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Sustainability of Growth Performance, Water Quality, and Productivity of Nile Tilapia-Spinach Affected by Feeding and Fasting Regimes in Nutrient Film Technique-Based Aquaponics

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Abstract: Aquaponic systems offer an innovative approach to sustainable agriculture, where the interplay between fish and plant cultivation can be optimized. The choice of feeding and fasting schedules plays a crucial role in system efficiency and overall productivity. This study aims to investigate the impacts of various feeding and fasting schedules on water quality, and the growth performance of Nile tilapia, *Oreochromis niloticus* fingerlings, and spinach productivity in an aquaponic system based on the nutrient film technique (NFT). *O. niloticus* fingerlings with an initial weight of 13.47 ± 0.14 g were randomly stocked at a density of 6 kg/m^3, and spinach plants (Spinacia oleracea) were included. The study employed a completely randomized block design with five replications. Various water quality parameters were monitored, and the effects of different feeding/fasting schedules on fish and spinach were assessed. The data revealed significant differences ($p < 0.05$) in water quality parameters, all of which remained within acceptable ranges for aquaponic systems. The one-day feeding/one-day fasting treatment resulted in reduced final body weight, weight gain percentage, and specific growth rate, compared to other treatment groups ($p < 0.05$). Higher levels of glucose and plasma cortisol were observed in this treatment. Economic efficiency was highest in the daily feeding treatment (40.05%), with no statistical difference ($p > 0.05$) observed in the group subjected to three-day feeding/one-day fasting (39.03%). Spinach yield varied significantly between treatments ($p < 0.05$), with the daily feeding treatment recording the highest yield (2.78 kg/m^2) and the one-day feeding/one-day fasting cycle having the lowest yield (1.57 kg/m^2). The findings suggest that the three-day feeding/one-day fasting regime in an NFT-based aquaponic system results in efficient nutrient utilization, higher productivity, and profitability for Nile tilapia. Additionally, this approach supports marketable biomass production for spinach. Different feeding and fasting schedules have distinct effects on water quality, fish growth, and spinach productivity in aquaponic systems. The three-day feeding/one-day fasting schedule emerges as an effective strategy for optimizing resource utilization and increasing overall productivity.

Keywords: aquaponics; feeding schedule; fasting; Nile tilapia; nutrient film technique; spinach yield; water quality

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1. Introduction

According to the Food and Agriculture Organization (FAO), food security includes having access to food at all times [1]. The Kingdom of Saudi Arabia experiences a variety of semi-arid to very dry climates with very little precipitation; as a result, the Kingdom is susceptible to threats to general food security due to its limited agricultural natural resources, high reliance on food imports, trade policies, water scarcity, rising rates of food losses, and the mounting effects of climate change [2]. In addition, over the past two decades, Saudi Arabia has experienced rapid economic growth that has led to an increased demand for protein-rich foods such as fish and vegetables that are high in value nutrients. According to the vision of the Kingdom of Saudi Arabia 2030, it aims to rely on soilless agriculture systems.

Modern agricultural techniques have shown that soilless agriculture (hydroponic and aquaponic systems) is one of the ideal solutions for sustainable food production in dry areas. According to refs. [3,4] hydroponic agriculture and aquaculture are combined in a system known as aquaponics, which is a great way to conserve water and dispose of aquaculture wastewater. In an aquaponic system, hydroponically produced plants receive nutrients from nutrient-rich aquaculture effluents. The Aquaponic system is regarded as an eco-innovative and resource-effective closed-loop manufacturing method because of its commitment to the environment. Instead of dumping aquaculture wastewater into nearby water bodies, this method recycles it and uses it to irrigate hydroponically produced plants [5–7]. Plants absorb these nutrients from aquaculture effluents before being put back into the fish tank. Although it is well known that recirculating aquaponic systems are more effective at using water and nutrients than conventional systems, managing recirculating aquaponic systems is difficult because of problems with water quality management, which must be properly resolved to increase nutrient recycling and crop productivity in the system [8,9]. Aquaponics is anticipated to help achieve food security and compensate for animal protein from fish and essential nutrients from crops. To put it another way, aquaponics offers greater advantages than separate aquaculture and hydroponic food production [10]. One of the best types of fish that can be raised in the aquaponic system is Nile tilapia (*Oreochromis niloticus*). Tilapia fish are relatively well-nourished, grow quickly, and are more weather-resistant than other fish. The filter-feeding Nile tilapia can be raised in a variety of intensified production systems [11,12]. Thus, increasing the sustainability of Nile tilapia production systems can be achieved by optimizing the absorption of endogenous feed supplies.

For economic and environmental sustainability, the creation of feeding systems that enable fish to utilize food more effectively is crucial [13]. Since fish feed is the primary source of nutrients for plants in aquaponic systems, the feeding schedule has a significant impact on nutrient availability and composition. Aquaculture sector growth is hampered by the high cost of feed and a lack of components, which also reduces profitability [14]. Fish farming income, water quality, and diet composition are all impacted by feeding rate and diet composition [15]. In aquaculture, overfeeding leads to poor digestion and absorption of nutrients as well as poor water quality assays [16]. On the other hand, undernourishment leads to a decrease in growth and immunity parameters, and more exposure to diseases and mortality [17].

Fish performance and productivity can be impacted by the duration of intermittent or continuous feeding regimens [18]. In aquaculture, cyclical fasting and refeeding can be another practical dietary approach [19]. After the fasting phase, fish can offset the growth by increasing their ability to digest and absorb nutrients, which increases their ability to consume food and acquire weight [20]. Generally, factors such as meal type, fish size, fasting length, water quality, and health status have a great impact on the fish's compensatory growth. Prior studies suggested that Nile tilapia had compensatory growth as a result of intermittent fasting and refeeding [21,22].

