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Growth performance and hematological, biochemical, and histological characters of the Nile tilapia (*Oreochromis niloticus*, L.) cultivated in an aquaponic system with green onion: The first study about the aquaponic system in Sohag, Egypt

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ABSTRACT

The aquaponic system has won the interest of researchers for its multiple efficiency in raising fish and producing crops simultaneously; however, information about the extent of this system's efficacy is scarce compared to other conventional food production methods. Hence, this study aimed to assess water quality, hematological and biochemical parameters, histological structure, and growth performances of the Nile tilapia (Oreochromis niloticus), as well as green onion (Allium cepa L.) yield. The species under study was reared in a newly developed deep-water culture (DWC), forming a prototype built in Upper Egypt, Center of Development owned by the ASRT in Sohag Governorate, Egypt. The current experiment consisted of two DWC systems. Fish were daily fed for 5days/ week with a protein content (30%), representing 2% of the total biomass. Except for water pH and temperature, the values of water quality were within the optimal ranges for tilapia culture. No significant difference was detected between the two systems with respect to the hematological, biochemical parameters, hepatic and intestinal histological structure, and growth performances, showing normal values. In addition, considering the two DWC systems, no significant difference was recorded in the growth and yield of green onion determined through fresh weight. It is deduced that aquaponics has a huge potential since it shows promising results for urban food development. At the same time, it provides new entrepreneurship and start-up opportunities. Nevertheless, more studies are recommended to overcome foreseen challenges despite its potential to be implemented in developing countries such as Egypt.

INTRODUCTION

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The concept of aquaponics is not popular (**Oladimeji** *et al.*, **2020**). In an aquaponics system, waste organic matters originated from aquaculture system, which are toxic to animals transform microbes into soluble nutrients for growing plants. At the same time, the hydroponics system treats water, recirculating the clean water to the

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aquaculture system (Somerville *et al.*, 2014). Beside its ecological merits, the aquaponics system has other additional economic advantages. It saves (input) treated water for aquaculture system and formulates a fertilizer for hydroponics system, benefiting from double outputs. Moreover, it makes use of a single input (fish feed) to harvest animal and plant together (Liang & Chien, 2013). These benefits lead to the production of food without external addition of chemicals or fertilizers. Hence, aquaponics is considered a form of organic farming (Al-Hafedh *et al.*, 2008). The deepwater culture (DWC), or raft method, involves suspending plants in polystyrene sheets with their roots hanging into the water. This method is the most common for large commercial aquaponics and is more cost-effective than large-scale media beds (Somerville *et al.*, 2014; Engle, 2015). The DWC method enables the roots of plants to absorb nutrients in water without clogging the water channel, although aeration for DWC units is vital (Somerville *et al.*, 2014).

Growth performance, serum biochemical parameters, hematological parameters, and histological structure are indicators of water balance, nutritional status, and overall health condition of fish (Chang & Hur, 1999; Denson *et al.*, 2003). Various plants and aquatic species can be grown together using an aquaponic system. The tilapia is likely the most prominent of all aquaculture fish in the 21st century (Fitzsimmons, 2000). The favorable characteristics of the Nile tilapia (*O. niloticus*) makes its cultivation the most tolerant to harmful environmental conditions. Additionally, it can survive at low euryhaline and low dissolved oxygen and shows relatively fast growth and effecient food conversion (Asad *et al.*, 2010). Notably, green onion (*Allium cepa* L.) is one of the most important crops in Egypt used for local consumption, and it is also an exportation commodity (Abo El-Hamd *et al.*, 2016). The most common maturity indices of green onion are the size of the plant and bulb diameter (Mohamed & Atress, 2016).

The present research was conducted to evaluate water quality, hematological and biochemical parameters, histological structure, and growth performances of the tilapia (*O. niloticus*) and onion (*Allium cepa* L.) yield, reared in a newly developed DWC, representing a prototype built in the Upper Egypt Center of Development owned by the ASRT in Sohag Governorate, Egypt.

