Role of stocking density of tilapia (*Oreochromis aureus*) on fish growth, water quality and tomato (*Solanum lycopersicum*) plant biomass in the aquaponic system

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Abstract— The present study reports the results of the production of Nile tilapia (*Oreochromis aureus*) and tomato (*Solanum lycopersicum*) in the classical aquaponic system (one-loop) with different fish density. The experiment as the first scientific aquaponics study in Turkey was conducted at the Ankara University, Faculty of Agriculture, using in-door, small-scale classical aquaponic systems. Ninety six tilapia juveniles (*O. aureus*) were stocked at different ratio; 25 kg/m$^3$ (Group I), 35 kg/m$^3$ (Group II) and 50 kg/m$^3$ (Group III) and fed with 45% raw protein feed at the level of 2% body weight for 126 days. Fish density affected the fish growth parameters and the most densest group showed the best results in terms of fish growth and feed efficiency. Water quality parameters measured fluctuated during the experiment even the exceed of the optimal ranges for the fish. However, tilapia tolerated the changes of water quality. Total plant biomass was low with the various limiting factors including insufficient lighting of in-door aquaponics system and low level of water potassium. The results of this study clearly illustrate the fish stocking rate has an impact on total biomass in the aquaponics and in one-loop aquaponics the water quality fluctuation is the main challenging factor.

Keywords— aquaponics, tilapia, tomato, fish growth.

1. INTRODUCTION

One of the main challenges of agriculture in 21th century to feed the growing population is finding more efficient and sustainable food production systems and adapting to climate change. There is also a gap in the availability of freshwater and land to increase the yield with minimal environmental effect [1]. To overcome the problems that the world is facing such as water scarcity, soil degradation, climate change and the population increase the aquaponics appear an alternative solution as the aquaponics are an environmental friendly and sustainable food production system [2,3].

Aquaponics, basically, the symbiotic growing of fish and vegetables in recirculating water systems is emerging as one of the most important areas of sustainable agriculture. Aquaponics is the systems that integrating aquaculture recirculating production systems with hydroponics. With aquaponics dual production of both fish and plants is possible by using the water from the fish tanks for plant growth. The essential elements of an aquaponic system consists of fish rearing tank, a suspended solid removal component, a biofilter, a hydronic component and a sump [4]. In the aquaponic system, nutrients, which are excreted directly by the fish or generated by the microbial breakdown of organic wastes, are absorbed by plants cultured hydroponically. Through microbial decomposition, the insoluble fish metabolite and unconsumed feed are converted into soluble nutrients which then can be absorbed by plant [5]. Fish feed provides most of the nutrients required for plant growth [6]. Aquaponics work on the principle of nitrogen cycle, where in dissolved waste generated from the production system is effectively converted to plant nutrients by beneficial nitrifying bacteria. Plants can utilize these nutrients for their growth [6, 7, 8]. Plants in hydroponics and aquaponics grow more rapidly compared to their counterparts which grow in the soil because the root system is in direct contact with nutrients and nutrient uptake is more efficient in an aqueous phase [9]. Water, energy and fish feed are the three main physical inputs for aquaponic systems although the aquaponic operations vary in size and type of production system [10]. Palm et al. [11] highlighted that economic sustainability of aquaponics depends on a variety of factors including system and feed design, animal welfare and pathogen control. There is a need to establish the macro- and micronutrient proportion that fish can release in the water for a given feed in a given system; this depends on fish species, fish density, temperature, and type of plants [12]. It is clear that feed and stocking rate of fish are directly related and to maintain the balance between metabolic products the
stocking rate is critical in the aquaponics as a reflection of feed. Therefore, the present study was carried out to assess the production of Nile tilapia (Oreochromis aureus) and tomato (Solanum lycopersicum) in the aquaponic system with different fish density.

II. MATERIAL AND METHODS
This research was carried out in the small-scale aquaponic system with a grow bed form, producing tilapia (O. aureus) and tomato (S. lycopersicum) in Ankara University, Faculty of Agriculture, Department of Fisheries and Aquaculture. Aquaponic system was installed in-door.

The protocol for the experiment was approved by the ethics committee of the Ankara University with the reference number of 2014-2-9.