To increase the system's ability to utilize nutrients, there is a fundamental need to manage aquaponic systems for crop cultivation more skillfully and to use nutrient sources more effectively. By reducing hazardous chemicals in aquatic solutions, proper nutrient management techniques can improve the high output and quality of plants in an aquaponic system and the water quality for fish, which is crucial for the long-term viability of recirculating aquaponic systems. Therefore, the objective of this study was to examine the impacts of feeding and fasting regimes on sustainability water quality and growth performance for Nile tilapia, *O. niloticus* fingerlings and the productivity of spinach (*S. oleracea*) in an NFT-based aquaponic system.

2. Materials and Methods

2.1. Aquaponic and Experimental Design

The trial was carried out in the King Faisal University greenhouse in Al-Ahsa, Saudi Arabia (26°23018.5600 N, 50°11016.0100 E), based on the nutrient film technique (NTF) aquaponic system for 60 days. Each aquaponic system was equipped with a 500 L circular fish tank (water volume 400 L), a 250 L mechanical filter, 500 L biofilter tanks, and a hydroponic culture system (NFT parallel plastic channels). Figure 1 displays a schematic diagram of an aquaponics system [23]. The hydroponic system (NFT) consisted of five 10 m long plastic channel troughs set in parallel. A submersible water pump (flow rate: 1500 L/m) was installed in each fish tank, and moved water at a rate of five m^3/hour to the mechanical filter and subsequently to the biological filter. By controlling the water flow, the fish tank's volume was altered twice every hour. Each fish tank had an air blower to maintain dissolved oxygen (DO) levels above 6 mg/L. The spinach in the hydroponic system absorbed nutrients that were dissolved in the aquaculture effluent, and purified water was then pumped back into the fish tank. During the trial period, water was not exchanged but rather recirculated between the hydroponic culture unit and fish tank. The trial used a completely randomized block design with five replications. Four treatments, depending on the different feeding and fasting regimes, were set: daily feeding, one-day, two-day, and three-day feeding versus one-day fasting.

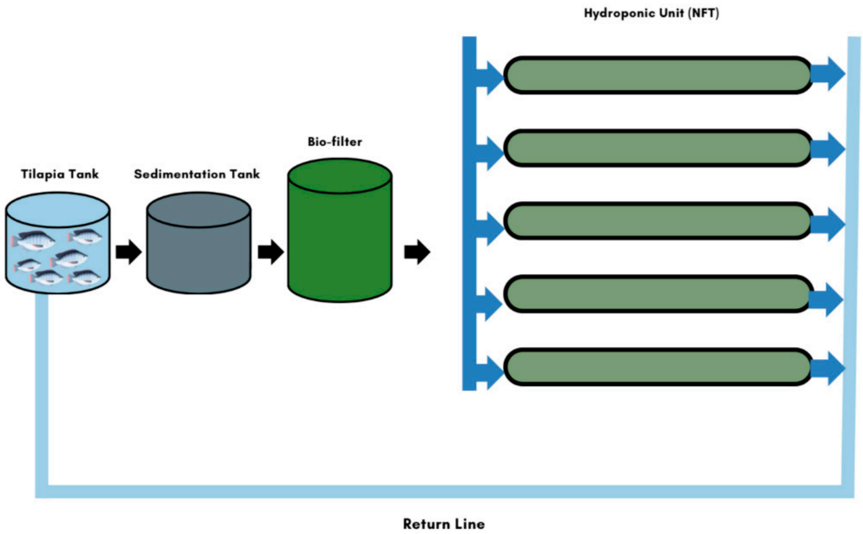

Figure 1. Schematic diagram of an aquaponics system.

2.2. Tilapia-Spinach Procedure

In nursery trays filled with coco peat and vermicompost, spinach seeds were planted. Spinach was planted rate of 36/m^2 (7.24 ± 0.11 cm). Four harvests of spinach were carried out on the 15th, 30th, 45th, and 60th days. In addition, the nutritional composition, and leaf pigment were estimated in spinach. All male-sex reverse Nile tilapia fingerlings with an initial weight of 13.47 ± 0.14 g were stocked randomly at 6 kg/m^3. For acclimation, fish were kept for two weeks. During the trial, the fish were fed twice daily at a rate of 4% of the fish body weight using a commercial feed that contains 35% protein and 4% crude fat. The fish growth characteristics were measured fortnightly, while the stress response of Nile tilapia under different feeding and fasting regimes was analyzed at the end of the trial period.

2.3. Water Quality Assay

Temperature (T), dissolved oxygen (DO), and electric conductivity (EC) were measured by a YSI 556 multiparameter meter (Atlanta, USA); pH was measured by an EcoSense pH10A Pen Tester; and total dissolved solids (TDS) were measured by Hach test kits. These water quality parameters were measured daily before fish feeding. Hardness, alkalinity ($CaCO_3$), total ammonia nitrogen (TAN), nitrite-nitrogen (NO2-N), nitrate-nitrogen (NO3-N), and phosphate (PO4-P) were all measured according to [6]. A flame photometer (Microcontroller Flame Photometer, Labard Instruchem Pvt. Ltd. (South Dumdum, India), Model: LIM-204) was used to analyze the calcium and potassium, in the water, according to [6].

