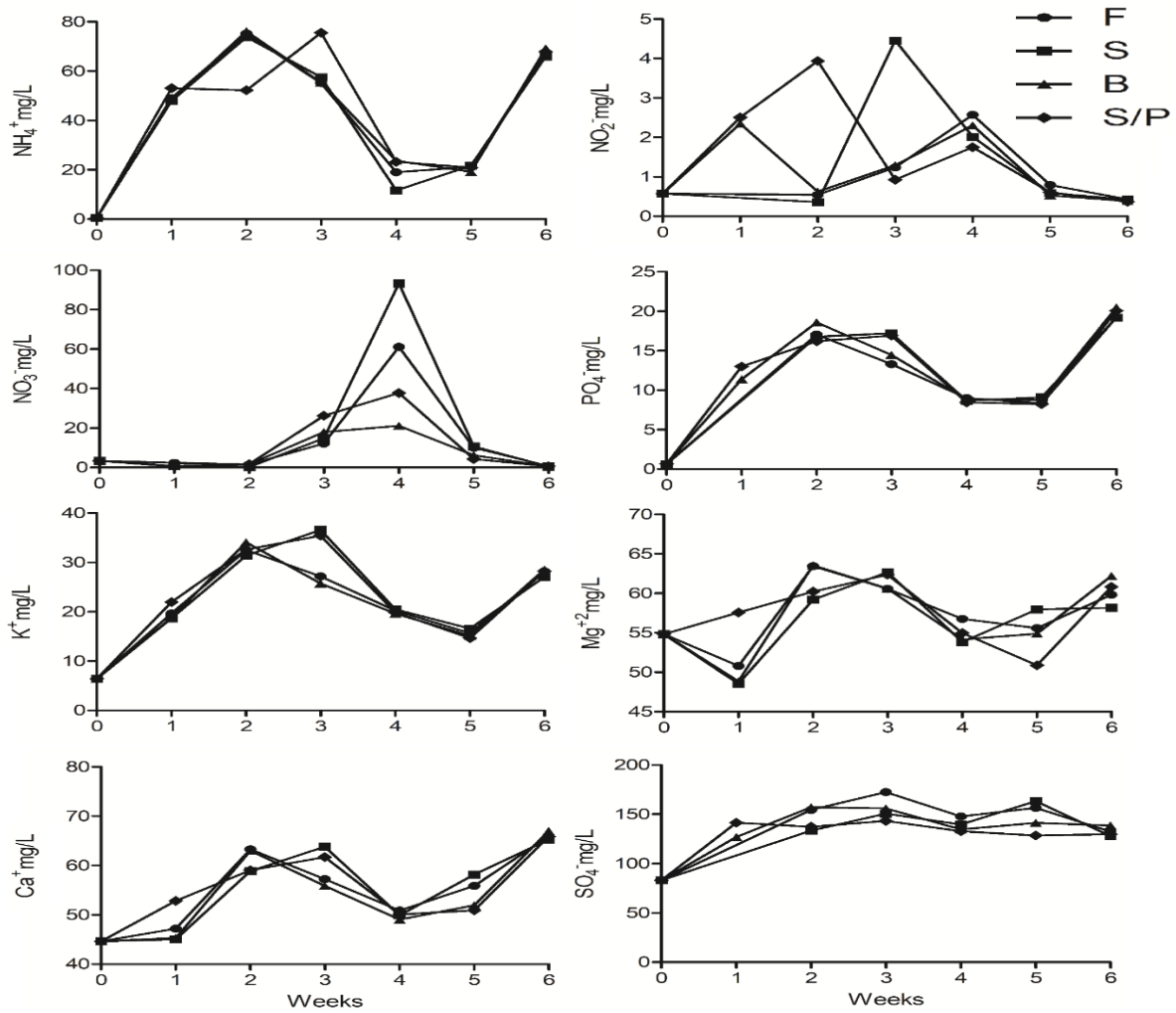


Figure 4. Ion concentrations in Aquaponics (F: Fish tank, S: Separation tank, B: Bio-filter, S/P: Sump/Pump)