Experimental set up
Ninety six tilapia juveniles (O. aureus) were stocked at different ratio: 25 kg/m³ (Group I), 35 kg/m³ (Group II) and 50 kg/m³ (Group III). Individual fish weight was 5-7 g at the beginning of the experiment. Fish were fed with commercial rainbow trout feed with 45% raw protein with 2% body weight for 126 days. Chemical composition of the feed is presented in Table 1. The aquaponics experimental system comprises of a nine fish tank (80x60x50 cm) and nine plastic tanks (65x40x35 cm) filled with hydraton for vegetable beds. Each vegetation tank planted with 4 plantlet (30-35 days old) of tomato (S. lycopersicum). Each fish tank was filled with 100 L of tap water and aerated continuously with air stone. Nitrifying bacteria; Nitrosomonas europaea and Nitrobacter winogradskyi were added to the system at the initial period. Experiments were run in three replicates. A lighting system made of eight Ostram HO 80w/865 lumilux cool daylight fluorescent lamps was placed above the units. Water loss due to sampling and evaporation was replenished with the addition of distilled water.

Analytic procedures
After 126 days of rearing the fish was harvested and their growth performance was measured with the parameters using the formulas as below.

i) Feed Conversion Ratios (FCR): FCR= food intake/ weight gain

ii) Protein efficiency ratio (PER): (PER) = (Wt-Wt0)/crude protein fed

iii) Feed efficiency (FE): FE= weight gain/food fed

iv) Specific growth rate (SGR%): SGR%= (lnWt – lnWt0 x 100) / t-t0

v) Average daily gain (ADG): ADG% = 100[Wt-Wt0/Wt x (t-t0)]

where, Wt=Mean final fish weight, Wt0 = Mean initial fish weight and t-t0 = number of days on feed

vi) Daily growth index DGI (%): DGI%= (final weight1/3 - initial weight1/3 ) x100/day

Table 1: Chemical composition of the feed

| Component (%) | Protein | Digestible energy kcal/kg | Lipid | Metabolic energy kcal/kg | Moisture | Vitamin A IU/kg | Ash | Vitamin D IU/kg | Cellulose | Vitamin E IU/kg | Nitrogen free extract % | Phosphorus | GE (Gross energy) kcal/kg | Labile P | Vc IU/kg |
|---------------|---------|----------------------------|-------|--------------------------|----------|----------------|-----|----------------|-----------|----------------|------------------------|------------|----------------------------|----------|---------|
|               | 45.0    | 4125                       | 20.0  | 3742                     | 8.5      | 5.000         | 11.0| 1.500          | 3.0       | 100           | 12.5                   | 1.5        | 5124                       |

At the end of the experiment, plant (S. lycopersicum) parts were weighted separately (as leaf, stem and root) for determination of fresh and dry weight. For measuring dry weight of the plant samples was dried in 65 ºC for 3 days.

Water Quality Measurements
Water quality parameters in fish tanks were routinely measured. During the experimental period the water temperature was kept at 23°C. Dissolved oxygen (DO), temperature (T) and pH were measured every week with portable equipments. Other water quality parameters; ammonia (NH3), Nitrat (NO3-), Nitrit (NO2-) and potassium (K) were measured every 15 days by using Standard Methods [13].

Statistical Analysis
This experiment were conducted as completely randomized design with three replicates. Data were analyzed by analysis of variance (ANOVA) with the SAS package. Duncan’s multiple-range test was used to compare differences among individual means. Treatment effects were considered significant at p<0.05. Percentage and ratio data were transformed to arcsine values prior to analysis[14].

III. RESULTS
Growth and production of tilapia in the aquaponic system are given in Table 2. The mean group weight gain was 544.1±57.9 in Group I (stocking rate: 25 kg/m³), 849.7±30.8 in the Group II (stocking rate: 35 kg/m³) and 1003.3±49.8 for Group III (stocking rate: 50kg/m³). The differences in mean group weight gain were statistically significant (p < 0.05) and the highest weight gain was in Group III with the highest fish density. Feed conversion ratio (FCR) differed among the groups (p < 0.05) however,
the FCR was similar in Group II and III. The FCR was higher in Group I than that of Group II and III. Thus, feed efficiency (FE) was lower in Group I. Protein efficiency ratio (PER) showed significant differences among the groups. PER was the lowest in Group I and the highest in Group III. Specific growth rate was higher in Group III. Average daily growth was the highest in Group III with the value of 12.83±0.829 %. Daily growth index (DGI) differed among the groups (p < 0.05) and the minimum DGI percentage was in Group I. Survival rate showed significant differences among the groups (p < 0.05) and was the highest in Group II.