2.4. Growth Index of Nile Tilapia

On the last day of the experiment, growth index and survival were evaluated and determined as follows:

$$\text{Final body weight (FBW)} = \frac{\text{total weight of fish in each tank (g)}}{\text{number of fish in each tank}}$$

$$\text{Weight gain(WG\%)} = 100 \times \left(\frac{\text{average final weight (g)} - \text{average initial weight (g)}}{\text{average initial weight (g)}}\right)$$

$$\text{Specific growth rate (SGR)} = \left(\frac{\text{Ln final weight (g)} - \text{Ln initial wgight (g)}}{\text{days}}\right) \times 100$$

$$\text{Feed conversion ratio (FCR)} = \frac{\text{feed intake (g)}}{\text{weight gain (g)}}$$

$$\text{Feed consumption (FC)} = \frac{\text{average total consumption (g)}}{(\text{number of fish at the end } + \text{ number of fish at the beginning}) \div 2}$$

$$\text{Survival rate} = \left(\frac{\text{number of fish at the end of trial}}{\text{number of fish at the beginning of trial}}\right) \times 100$$

2.5. Determination of Stress Parameters in Nile Tilapia

The parameters associated with stress in fish were investigated on the last day of the trial. The tilapia fasted for the day before sampling, and 15 fish from each treatment were chosen (three fish per replication). After applying clove oil for the fish's anesthesia, an anticoagulant was added and blood was drawn from the caudal vein. The samples were centrifuged for 10 min at 5000 rpm. The collected plasma was stored for subsequent examination in a freezer at 20 °C. A commercial Cortisol ELISA kit (ZELLX, Berlin, Germany) was used to measure cortisol using an immune enzymatic assay. A spectrophotometer (LabX® Germany) was used to measure the levels of triglycerides and glucose [24,25].

2.6. Economic Efficiency of the Feed

The following equations were used to calculate the economic efficiency in accordance with Kishawy et al. [26] and Allam et al. [27]:

$$\text{Feed cost for 1 kg weight gain/USD} = \text{FCR} \times \text{ feed cost of 1 kg}$$

$$\text{Net profit (kg WG) United States dollar(USD)} = \text{Sale price of kg WG} - \text{gross cost/kgWG}$$

$$\text{Economic efficiency} = \frac{\text{Profit per kg WG}}{\text{Gross cost per kg WG}}$$

where: The cost of 1 kg feed was 1.35 USD/kg feed. The gross cost was computed using 60% of the feed cost and 40% of the remaining cost. The difference between gross and sale costs (3.6 USD/kg WG) for every kg WG was used to compute net profit. The prices of feed ingredients and sale of fish were set in accordance with the going rates in the Saudi Arabian local market during April 2023. The rate of one USD was 3.77 Saudi Arabia riyal (SR).

2.7. Growth and Nutritional Composition Analysis of Spinach

The spinach yield was determined at each harvest. A total of twelve spinach plants were randomly selected from each replicate to estimate the growth of the spinach plant. To determine the plant's height, a graduated ruler was employed as well as the number, length, and width of the leaf. The content of spinach leaf in various mineral elements (nitrogen, phosphorus, calcium, potassium, magnesium, copper, sulfur, and zinc) was estimated, as described by [28,29].

2.8. Leaf Pigment Analysis of Spinach

According to the method of [30], which was used to extract chlorophyll, 25 milligrams of leaf tissue were added to 3 milliliters of dimethyl sulfoxide (DMSO) and kept at room temperature for 15 h until chlorophyll was removed. Absorption was measured by using a spectrophotometer at specific wavelengths, and calculations were made for different pigments, such as chlorophyll a, chlorophyll b, and carotenoids, using the equations of [31].

2.9. Statistical Analysis

Levene's test was used to determine the normality and homogeneity of variance of all the data. The program SPSS version 26.0 and one-way analysis of variance (ANOVA) was used to assess the data, before Duncan's multiple comparison post hoc test was performed. Significance was set at $p < 0.05$. The means $\pm$ standard errors (S.E.) were used to express all the data.

3. Results

3.1. Water Quality Assays

Table 1 shows the water characteristics of fish tanks under the influence of different feed and fasting regimes over a period of 60 days. The water characteristics (temperature, pH, electric conductivity, alkalinity, nitrate, calcium and potassium) varied insignificantly ($p > 0.05$), and nitrite, phosphate, total dissolved solids, total ammonia nitrogen, and dissolved oxygen all showed a significant difference ($p < 0.05$). The values of dissolved oxygen showed the highest mean in fish exposed to one-day feeding/one-day fasting (6.44 ± 0.30), while the lowest mean was in fish treated with three-day feeding/one-day fasting (5.67 ± 0.27) ($p < 0.05$). It was observed that there were no statistical differences in dissolved oxygen, total dissolved solids, and total ammonia nitrogen among the daily feeding, two-day feeding/one-day fasting, and three-day feeding/one-day fasting treatments. All water quality parameters of the aquaponics were found to have levels within the permissible limit.

3.2. Growth Performance of Nile Tilapia

The impacts of different feeding schedules for Nile tilapia on growth index (FBW, WG, SGR, FCR, and survival rate), are presented in Table 2. After 60 days of the experiment, the final body weight (FBW) for Nile tilapia increased between two and four times the initial body weight. It was found that final body weight increases significantly with increased feeding days in the feeding regime ($p < 0.05$). The percentage of weight gain in tilapia exhibited a comparable pattern to final body weight and was found to be higher in the daily feeding treatment ($360.93 \pm 12.34\%$) and lower in one-day feeding/one-day fasting treatment ($192.96 \pm 9.58\%$) ($p < 0.05$). A significant difference ($p < 0.05$) was found in specific growth rate (SGR) and feed conversion ratio (FCR), which ranged from 2.38% to 2.09% and 1.46 to 1.36 respectively. The daily feeding and three-day feeding/one-day fasting treatments were not significantly different ($p > 0.05$) for FBW, WG%, SGR, and FCR. The percentage of survival at the end of the trial showed no statistical difference among treatments (Table 2).

Table 1. The effect of different feeding and fasting regimes (for Nile tilapia reared in the aquaponic system) on the water quality and nutrient profile during a 60-day experimental period.