Table 2. Average plant growth parameters with standard deviations

HLR	IPL (mm)	FPL (mm)	IPW (g)	FPW (g)	ILN	FLN	ISD (mm)	FSD (mm)	ILA (mm ²)	FLA (mm ²)
2 (m ³ /m ² /day)	57.3±6.4 ^a	173.0±48.0 ^a	0.1±0.0 ^a	15.5±1.7 ^a	3.5±0.9 ^a	38.0±18.4 ^a	1.3±0.0 ^a	5.6±0.3 ^a	63.7±3.3 ^a	870±71 ^a
4 (m ³ /m ² /day)	59.2±7.1 ^a	195.9±60.9 ^a	0.1±0.0 ^a	23.0±2.5 ^b	3.6±0.8 ^a	44.5±24.0 ^a	1.2±0.0 ^a	5.7±0.2 ^a	59.4±3.7 ^a	1314±145 ^b
8 (m ³ /m ² /day)	63.2±5.0 ^a	175.6±32.5 ^a	0.1±0.0 ^a	15.7±1.5 ^{ab}	3.6±0.8 ^a	36.4±19.6 ^a	1.2±0.0 ^a	5.3±0.2 ^a	59.4±3.0 ^a	1119±95 ^{ab}
12 (m ³ /m ² /day)	61.6±6.5 ^a	196.0±35.7 ^a	0.1±0.0 ^a	19.9±2.1 ^{ab}	3.9±0.1 ^a	40.9±17.8 ^a	1.2±0.0 ^a	5.5±0.2 ^a	62.4±3.7 ^a	1167±146 ^{ab}

IPL: Initial plant length, FPL: Final plant length, IPW: Initial plant weight, FPW: Final plant weight, ILN: Initial leaf number, FLN: Final leaf number, ISD: Initial stem diameter, FSD: Final stem diameter, ILA: Initial leaf area, FLA: Final leaf area

Yields of basil ranged as 0.32±0.04 kg/m², 0.47±0.14 kg/m², 0.33±0.03 kg/m², and 0.41±0.13 kg/m² in the 2, 4, 8, and 12 m³/m²/day HLRs, respectively without significant differences (p>0.05). HLR treatments of 2, 4, 8, and 12 m³/m²/day consumed average electricity of 20.54±2.02, 30.72±14.0, 79.50±9.48, and 102.9±34.8 kW/kg basil, respectively, with significant differences among the treatments. The third-degree polynomial regression between HLR and total yield (kg/m²), although not strong (R²=0.28; p=0.42), suggested the best hydraulic loading rate as 4.41 m³/m²/day

(Figure 5). A broken line regression model for energy consumption per kg of basil production generated a breakpoint at 4 m³/m²/day (Figure 5), which suggests increasing HLR above 4 m³/m²/day will result in excessive energy expenditure for basil production.

Energy consumption of HLRs of 8 and 12 m³/m²/day was significantly higher than that of 2 m³/m²/day (Figure 6). 4.41 m³/m²/day HLR was determined as the best for energy consumption and productivity.

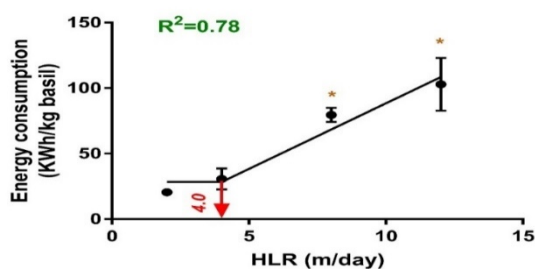


Figure 5. Relationship between HLR and energy consumption per kg of basil production in an aquaponics system (Values with * are significantly different from the treatment of 2 m³/m²/day based on Dunnet's test)