MATERIALS AND METHODS

Aquaponic (Deepwater Culture System)

Two deepwater culture systems have been set up within the greenhouse in the Upper Egypt Center of Development owned by the ASRT in Sohag Governorate, Egypt. In each DWC system (also known as the raft system), the plants were grown on Styrofoam rafts floating on top of water (Fig. 1). A two–cubic meter tank was used as a fish tank. Water flows by gravity to a mechanical filter used to remove the solid waste from water. First, the water flows from the mechanical filter to the net filter. Then, the water flows from the biofilter to the building's grow bed that is covered with vinyl. The grow bed enables plants to grow in net cups, fixed in holes within the Styrofoam rafts that

float over water in the plants' grow bed. Plants' roots absorb soluble nutrients in water after the nitrification process. Afterwards, water is pumped again to the fish tank through a submersible pump. The whole system is aerated using an air pump to maintain enough dissolved oxygen in both the fish tank and the plants' grow bed.

Experimental Fish and Feeding

Healthy Tilapia (*O. niloticus*) and those without visible injuries were collected from a fish farm (Alahywa Fish Hatchery Station, Sohag Governorate) and transported to the aquaponics unit at the Upper Egypt Center of Development. A number of 200 tilapia fish, with an average weight $(47 \pm 4 \text{ g})$ were stocked in each tank. During the period of acclimatization, the fish's feed was of appropriate size and content. The trial continued for 110 days starting on the 24th of November 2020 while the harvest was on the 16th of March 2021. The samples were fed manually once per day. with a daily feed ratio of 2% from fish body weight.

Hydrochemical Parameters

The dissolved oxygen (mg. l^{-1}) was measured with a portable dissolved oxygen meter. The pH and water temperature (°C) were measured with a portable meter pH meter. TDS, salinity, and electrical conductivity (μ S.cm⁻¹) were recorded with a portable conductivity meter. The concentration of ammonium (mg. l^{-1}), and nitrate (mg. l^{-1}) were registered using UV-spectrophotometer (Unico1200).

Fish growth performance

The average individual weight (g) of the fish was gauged at the start and the end of the experiment. At the end of the trial, the weight gain (g), growth rate (g/day), feed conversion ratio, feed efficiency ratio, and survival rate (percentage) were calculated.

Weight gain (g) = final weight - initial weight.

Growth rate (g/day) = final weight - initial weight/experiment period.

Feed conversion ratio (FCR) = food intake/weight gain.

Feed efficiency (FE) = weight gain/feed fed.

Hemato-Biochemical Parameters

The following hematological parameters were measured and calculated: red blood cells (RBCs), white blood cells (WBCs), platelets (PL), hematocrit (HCT), hemoglobin (Hb), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC), using the following formula of **Dacie and Lewis (1991)** and **Fazio (2019)**:

MCV (μ m³) = HCT/RBCs × 10, MCH (pg) = Hb/RBCs × 10.

MCHC (%) = Hb/HCT \times 100.

The biochemical parameters, including creatinine (CR), uric acid (UA), aspartic aminotransferase (AST), alanine aminotransferase (ALT), glucose (GL), alkaline phosphatase (ALP), cholesterol, and total protein (TP) were measured using a spectrophotometer (Jasco- V530, Ottawa, Canada) as defined in the study of Hamed *et al.* (2019).

Hepatic and Intestinal Histomorphology

The liver and intestine were rinsed and fixed in a 10% neutral buffered formalin for48 hours, dehydrated in the graded ethanol, and then cleared in xylene before embedding in paraffin. Dewaxed sections (5 µm) were stained with Harris's Hematoxylin and Eosin stains (H&E). Sections were visualized and studied using an Olympus microscope (BX50F4, Olympus Optical Co. LTP, Japan).

Plant Yield

Fresh weights of green onion (kg) were weighed using a digital balance.

Statistical Analysis

Means and standard errors were used to present the data. A t-test was used to estimate the differences between different treatments.