Parameter	Feeding Regimes			
	Daily Feeding	One-Day Feeding/ One-Day Fasting	Two-Day Feeding/ One-Day Fasting	Three-Day Feeding/ One-Day Fasting
T (°C)	26.74 ± 0.04	26.68 ± 0.06	26.62 ± 0.06	26.88 ± 0.10
pH	7.31–8.09	7.27–7.78	7.24–7.95	7.20–8.05
DO (mg L^{-1})	5.71 ± 0.24 ^{b}	6.44 ± 0.30 ^{a}	5.82 ± 0.21 ^{b}	5.67 ± 0.27 ^{b}
EC (µs cm^{-1})	654.56 ± 4.68	643.70 ± 41.82	645.83 ± 39.88	636.33 ± 39.37
TDS (mg L^{-1})	361.72 ± 8.07 ^{a}	321.02 ± 3.45 ^{b}	348.21 ± 8.12 ^{ab}	358.88 ± 6.51 ^{a}
CaCO3 (mg L^{-1})	36.87 ± 0.61	36.16 ± 0.42	36.66 ± 0.16	35.62 ± 0.26
TAN (mg L^{-1})	0.121 ± 0.05 ^{a}	0.103 ± 0.08 ^{b}	0.125 ± 0.06 ^{a}	0.119 ± 0.09 ^{a}
NO2-N (mg L^{-1})	0.088 ± 0.004 ^{a}	0.055 ± 0.006 ^{c}	0.074 ± 0.005 ^{b}	0.087 ± 0.004 ^{a}
NO3-N (mg L^{-1})	0.37 ± 0.02	0.31 ± 0.02	0.34 ± 0.02	0.36 ± 0.02
PO4-P (mg L^{-1})	0.63 ± 0.03 ^{a}	0.51 ± 0.02 ^{c}	0.56 ± 0.02 ^{b}	0.59 ± 0.03 ^{a}
Calcium (mg L^{-1})	38.23 ± 0.62	37.84 ± 0.57	38.69 ± 0.39	38.35 ± 0.71
Potassium (mg L^{-1})	11.42 ± 0.11	11.87 ± 0.41	11.69 ± 0.37	11.51 ± 0.62

There is a significant difference (p < 0.05) in the mean values (mean ± S.E.) of each row's non-shared superscript.

Table 2. The effect of different feeding and fasting regimes (for Nile tilapia reared in the aquaponic system) on growth parameters during a 60-day experimental period.

Parameter	Feeding Regimes			
	Daily Feeding	One-Day Feeding/ One-Day Fasting	Two-Day Feeding/ One-Day Fasting	Three-Day Feeding/ One-Day Fasting
IBW (g)	13.44 ± 0.13	13.51 ± 0.16	13.56 ± 0.14	13.40 ± 0.14
FBW (g)	61.95 ± 1.27 ^{a}	39.58 ± 1.32 ^{c}	48.47 ± 0.47 ^{b}	59.67 ± 1.42 ^{a}
WG (%)	360.93 ± 12.34 ^{a}	192.96 ± 9.58 ^{c}	257.44 ± 14.31 ^{b}	345.29 ± 17.72 ^{a}
SGR (%/day)	2.38 ± 0.02 ^{a}	2.09 ± 0.07 ^{b}	2.31 ± 0.05 ^{a}	2.35 ± 0.03 ^{a}
FCR (g feed/g WG)	1.36 ± 0.03 ^{c}	1.46 ± 0.05 ^{a}	1.43 ± 0.02 ^{b}	1.37 ± 0.02 ^{c}
Survival (%)	99.92 ± 0.05	99.95 ± 0.04	99.93 ± 0.05	99.91 ± 0.08

There is a significant difference (p < 0.05) in the mean values (mean ± S.E.) of each row's non-shared superscript.

3.3. Stress in Nile Tilapia

Plasma cortisol, glucose, and triglyceride levels were collected and analyzed on the last day of the trial and the mean values ± standard error of mean (S.E.) for plasma cortisol, glucose, and triglycerides are shown in Figure 2.

There was a significant difference (p < 0.05) in plasma cortisol and glucose levels between the one-day feeding/one-day fasting therapy and the other treatments. The triglyceride levels did not show a statistical difference during the trial between groups. The highest values for plasma cortisol and glucose were 5.31 ± 0.59 and 89.11 ± 3.94, respectively in one-day feeding/one-day fasting treatment.

3.4. Economic Efficiency

All economic factors (feed cost, gross cost, net profit, and economic efficiency) were affected significantly (p < 0.05) by feeding and fasting schedules for Nile tilapia (Table 3). The feed cost was significantly lower in daily feeding and three-day feeding with one-day fasting, compared to other treatments. Similarly, the gross cost of tilapia production from daily feeding and three-day feeding with one-day fasting was significantly lower than the persistent treatments. The net profit of tilapia fed on daily feeding and three-day feeding with one-day fasting is higher (p < 0.05) than the remaining treatments. The maximum value was recorded for economic efficiency in tilapia daily feeding (40.05%) treatment, followed by three-day feeding with one-day fasting (39.03%), without a significant difference being noted. The minimum economic efficiency ($p < 0.05$) was recorded in tilapia fed on one-day feeding and one-day fasting (30.46%).