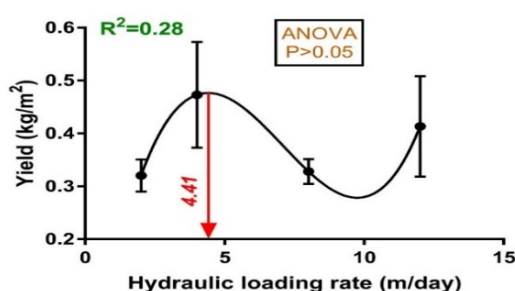


Figure 6. Relationship between HLR and basil yield in an aquaponics system

DISCUSSION

Water quality parameters

It was found that the physical and chemical parameters of the water were relatively adequate for the growth of *O. basilicum* and *C. gariepinus* (Knaus et al., 2020a; Pasch et al., 2021). The hydraulic loading rate is another important factor in the growth of basil plants. But only limited data is available on basil and HLRs in aquaponics production. But dissolved oxygen concentration in aquaponic system caused major difficulties on nitrifying bacteria effectiveness. The water temperature and the lack of aeration in the system caused the low wavy trend of oxygen concentrations in aquaponics. Low oxygen concentrations and pH of water in aquaponics restricted the nitrifying bacteria metabolism to convert ammonia to nitrate, effectively (Tyson et al., 2004). pH in aquaponics was measured as between 7 to 8 which are suitable for nitrification bacteria (Timmons et al., 2002) and African catfish (Endut et al., 2010). The water quality parameters are consistent except for oxygen concentration and saturation with those reported by Knaus et al. (2020b) for aquaponic systems. Low oxygen is a frequent reason for fish death, but African catfish can practice aerial as well as aquatic respiration (Bovendeur et al., 1987). African catfish was reported to be highly tolerant to low dissolved oxygen as low as 0.5 mg/L (Boyd and Tucker, 1998) and able to grow well even in low oxygen levels (Akinwale and Faturoti, 2007; Ibrahim and Nagggar, 2010). Average oxygen saturation levels were found as 21.16±8.39% in the fish tank, 10.03 ± 9.05% in

the separation tank, 3.40±2.90% in the bio-filter tank, and 76.26±11.09% in the sump/pump tank.

In water with high levels of ammonia, nitrite, nitrate and urea, African catfish is known to be highly tolerant (Figure 4) (Bakar et al., 2015; Ip et al., 2004). Short-time NH₄-N increase in aquaponics can be acceptable (Zou et al., 2016), however in prolong duration of excess NH₄-N concentration might be toxic for fish (Yang, 2019). 80 mg/L of NH₄-N and 10 mg/L of NO₂-N were determined as upper lethal limits for African catfish in Recirculating Aquaculture Systems (Palm et al., 2018). Aquaponic systems integrated with African catfish including a wide variety of ammonium concentrations such as 0.9-1.8 mg/L by Su et al. (2020), 0.91±0.26 mg/L by Baßmann et al. (2017), and 20.46±6.53 mg/L by Knaus et al. (2020b) were compared to this study. When the concentrations in the units are considered, average ammonium concentrations were measured as 41.04±10.51 mg/L in the fish tank, 39.88±10.78 mg/L in the separation tank, 41.73±10.58 mg/L in the bio-filter tank, and 41.91±10.40 mg/L in the sump/pump tank, being higher than the findings of previous studies. In this study, these nitrogenous compounds' upper limits were not exceeded even without aeration in the system.

Nitrite and nitrate concentrations in the system were compatible with those reported by Oladimeji et al., (2020), and Yang and Kim (2020a). Mean phosphate concentrations were found as 11.21±2.33 mg/L in the fish tank, 11.73±2.45 mg/L in the separation tank, 11.88±2.54 mg/L in the bio-filter tank, and 11.94±2.50 mg/L in the sump/pump tank (Figure 4), being comparable to the results of other studies (Villarreal et al., 2011; Strauch et al., 2019; Yang and Kim, 2020a).