RESULTS

Hydrochemical Parameters

Temperature values varied from 17.5°C to 22°C. The pH range appeared within 8.2– 8.62. For the dissolved oxygen concentration (OD), its value ranged from 6.3 mg L^{-1} to 6.6 mg L⁻¹. The electric conductivity (EC) value was 536 (μ S); TDS value was 739; salinity value was 0.3; ammonia value was 0.93, and nitrate values varied from 3.1 to 7.7(Table 1).

Table 1. Water physicochemical parameters for the two aquaponic systems								
Items	T (°C)	DO (mg/L)	EC (µS)	pН	TDS (ppm)	Salinity (ppm)	Ammonia (mg/L)	NO ₃
Values	17.5-22	6.3-6.6	536	8.2- 8.62	739	0.3	0.93	3.1-7.7



Fig. 1. A newly developed DWC, a prototype built in the Upper Egypt Center of Development owned by the ASRT in Sohag Governorate, Egypt (1- Fish tank, 2- Mechanical filter, 3- Net filter, 4- Biofilter, 5- Hydroponic part).

Growth Performance

The growth performance parameters of the tilapia (O. niloticus) in the two systems are illustrated in Table (2) and Fig. (2). Weight gain (g) values varied from 140 ± 32 to 146 \pm 21. Growth rate (g per day) values varied from 1.3 \pm 0.3 to 1.3 \pm 0.2. The food conversion ratio was the same in both systems. FER (in percentages) values varied from 94.5 ± 21 to 98± 14.



Fig. 2. Green onion and tilapia (*O. niloticus*) in a newly developed DWC, a prototype built in the Upper Egypt Center of Development owned by the ASRT in Sohag Governorate, Egypt

Parameters	Initial weight (g)	Final weight (g)	Weight gain (g)	GR (g per day)	FCR	FER (%)	Survival (%)
Values	45±4 - 48±5	185±36 - 194±26	140±32 - 146±21	1.3±0.3 - 1.3±0.2	1.1±0.02 - 1.1±0.2	94.5±21 - 98±14	94-98

Table 2. Growth performance parameters of tilapia (O. niloticus) for the two aquaponic systems

Hematological Parameters

The determined hematological parameters of *O. niloticus* in the two systems are shown in Table (3). All the analyzed hematological parameters were within the normal values in the two systems.

Table 3. Hematological parameters of tilapia (O. niloticus) for the two aquaponic systems

Hematological parameters	Values		
(RBCs) (million/mm ³)	$1.6 \pm 0.6 - 1.5 \pm 0.1$		
Hemoglobin (Hb) (g/dl)	8.4 ± 0.6 - 8.2 ± 0.2		
Ht (PCV) (%)	$25 \pm 0.7 - 25 \pm 0.7$		
MCV (µm ³)	$159 \pm 0.6 - 166 \pm 12.7$		
MCH (Pg)	$53.7 \pm 0.9 - 54 \pm 4.1$		
MCHC (%)	$33.7 \pm 0.3 - 32.5 \pm 0.4$		
Thrombocytes (Thou./mm3)	$314 \pm 0.3 - 307 \pm 7.9$		
(WBC's) (Thou./mm3)	$840 \pm 0.4 - 832 \pm 6.8$		
Lymphocytes (%)	$90 \pm 0.1 - 92 \pm 0.0$		
Monocytes (%)	$2.7 \pm 0.1 - 1 \pm 0.6$		
Neutrophils (%)	$6.3 \pm 0.6 - 6 \pm 0.6$		
Eosinophils (%)	$1 \pm 0.0 - 1 \pm 0.0$		

Biochemical Parameters

Table (4) illustrates the investigated biochemical parameters in the serum of tilapia (*O. niloticus*). All the analyzed biochemical parameters were within the normal values in the two systems.