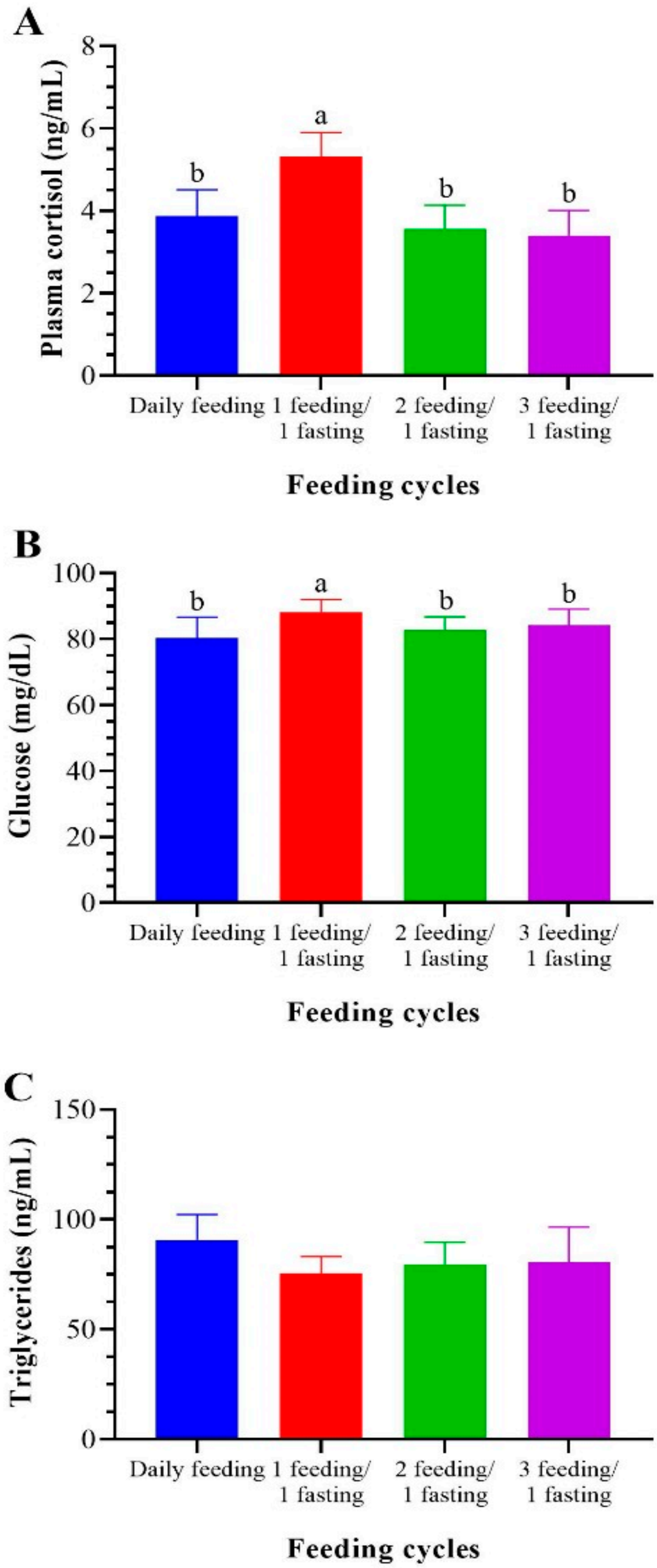

Figure 2. The effect of different feeding and fasting cycles for Nile tilapia reared in the aquaponic system on stress parameters: (**A**), plasma cortisol, (**B**), glucose and (**C**), triglyceride. Different letters above columns indicate significant differences at $p < 0.05$.

Table 3. The effect of different feeding and fasting regimes (for Nile tilapia reared in the aquaponic system) on economic efficiency during a 60-day experimental period.

Parameter	Feeding Regimes			
	Daily Feeding	One-Day Feeding/ One-Day Fasting	Two-Day Feeding/ One-Day Fasting	Three-Day Feeding/ One-Day Fasting
Feed cost (USD/kg WG)	1.78 ± 0.02^{b}	1.91 ± 0.07^{a}	1.87 ± 0.04^{a}	1.79 ± 0.03^{b}
Gross cost (USD/kg WG)	2.49 ± 0.07^{b}	2.67 ± 0.03^{a}	2.62 ± 0.05^{a}	2.51 ± 0.09^{b}
Net profit (USD/kg WG)	1.02 ± 0.08^{a}	0.84 ± 0.06^{b}	0.89 ± 0.05^{b}	1.01 ± 0.04^{a}
Economic efficiency (%/kg WG)	40.05 ± 1.02^{a}	30.46 ± 1.97^{c}	33.20 ± 2.13^{b}	39.03 ± 1.13^{a}

There is a significant difference ($p < 0.05$) in the mean values (mean ± S.E.) of each row's non-shared superscript.

3.5. Spinach Productivity

Spinach was harvested four times during the experiment period (each for 15 days), and the results showed a significant difference between the different treatments in spinach yield from the second harvest. It was noted that there was an insignificant difference between the daily feeding treatment and the 3-day feeding/one-day fasting treatment in all spinach harvests ($p > 0.05$). The daily feeding treatment recorded the highest yield (2.78 ± 0.014 kg/m^2), and the one-day feeding/one-day fasting treatment had the lowest (1.57 ± 0.023 kg/m^2) (Figure 3). Treatments differed significantly ($p < 0.05$) for spinach in terms of height increase plant and leaf of width, length, and number in the second, third, and fourth harvests (Table 4). The treatments of daily feeding and three-day feeding/one-day fasting displayed higher productivity compared to the other treatment. However, an insignificant difference was noted in the height gain, width, length, or number of spinach leaves for these treatments ($p > 0.05$), and potassium, magnesium, and sulfur concentration varied significantly ($p < 0.05$) between treatments (Table 5). The maximum content of potassium and magnesium in spinach leaves was found in the daily feeding treatment, at 3.19 ± 0.18 and 1.46 ± 0.07% respectively, and the minimum value was recorded in the one-day feeding/one-day fasting, at 2.63 ± 0.29 and 1.23 ± 0.04% respectively. No significant difference in nutritional composition of spinach leaves was observed among the daily feeding, two-day feeding /one-day fasting, and three-day feeding/one-day fasting treatments. The different feeding and fasting regimes significantly affected chlorophyll a, chlorophyll b, and carotenoid in spinach leaves during the 60-day experiment ($p < 0.05$). Significantly higher chlorophyll-a, b, and carotenoid content were observed in daily feeding and lower in one-day feeding/one-day fasting (Table 6). There was an insignificant difference in chlorophyll a, b, and carotenoid values between the groups daily feeding and three-day feeding/one-day fasting. The highest Chlorophyll-a content in spinach leaves was found in the daily feeding treatment (1.16 ± 0.06 mg/g) and the lowest in the one-day feeding/one-day fasting treatment (0.93 ± 0.12 mg/g). The chlorophyll b and carotenoid content of leaves also displayed a similar trend; the highest values were found in the daily feeding treatment (0.22 ± 0.03, and 0.49 ± 0.02 mg/g, respectively), and the lowest in the one-day feeding/one-day fasting treatment (0.15 ± 0.02, and 0.33 ± 0.02 mg/g, respectively) (Table 6).