Not only temperature, pH, and hydrogen ions in aquaponics but also biofilter activation affected the ammonium concentrations in the system (Yavuzcan Yildiz et al., 2017). Ammonia levels of aquaponics in all units decreased at the 4th week of the experiment thanks to the activation of ammonia-oxidized bacteria (Su et al., 2020). The poor oxygen concentration related to no aeration in the system affected the used previous aquaponics' biofilter tank in a negative way. Low level of dissolved oxygen in aquaponics caused low pH as well. And pH directly affects the nitrification process in the aquaponics (Tyson et al., 2004). Average magnesium, potassium, calcium, and sulfate concentrations that directly affect plant production were found compatible with the other aquaponics studies integrating basil in aquaponics (Baßmann et al., 2018; Knaus et al., 2020a) (Figure 4).

The main aim of the biofilter tank is to enhance the oxidation of NH₄-N and NO₂-N in the recirculating water of the aquaponics (Kasozi et al., 2021). However, adequate biofiltration could not be achieved well because of the hot-summer Mediterranean climate, the increase in the amount of ammonium in parallel with the increase in fish biomass, and not including aeration in the system. In this study, the main aim was to produce fish and plants together with limited energy consumption in hot climate conditions by trying to keep the costs to a minimum.

Air parameters

The environmental factors, air temperature, relative humidity, and wind speed affected the water temperature of aquaponics directly (Figure 2, Figure 3). These parameters have been found compatible with the recordings of studies by Ghamarnia et al. (2014) and Ferrarezi and Bailey (2019), who maintained the basil in an aquaponics system and soil agriculture in a semi-arid climate. For the basil plant, the air temperature met the optimum temperature requirement throughout the study (Chang et al., 2005; Barickman et al., 2021). Air humidity changed between 20% to 80% throughout the study which is suitable for required for the optimum growth of basil (Solis-Toapanta et al., 2020; Lin et al., 2021). Wind speed (air velocity) parameters recorded between 0-20 km/h that is founded as very low compared to other studies (Cohen and Ben-Naim, 2016).

Fish growth

African catfish is a very sturdy fish with the ability to survive in poor water quality conditions and therefore it has a good survival rate in recirculating aquaculture systems (Akinwole and Faturoti, 2007). 25–30 °C of water temperature is specified as the optimum temperature range for the catfish culture (Putri et al., 2021). Water temperature in the system was within the optimum range for the growth of African catfish (Hogendoorn et al., 1983). Despite various water conditions, the African catfish is highly tolerant. The growth of *C. gariepinus* was satisfactory throughout the study. The feed conversion ratio (FCR) was recorded as 0.695, being consistent with our previous study by Yeşiltaş et al. (2021) and better than those reported for the same species by Endut et al. (2010) with 1.23-1.39, Palm et al. (2014) with 1.00, Baßmann et al. (2017) with 1.02, and Knaus et al. (2020a) with 0.74-0.91. Therefore, the overall growth and nutrient utilization performance of fish in this study are quite acceptable when compared with literature findings (Baßmann et al., 2017; Endut et al. 2010; Palm et al., 2014; Enyidi et al., 2017; Knaus et al., 2020a).

Plant growth

Basil adapted very well to the aquaponic system run under the summer climate conditions of the Eastern Mediterranean. During the experiment, basil seedlings in all rafts grew well and seemed healthy. Lower or higher HLRs than 4 m³/m²/day seemed to have resulted in lower growth performance in basil in an NFT aquaponics system. Nitrogen is assumed to be the major nutrient in the aquaponics systems that influence plant growth. At low HLRs, the plant growth can be weakened due to the formation of anoxic zones with the low water current and the development of denitrification which leads to N losses from the system (Endut et al., 2010). Conversely, higher HLRs than the optimum can reduce the contact time of nutrients with the plant roots (Shete et al., 2016; Yang and Kim, 2020a), and increase the energy expenditure for each unit of plant production (Yang and Kim, 2020a).