Biochemical parameters	Values
AST activity (U/l)	55.6±0.1 - 53.1±1.1
ALT activity (U/l)	30.7±0.1 - 27.7±1.0
ALP activity (U/l)	30±0.7 - 30.4±0.4
Glucose (mg/dl)	97.7±0.2 - 106.8±5.4
Cholesterol(mg/dl)	194±0.1 - 205±4.0
Total protein (mg/dl)	5.9±0.1 - 6.4±0.2
Creatinine (mg/dl)	0.6±0.1 - 0.6±0.1
Uric acid (mg/dl)	16.6±0.8 - 16.9±0.5

Table 4. Biochemical parameters of tilapia (O. niloticus) for the two aquaponic systems

Hepatic and Intestinal Histology

The histological appearance of the intestine and hepatic samples is illustrated in Fig. (3). Hepatocytes from the two systems were normal and loose; polyhedral in shape with vacuolated cytoplasm. There was no obvious degeneration or inflammatory cell infiltration in all experimental groups. The intestinal microvilli were most regular in shape from the two systems with normal villus structure.

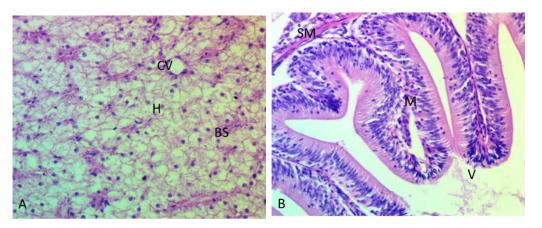


Fig. 3. Photomicrographs showing (**A**) liver tissue showing the normal architecture of liver hepatocytes (H), normal blood sinusoids (BS), and central vein (CV), and (**B**) intestine tissue showing the normal architecture of villi (V), mucosa (M), and submucosa (SM) of tilapia (*O. niloticus*) for the two aquaponic systems (H&E stain, scale bar 50 mm)

Plant Yield

Fresh weights of green onion (kg) are shown in Figs. (2, 4). The growth of green onion was similar in both systems, but it was better in system II.

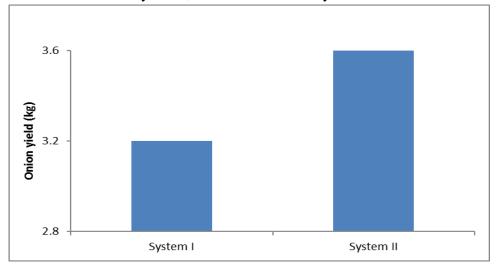


Fig. 4. Fresh weights of green onion (kg) for the two aquaponic systems

DISCUSSION

The parameters of water quality of the rearing tanks in the aquaponics systems were within the recommended range for aquaculture, except for the pH and temperature. The pH range was appropriate for the tilapia. However, some plants may have been impaired due to the restricted uptake of nutrients; this was evident by chlorosis as a result of chlorophyll inhibition and/or iron deficiency. The study of **Cerozi & Fitzsimmons** (**2016**) determined that, pH between 6.5 and 7.2 is the optimum for the process of nitrification, fish growth, and maximum plant biomass production. In the present study, dissolved oxygen ranged between 6.3 and 6.6 mg/l. Moreover, **Pattillo (2016)** stated that the average values of dissolved oxygen ranged between 6.8 and 7.4 mg/l. **Boyd (1982)** suggested that dissolved oxygen should be above 5 mg/L to support the survival and the development of aquatic life in any culture system. **Kamal (2006)** reported that, 6.6–6.8 mg DO/l is suitable for the growth of the Nile tilapia (*O. niloticus*).