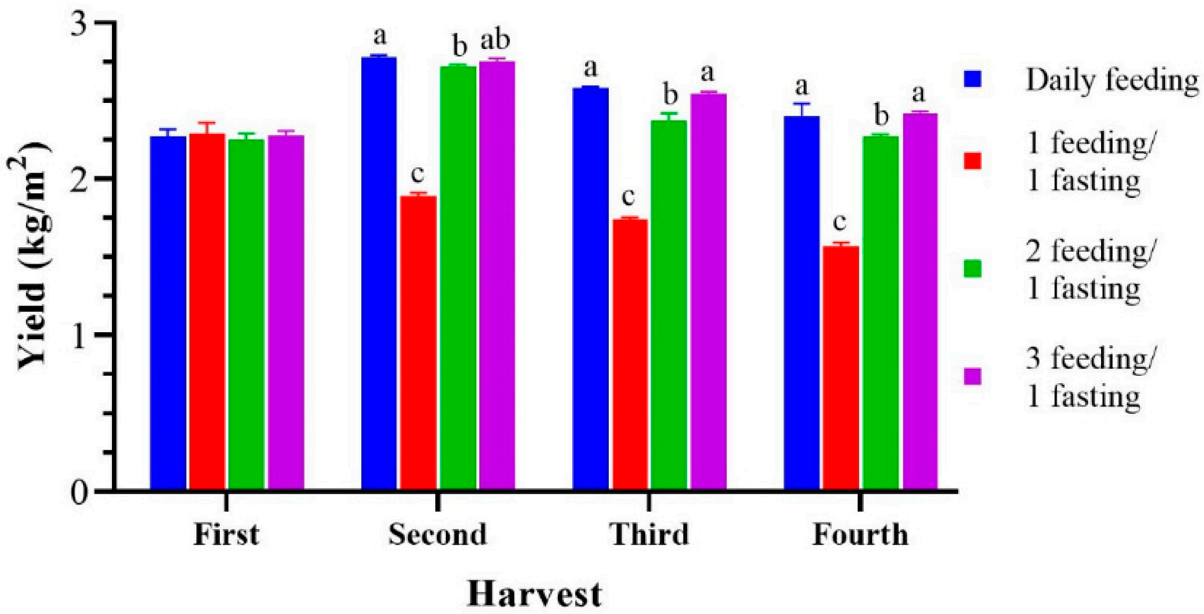

Figure 3. Spinach yield at different feeding and fasting regimes of Nile tilapia in the aquaponic system. Different letters above columns indicate significant differences at $p < 0.05$.

Table 4. The effect of different feeding and fasting regimes (for Nile tilapia reared in the aquaponic system) on the spinach growth parameters during a 60-day experimental period.

Parameters		Feeding Regimes			
		Daily Feeding	One-Day Feeding/ One-Day Fasting	Two-Day Feeding/ One-Day Fasting	Three-Day Feeding/ One-Day Fasting
Plant height (cm)	Initial	7.24 ± 0.13	7.29 ± 0.09	7.20 ± 0.10	7.26 ± 0.08
	15 Days	10.57 ± 0.26	12.15 ± 0.90	12.07 ± 0.53	10.86 ± 0.34
	30 Days	25.15 ± 1.56 ^{a}	20.40 ± 0.76 ^{b}	22.72 ± 1.09 ^{ab}	23.89 ± 1.46 ^{ab}
	45 Days	38.04 ± 1.82 ^{a}	24.96 ± 1.09 ^{b}	34.43 ± 1.48 ^{a}	35.74 ± 0.65 ^{a}
	60 Days	38.74 ± 2.01 ^{a}	32.31 ± 0.51 ^{b}	36.95 ± 1.94 ^{a}	38.13 ± 2.81 ^{a}
Leaf length (cm)	Initial	1.61 ± 0.03	1.63 ± 0.04	1.55 ± 0.06	1.63 ± 0.08
	15 Days	3.21 ± 0.08	3.02 ± 0.11	3.10 ± 0.14	3.03 ± 0.22
	30 Days	8.42 ± 0.92 ^{a}	6.96 ± 0.47 ^{b}	7.86 ± 1.19 ^{b}	8.22 ± 1.23 ^{b}
	45 Days	15.28 ± 1.08 ^{a}	9.65 ± 0.36 ^{b}	14.87 ± 0.33 ^{a}	15.06 ± 0.87 ^{a}
	60 Days	13.75 ± 0.52 ^{a}	9.06 ± 0.39 ^{b}	11.61 ± 0.26 ^{ab}	12.50 ± 0.91 ^{a}
Leaf width	Initial	0.40 ± 0.03	0.41 ± 0.02	0.44 ± 0.04	0.42 ± 0.02
	15 Days	1.49 ± 0.02	1.48 ± 0.10	1.50 ± 0.10	1.52 ± 0.10
	30 Days	5.33 ± 0.53 ^{a}	4.17 ± 0.26 ^{b}	5.21 ± 0.87 ^{a}	5.16 ± 0.68 ^{a}
	45 Days	10.63 ± 0.11 ^{a}	8.49 ± 0.36 ^{c}	9.46 ± 0.15 ^{b}	10.09 ± 0.35 ^{ab}
	60 Days	9.60 ± 0.44 ^{a}	7.42 ± 0.34 ^{bc}	8.70 ± 0.42 ^{ab}	9.55 ± 0.55a
Leaf number	Initial	2.31 ± 0.11	2.20 ± 0.07	2.29 ± 0.13	2.35 ± 0.11
	15 Days	4.21 ± 0.16	4.14 ± 0.11	4.22 ± 0.16	4.32 ± 0.14
	30 Days	6.10 ± 0.13 ^{a}	5.21 ± 0.19 ^{b}	6.15 ± 0.38 ^{a}	6.12 ± 0.49 ^{a}
	45 Days	19.20 ± 0.34 ^{a}	14.31 ± 0.84 ^{c}	16.32 ± 0.85 ^{b}	18.66 ± 0.89 ^{a}
	60 Days	16.87 ± 0.32 ^{a}	14.09 ± 1.52 ^{b}	14.22 ± 1.58 ^{b}	16.43 ± 0.38 ^{a}