Nutrient concentration and salinity stress in aquaponics affected the yield of the basil. Increasing trend of ammonia

nitrogen, nitrite nitrogen, and phosphate phosphorus in the first three weeks and then increasing nitrate concentration in the 4th week of the experiment contributed to the growth of basil plant biomass. Electrical conductivity was not the main factor that is affecting the basil plant biomass, however, anion and cation combinations were playing important roles in obtaining a better yield (Yang and Kim, 2020b). Increasing ammonium concentrations in aquaponics in the first three weeks did not contribute as expected to basil biomass production when this data was compared to other studies (Rakocy et al., 2003). At the end of the study, the total average yield of the basil plant in the aquaponics was calculated as 311.7±54.7 g/m², 463.9±154.1 g/m², 306.1±30.47 g/m², and 413.6±134 g/m² in different HLR ratios, 2, 4, 8, and 12 m³/m²/day, respectively. These results were much lower than the studies conducted in the deep water technique in the literature (Rakocy et al., 2004; Rodgers et al., 2022).

Ammonium nitrogen concentration was in increasing trend in the first three weeks due to the lack of nitrifying bacteria in the biofilter. One of the other reasons for the high concentration of ammonium in water can be related to the low ammonium intake of basil plants (Nurzyńska-Wierdak et al., 2011). Nitrate concentration in aquaponics reached nearly 100 mg/L and it was higher than Roosta (2014) with a value of 34.6±3.1 mg/L. The maximum nitrate concentrations were in aquaponics, in the sump/pump tank with a range of nearly 20 to 100 mg/L.

Generally, the oxygen concentration in the root zone of plants is considered essential. Thanks to its less sensitivity to oxygen availability in water basil showed a good growth performance at low HLRs (Puccinelli et al., 2021). By using the advantage of NFT aquaponics, plant roots had enough space to respiratory from the air in the rafts. Plant length, weight, stem diameter, leaf area, and leaf number are important growth factors for basil. 27 ± 1 °C average water temperature in all tanks of the study have affected in good way to the leaf areas (Chang et al., 2005). In this study, upper and lower basil production temperatures were not exceeded.

It is difficult to compare these results with the literature findings due to different expressions used for the period per unit of yield. For instance, basil yield was founded as 42 kg/m² year by Savidov et al., (2005), 14.91 kg/m² year by Ferrerazi and Bailey (2019) and 15.2 kg/ m² year by Yang and Kim (2020c). Conversion of yield to a year production in the present study is impossible since the system used was outdoor that was open to seasonal environmental changes. A comparison of the treatments in the present study with the literature from an energy consumption perspective could be more reasonable.

Higher electricity consumption was calculated as 162.10 kWh/kg basil/year by Xie and Rosentrater (2015), who conducted a study on life cycle assessment and techno-economic analysis of tilapia-basil aquaponics.

CONCLUSION

The effectiveness of four different HLRs in an NFT aquaponics system was tested under the summer climate

conditions of the Eastern Mediterranean. African catfish and basil were successfully produced without aeration in very low oxygen concentrations in aquaponics. Energy savings were achieved by not using aeration. The optimum HLR for basil in the aquaponic system was between 4 and 4.41 m³/m²/day when the assessment was based on basil yield and energy consumption for basil production. The finding is important in terms of maximization of basil production with acceptable energy expenditure in the aquaponics. African catfish in the system showed excellent growth and feed utilization performance even in low oxygen concentrations. African catfish and basil seem a very good couple for NFT aquaponic systems thanks to their thermo-tolerant characteristics under the Eastern Mediterranean climate conditions. This type of system and HLR may be recommended to those locations that are struggling with drought and limited energy.

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CREDIT AUTHORSHIP CONTRIBUTION STATEMENT

Murat Yeşiltaş, Hüseyin Sevgili, Mehmet Ali Turan Koçer, and Edis Koru contributed to project. Murat Yeşiltaş, Mehmet Ali Turan Koçer, and Hüseyin Sevgili contributed to perform experiment. Murat Yeşiltaş and Hüseyin Sevgili contributed to data analyses, interpretation and manuscript writing.

DECLARATION OF INTEREST

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

ETHICS APPROVAL

Ethics approval for this research was obtained through Mediterranean Fisheries Research Production and Training Institute Local Ethics Committee for Animal Experiments (212809), and consent forms were signed by all participants.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are openly available in Ege Journal of Fisheries and Aquatic Sciences.

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