Meanwhile, the temperature values recorded were slightly outside the optimum range since the experiment was conducted during the months of winter, where heaters were used to overcome the cold weather. **Gui** *et al.* (1989) found that. the average temperature of 28°C was optimal for the growth of the Nile tilapia. On the other hand, ammonia (NH₃) was within the optimum level of the aquaponic system during the experiment, which was less than 1.0 ppm determined in the study of (Somervilla *et al.*, 2014). Ionized ammonia (NH₄⁺) stimulates plant growth, but very low levels of unionized ammonia (NH₃) may cause stress and death of fish. Levels can be reduced by reducing feeding, lowering stocking density, improving biological filtration, use of ion exchange

materials to selectively remove ammonia in addition to changing water to affect dilution. **Syafiqah** *et al.* (2015) noted that, plants in the aquaponics system act as biological filters, thereby absorbing nutrients such as nitrate and NH₃ from the system. This justifies the observed low levels of nitrate in the aquaponics system. **Visvanathan** *et al.* (2008) and **Mullen** (2009) noticed an upper lethal limit of nitrate at about 500 mg/l. Liedl *et al.* (2004) and **Rakocy** *et al.* (2006) suggested that, the acceptable levels of nitrogen for plants at the outset are optimal at around 100 mg/l, while it should be at 200 mg/l during growth. Yet, the apparent health of most of the plants (especially the leafy vegetables) indicated that even these low average levels of nitrate around 5.0 mg/l were enough to yield healthy vegetation, especially in the Swiss chard, lettuce, celery and spring onions. Increasing the fish density would have increased the nitrate supply, and most probably, further surged the growth of most of the vegetables and herbs; the increase of fish densities is actually possible (Kotzen & Appelbaum, 2010). Rakocy (2010) suggests that it is viable to stock the tilapia at around 75 to 150/m³ of water, varying with the species.

The EC for the freshwater system was 536 μ S/cm. As noted in Kotzen & Appelbaum (2010), Rakocy (2006) states that EC should be from 1500 to 3000 μ S/cm in aquaponic systems. Aquaponics system varies according to two factors: a) the nutrients in aquaponics are generated constantly (**Rakocy, 2010**). In this respect, the organic nature of the nutrients is generated besides low concentrations of salts, causing low conductivity values (**Nelson, 2008**). Therefore, the observed EC values indicate the existance of good concentration of water nutrients, which were efficiently utilized by plants even when roots were in a medium-to-slightly alkaline pH, showing that used herbaceous adapt to such systems.

In the present study, weight gain (g) values varied from 140 ± 32 to 146 ± 21 . Growth rate (g/day) values varied from 1.3 ± 0.3 to 1.3 ± 0.2 . The food conversion ratio was the same in both systems. FER (in percentages) values varied from 94.5 \pm 21 to 98 \pm 14. Rakocy et al. (2004) stated that, the mean harvest weight was 813.8 g and the survival rate reached 98.3%, while feed conversion ratio was lower at 1.7 in the tilapia. Rahmatullah et al. (2010) postulated that, weight gain (g) of the tilapia was within 19.41-32.6 7 g and was inversely related to stocking density. Percentage weight gain varied between 2553.99 and 4298.68% and was significantly different in the treatments. SGR ranged from 3.09 to 3.59% per day and varied significantly. FCR varied from 2.19 to 2.69. Goda et al. (2015) observed that, feed conversion ratio (FCR) recorded values of 1.69 and 1.8 for the Nile tilapia in the aquaponic system. Kotzen and Appelbaum (2016) recorded a surge of 0.78 g/day/fish for the brackish water system and 1.1 g/day/fish in the freshwater system. The tilapia biomass increased by almost 47 (g), and each fish showed an increase in the total length by 3 cm. Lower FCR means that the feed was utilized more efficiently by the fish. FCR became better at the end of the experiment, which was equal to 1.74. FCR is related to the feed and the fish water quality. Moreover, the tilapia would have an FCR within 1.4–1.8 if the conditions were better and more optimal (Awad, 2017). Stathopoulou *et al.* (2018) stated that no fish mortality occurred. Weight gain was statistically and significantly higher in the first aquaponics system (system I) (WG) (95.8 \pm 13.62 g), compared to the second system II, where it was 51.7 \pm 9.90 g. The specific growth rate was higher in system I (SGR%/day, 1.8 \pm 0.17) than system II (1.2 \pm 0.16%/ day), but it was not statistically significant. Feed consumption rates (FCR) were 0.7 and 1.1 for systems I and II, respectively. Akter *et al.* (2018) mentioned that the length and weight percentage gains were 33.81% and 174.06%, respectively. It was found that the survival rate and FCR were 98.33% and 1.56, respectively. The total production of fish was 29.44 tons/ha/90 days.