There is a significant difference ($p < 0.05$) in the mean values (mean ± S.E.) of each row’s non-shared superscript.

Table 5. The effect of different feeding and fasting regimes (for Nile tilapia reared in the aquaponic system) on the nutritional composition of spinach leaves during a 60-day experimental period.

Parameters	Feeding Regimes			
	Daily Feeding	One-Day Feeding/ One-Day Fasting	Two-Day Feeding/ One-Day Fasting	Three-Day Feeding/ One-Day Fasting
Nitrogen (%)	5.04 ± 0.38	4.72 ± 0.31	4.77 ± 0.43	4.87 ± 0.33
Phosphorus (%)	0.53 ± 0.08	0.43 ± 0.07	0.45 ± 0.06	0.47 ± 0.05
Potassium (%)	3.19 ± 0.18 [a]	2.63 ± 0.29 [b]	2.89 ± 0.21 [ab]	3.13 ± 0.18 [a]
Calcium (%)	2.71 ± 0.21	2.40 ± 0.19	2.54 ± 0.29	2.47 ± 0.23
Magnesium (%)	1.46 ± 0.07 [a]	1.23 ± 0.04[b]	1.37 ± 0.09 [ab]	1.41 ± 0.06 [ab]
Sulphur (%)	0.32 ± 0.03 [a]	0.24 ± 0.02[b]	0.35 ± 0.03 [a]	0.38 ± 0.02 [a]
Copper (mg/ L)	9.26 ± 0.56	8.81 ± 0.17	8.46 ± 0.89	9.35 ± 0.21
Zinc (mg/L)	135.73 ± 15.89	141.09 ± 20.40	145.63 ± 9.91	139.70 ± 8.40

There is a significant difference ($p < 0.05$) in the mean values (mean ± S.E.) of each row's non-shared superscript.

Table 6. The effect of different feeding and fasting regimes (for Nile tilapia reared in the aquaponic system) on the pigment of spinach leaves during a 60-day experimental period.

Parameters	Feeding Regimes			
	Daily Feeding	One-Day Feeding/ One-Day Fasting	Two-Day Feeding/ One-Day Fasting	Three-Day Feeding/ One-Day Fasting
Chlorophyll a (mg/g FW)	1.16 ± 0.06 [a]	0.93 ± 0.12 [b]	0.95 ± 0.08 [b]	1.06 ± 0.11 [ab]
Chlorophyll b (mg/g FW)	0.22 ± 0.03 [a]	0.15 ± 0.02 [b]	0.19 ± 0.04 [ab]	0.20 ± 0.07 [ab]
Carotenoid (mg/g FW)	0.49 ± 0.02 [a]	0.33 ± 0.02 [c]	0.44 ± 0.02 [b]	0.46 ± 0.01 [ab]

There is a significant difference ($p < 0.05$) in the mean values (mean ± S.E.) of each row's non-shared superscript.

4. Discussion

There has been significant development of food production methods to meet the challenges of declining groundwater supplies, rising water demands, loss of soil fertility, and climate change. The aquaponic system has been one of the most important productive, creative, and sustainable development methods that has been applied in an advanced aquaculture and agricultural system to overcome food security concerns.