### Table 2: The growth parameters of tilapia (O. aureus) in the aquaponics system by the stocking ratio

| Growth Parameters                          | Experimental groups                      |
|--------------------------------------------|------------------------------------------|
|                                            | Group I Stocking rate: 25 kg/m³           | Group II Stocking rate: 35 kg/m³           | Group III Stocking rate: 50 kg/m³          |
| Mean group initial body weight (g)         | 44.96±1.08b                              | 68.73±0.994a                              | 70.07±3.18a                               |
| Mean group final body weight (g)           | 589.0±58.4b                              | 918.4±31.8a                               | 1073.4±50.0a                              |
| Mean group weight gain (g)                 | 544.1±57.9c                              | 849.7±30.8b                               | 1003.3±49.8a                              |
| Food consumed (g)²                         | 621.8±12.0c                              | 788.9±12.1b                               | 913.8±23.9a                               |
| Feed conversion ratios (FCR)¹              | 1.1600±0.077a                            | 0.9300±0.023b                             | 0.9133±0.0406b                            |
| Feed efficiency (FE)¹                      | 0.8710±0.0618b                           | 1.0758±0.0257a                            | 1.0977±0.0516a                            |
| Protein Efficiency Ratio (PER)¹            | 11.828±1.26c                             | 18.471±0.669b                             | 21.812±1.08a                              |
| Specific Growth Rate (SGR %)               | 2.2891±0.0763c                           | 2.3138±0.0176b                            | 2.4366±0.0533a                            |
| Percentage average daily growth (ADG %)    | 10.788±1.02b                             | 11.030±0.238b                             | 12.833±0.829a                             |
| Daily growth index (DGI %)                 | 4.294±0.228c                             | 5.0193±0.0815b                            | 5.4583±0.145a                             |
| Survival (%)                               | 80.952±9.52a                             | 96.970±3.03b                              | 85.714±10.9a                              |

*Values with different superscripts in a row differ significantly (p<0.05)
1 Expressed as the percent of the initial body weight after 126 days.
2 Moisture-free basis.

The tomato (S. lycopersicum) plant biomass as fresh and dry weight of tomato plant leaf, stem and root branches were presented in Table 3. Significant differences were observed in the fresh weight and dry weight of tomato plant (p < 0.05). Final total weight values were the maximum in Group III. Fresh and dry weight of total plant correlated with fish density (R²=0.92).

### Table 3: Biomass of tomato (S. lycopersicum) plants grown in the aquaponic system by fish stocking density groups.

| Group | Leaf | Stem | Root | Total | Leaf | Stem | Root | Total |
|-------|------|------|------|-------|------|------|------|-------|
| I     | 1252.5 | 621.6 | 131.2 | 2005.3a | 192.5 | 66.1 | 20.4 | 278.9a |
| II    | 1405.9 | 902.6 | 90.0 | 2398.5b | 210.0 | 95.9 | 14.0 | 326.0b |
| III   | 1728.1c | 1108.3 | 139.8 | 2976.2c | 265.6 | 117.8 | 21.7 | 405.1c |

*Different letters in a column indicate significant differences (p < 0.05) among the groups.

Water quality parameters measured in the experiment (DO, pH, ammonium, nitrite, nitrate, potassium) are presented in the Fig 1. Water quality parameters except water temperature showed significant differences by the time (p<0.05) and the experimental groups (p<0.05). During the experimental period the water temperature was kept around 24-25°C. The range of pH was between 5.83 and 7.31 in Group I, 5.60-7.22 in Group II and 5.50-7.12 in Group III. Dissolved oxygen level providing with artificial aeration ranged between 5.80 mg/L (min) and 7.13 mg/L (max). Ammonium levels during the experiment varied between 0.68 and 3.70 mg/L in Group I, 0.15 and 3.49 mg/L in Group II and 0.40 and 2.92 mg/L in Group III. Nitrite levels were between 0.05 and 0.80 mg/L in Group I, 0.16 and 0.90 in Group II and 0.10 and 0.53 mg/L in Group III. Nitrate levels ranged from 1.85
to 275 mg/L in Group I, from 2.33 to 419 mg/L Group II and from 2.38 to 400.93 mg/L in Group III. Potassium values in water ranged from 0.13 to 0.36 meq/L in Group I, from 0.10 to 0.37 meq/L in Group II and from 0.10 to 0.38 meq/L in Group III.