Setiadi et al. (2018) reported that, the highest survival rate, weight, and length were $96.00 \pm 1.73\%$, $32.31 \pm 0.74g$, and 7.57 ± 0.21 cm in the red tilapia, respectively. Hundley et al. (2018) observed that, the mean gain in weight record was 54.11 ± 22 ; while, the gain value of 164.01 ± 55.25 g was reported in the work of **Palm** et al. (2014) for the Nile tilapia cultured in an aquaponic system. Saufie et al. (2020) mentioned that the low value of FCR (1.04–1.13) is a good indication of the high-quality feed that the fish can efficiently convert into tissues, and thus requires it in smaller quantities to produce a unit weight gain. It can help in predicting the growth of the fish and the profitability of aquaponics. Thus, in this study on GIFT, the high value of SGR (2.02-2.04) can be interpreted in terms of efficient nutritional management as reflected by low FCR. Hussein et al. (2020) stated that the final mean weight for the mixed-sex tilapia, yielded from seven harvests in the aquaponic system was 285.3 g/fish. FCR averaged 1.4 for the mixed-sex population and 1.3 for all-male tilapia. The specific growth rate (SGR) values were 1 and 1.1% for mixed-sex and all-male tilapia, respectively. Survival was at its lowest for mixed-sex tilapia, averaging 95.8% compared to 99% for all-male tilapia. FCR averaged 1.4, with ranges from 1 to 1.7 in the aquaponic system.

To our knowledge, few studies investigated the hematological, biochemical, and histological profiles of aquaculture animals in aquaponics. In the present study, all the biochemical parameters analyzed in two systems showed non-significant differences. Furthermore, **Osman** *et al.* (2021) detected no significant difference in the values of nearly all the hematological parameters in the blood of fish reared in the aquaponic system and those reared in pond systems, being always within the normal ranges for the healthy Nile tilapia. Most of the measured biochemical variables exhibited an improvement in the blood of fish reared in the ASTF-PRO system, compared to that of the pond. **Anantharaja** *et al.* (2017) determined that, the RBC and the hemoglobin content in blood were higher in fish reared in aquaponic systems. **Gao** *et al.* (2020) noted that the GLU levels of Qihe crucian carp C. auratus ranged between 3.31 and 6.14 mmol L^{-1} in the aquaponics system.

The growth of green onion was similar in both systems, but it was better in system II. Kotzen & Appelbaum (2016) mentioned that, the growth of the plants in the two

aquaponic systems, compared to plants grown in soil was remarkable. All the plants in the aquaponic systems were larger, more advanced, and healthier in the aquaponic systems, including the kohlrabi, leeks, lettuce, cabbages, cauliflower, spring onions and various herbs. Awad (2017) stated that, the average values of planted chili peppers heights ranged from 6.1 to 10.5 cm during the experiment. The average values of planted eggplant heights ranged between 6.5 to 12.1cm during the experiment period. Oladimeji *et al.* (2020) observed that, pumpkin production in the aquaponics system was about five times the performance in irrigated land and eleven times that in no irrigated land. Hussein *et al.* (2020) reported that, the lowest density of fish (3.5 kg/1,000L) produced significantly smaller lettuces with nitrogen deficiency symptoms. The middle (6.5 kg/1,000L) and highest densities (13 kg/1,000L) produced bigger lettuces, with no significant differences between their weights.

CONCLUSION

The present study demonstrated that the combination of the tilapia fish and onion in deepwater aquaponics used in this study was extremely effective in fish waste treatment and also water conservation. The findings are valuable and applicable in the development of aquaponics and biological waste reuse in a newly developed DWC, a prototype built in the Upper Egypt Center of Development owned by the ASRT in Sohag Governorate, Egypt.

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