IV. DISCUSSION

In this aquaponic system, three different stocking rate of tilapia were analysed for i) Feed Conversion Ratios (FCR), ii) Protein efficiency ratio (PER) iii) Feed efficiency (FE) iv) Specific growth rate (SGR %), v) Average daily gain (ADG), vi) Daily growth index (DGI (%)) and all fish were fed with the same feed containing 45% raw protein. We observed that the growth parameters were better in the group having the maximum fish density with 50 kg/m³. Total plant biomass values were also better in the group of 50 kg/m³ than the groups of 25 and 35 kg/m³. Nevertheless, tilapia in oxygenated water can be grown at the 120 kg/m³ by providing better nutrient supply [15]. FCR as one of the most important parameters in terms of economy of the aquaponic system should optimize in parallel to fish density and feeding ratio. Thus, in our case, the minimum FCR was observed in the group
of the highest stocking rate (50 kg/m³) with the feeding ratio of 2% of total body weight daily. The average FCR as 1.2-1.3 in the couple system with 40 kg fish/m³ in the study of Monsees et al. [13] was considered as favourable for the commercial aquaculture. Endut et al. [16] reported that feed conversion ratio (FCR) values were in the range of 1.23–1.39 for catfish (Clarias gariepinus) in the aquaponic system with stocking ratio of 25 kg/m³ at different flow rates, by stating that the FCR values were close to the ideal value for aquaculture. Thus, in our study FCR of the all groups (Group I 1.16; Group II 0.93 and Group III 0.91) are appropriate when compared to the economic FCR values in aquaculture. Here, SGR values were 2.28 (Group I), 2.31 (Group II) and 2.43 (Group III), presenting good growth performance. SGR values are higher than the values assessed by Al-Hafedh [17], Monsees [15] and Endut [16] for the aquaponic production.

pH values fluctuated in all groups during the present study. pH is one of the crucial factors in aquaponics and should be kept around 7 for the success in nitrification; converting ammonia and providing nitrate for the plants ([12, 15, 18]). Although the pH values were below the optimal value for the fish in this experiment tilapia tolerated the pH changes. On the other hand, pH values were suitable for the plant in the present study hence, most plants need a pH value between 6 and 6.5 in order to enhance the uptake of nutrients [12]. It is known that pH<6.5 disrupts the nitrification process with eventual risk of ammonia and nitrite toxicity. Here, ammonia and nitrite exhibited high values in parallel to low pH, the peak of ammonia and nitrite corresponds to the lowest pH values. However, in our case, the nitrate values reached higher values and this may be explained by the insufficient nitrate uptake of the plant due to weak lighting. Thus, the interaction of the water quality parameters in the aquaponics with media based growing bed is more complicated and difficult to keep within optimal ranges. In terms of optimal production parameters decoupled systems are taken into consideration, as stated by Monsees et al. [15]. In this study, water potassium showed low levels. This was also reported by Graber and Junge [19] to explain a poor vegetable quality in aquaponics. In the present study, total plant biomass was low when compared with the previous studies on tomato plant in the aquaponics [20, 21]. Total biomass of the plant showed differences depending on the fish stocking ratio and total plant biomass increased with decreasing the fish density. However, the proportion of root to total biomass decreased with fish density. Here, more leave portion was observed in one-loop system. This has been reported before by the fact that of suboptimal nutrient supply [22].

Leaves portion to total biomass increased in the one-loop system here. Thus, Bloom et al [23] reported that when exposed to low light, plants usually respond by increasing allocation of biomass to leaves, by actively creating a dynamic balance where all resources should be equally limiting to growth. Goddek et al [12] reported that every plant and fish species have different nutritional needs that are also dependent on the growth stage/life-cycle and external factors (including system design). Hence, the optimization of whole aquaponics system to dual production is highly complicated. Regarding the fish reaction to water quality fluctuations, tilapia tolerated the sharp changes in water quality as reported by Rakocy [24]. Survival ratio is considered in normal ranges as found in RAS.

V. CONCLUSION

The effects of stocking rate were determined for the tilapia growth and plant biomass in one-loop system. The growth performance and feed conversion assessed in this study were better in the group with the maximum density (initial stocking rate, 50 kg/m³). Total plant biomass was found to be low with the various limiting factors including insufficient lighting of aquaponics system used. The most important factor was to control the water quality, particularly pH and nitrogenous substances. Thereby, the dynamic action of water quality in one-loop systems may not meet the expectations in terms of co-production performance. To optimize fish stocking density in the aquaponics the complexity of the water quality should be considered in one-loop system.

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