4.1. Water Quality in Tilapia Tank

Aquaponics crop production relies heavily on water quality, particularly in recirculating water systems. According to [32], declining water quality metrics have an impact on crop performance, quality, and/or yield, in addition to fish physiology, growth rate, and feed efficiency. Water quality during the experiment stayed within established standards for the rearing of *O. niloticus*, demonstrating that the water quality management of the culture system was adequate, regardless of feeding and fasting regimes [33,34]. During the experiment, the difference in feeding and fasting regimes of Nile tilapia did not significantly affect the pH, as the range was 7.20 to 8.09, which is generally considered a suitable range for the aquaponic system [35], asplants are considered to have access to nutrients in the pH range of 5.8 to 6.5. According to [36], nitrifying bacteria prefer a pH range of 7.0 to 9.0, whereas fish require a pH range of between 6.5 and 9.0 (Table 1). Dissolved oxygen levels decreased up to 12% as feeding days increased due to the oxygen needs of fish and microbes. Generally, the average DO in all treatments was slightly higher than the average DO level suggested for aquaculture, which is greater than 5 mg/L. Nitrifying bacteria need a DO range of 4–8 mg/L to maximize the nitrification operation [37]. The dissolved oxygen levels were sufficient in the aquaponics system. Many studies have reported that the electrical conductivity (EC) measured between 300 and 1100 s/cm is suitable for aquaponic solutions [36,38]. In a hydroponic solution, the TDS should be between 1000 and 1500 mg/L. Relatively low TDS concentrations (200–400 mg/L) will yield good results because nutrients are continuously created in the aquaponics system [36]. EC (636.33 ± 39.37–654.56 ± 4.68 µs/cm) and TDS (321.02 ± 3.45–361.72 ± 8.07 mg/L) were within the range of favorable conditions (Table 1). The calculated alkalinity was within the permissible range of 5–500 mg/L, and ranged from 35.62 0.26 to 36.87 0.61 ($p > 0.05$) [39]. In the aquaponics system, nitrification is a biological reaction that converts water from the toxic form (ammonia nitrogen, NH3-N) caused by fish in bio-filtration units to the non-toxic form nitrate (NO3-N), which the plant benefits from. According to [40], nitrite (NO2-N), a nitrification intermediate product, is also widely recognized as being toxic (at low concentrations) to both fish and plants: according to [38], the acceptable limit for ammonia in the aquaponics system is less than 1.0 mg/L. In our research, the level of total ammonia nitrogen increased ($p < 0.05$) as feeding days increased (0.103 ± 0.08–0.121 ± 0.05 mg/L) due to the increase in feed provided to the fish, but they were within the limits of nitrification. This was evidenced in the fact that no statistical differences appeared in the level of nitrate between the treatments ($p > 0.05$). According to [41], nitrate, produced as a result of nitrification, is the preferred type of nitrogen for plants due to its low toxicity levels. [38] concluded that an aquaponics system has an endurance limit of 150 mg/L for nitrate, whereas [42] found that the level of nitrate in aquaculture water should be 0.1–4.00 mg/L of nitrate. In the current experiment, the nitrate-nitrogen concentration in different feeding and fasting regimes ranged from 0.31 ± 0.02 to 0.37 ± 0.02 mg/L ($p > 0.05$). According to [43] the plant absorbs phosphorus in the form of ionic orthophosphate, whereas [44] determined that in an unprocessed aquaponic system, the orthophosphate level never rises above 2.0 mg/L but often hovers around 1.0 mg/L. In this experiment, it was found that the level of phosphate decreases significantly as feeding days decrease (0.63 ± 0.03–0.51 ± 0.02 mg/L), as a result of plant absorption. However, the phosphate level was, in all treatments, found to be within the desired range. The findings of the current study established that the calcium level ranged between 37.84 ± 0.57 and 38.35 ± 0.71 mg/L ($p > 0.05$). According to [45], the acceptable range for free calcium in fish culture water is 25–100 mg/L. A calcium dosage of 27 mg/L was recommended by [46] to enhance fish growth without damaging the spinach yield in the aquaponic system. According to [44], the normal aquaponic nutrition solution contains 12 mg/L of potassium. In this trial, there was no external potassium supplementation, hence the potassium level was lower (11.87 ± 0.41–11.42 ± 0.11 mg/L).

4.2. Nile Tilapia Performance

A 60-day experimental study was designed to minimize fish feed consumption based on feeding and short-term fasting regimes while maintaining maximum Nile tilapia growth performance and spinach fertilization, and maximize benefits through optimal and efficient use of resources from the NFT-based aquaponics system. To the best of the authors' knowledge, this is the first study to use feeding and fasting regimes in an NFT-based aquaponic system. In the aquaponic system, the different feeding and short-term fasting regimes significantly affected the growth performance (FBW, WG, SGR, and FCR) of Nile tilapia fingerlings during the current study. Strategic short-term fasting is used in fish farming to cut down on overfeeding expenses as well as to minimize handling stress, poor water quality, and infection-related mortality [47]. Fish definitely need energy during hunger in order to provide proteins that help reduce oxidative damage and metabolic irregularities in their cells [48]. In starvation, the body tries to maintain a good metabolism, as the body uses deposited proteins, fats and glycogen to adjust physiological balance. [19,49]. The fish's size, dietary habits, species, and environment all affect their endogenous metabolism for energy release [50], Some fish species rely on lipids, while others rely on protein [51]. Numerous studies have demonstrated that protein content is an appropriate primary nutrient for starvation compensation [19]. The daily feeding and three-day feeding/one-day fasting regimes of Nile tilapia did not notably influence the growth performance, and fish still displayed no statistical differences ($p > 0.05$). However, Nile tilapia fed on one-day feeding/one-day fasting showed lower growth performance than those in other regimes. The results did not show compensatory growth performance between the different regimes, which is not consistent with previous studies that reported saturated and timely refeeding programs influenced the expeditious compensatory growth of fish [52,53]. To the same extent, Nile tilapia showed compensatory growth performance following starvation regimes [21,22]. There are two reasons that may explain the absence of compensatory growth: (1) the short duration of fasting, and (2) the aquaponic system, where all excess feed and fish waste are passed periodically throughout the day to the mechanical and biological filter, which the plant depends on to supply it with nutrients.

Glucose, cortisol, and triglycerides are plasma biomarkers that are used as general indicators to monitor fish health, physiological state, and stress responses [54]. An increase in the level of these biomarkers indicators means a stress response for the fish [55]. The plasma cortisol concentration level was significantly higher in the one-day feeding/one-day fasting treatment than in other treatments for the Nile tilapia. The mean ± S.E. (3.87 ± 0.64 to 5.31 ± 0.59) plasma cortisol values obtained from four treatments compared positively with the reported normal mean basal cortisol range of 5–60 ng/mL for Nile tilapia [56]. Because glucose is an innate immunological parameter driven by stress, a rise in glucose concentration is a secondary stress response, and the degree of the increase is a stress indicator [57]. According to [56], electroshock and social stressors resulted in mean basal glucose concentrations of 39.6 and 34.2 mg/dL, respectively, which caused acute stress in Nile tilapia. These results are positive comparable to the total blood glucose mean level of 83.52 ± 4.63 mg/dL obtained from the four Nile tilapia treatments. The mean ± S.E. for blood glucose concentration level in the current study was statistically the highest in on-day feeding and one-day fasting at 89.11 ± 3.94 mg/dL than in other treatments (Figure 2). In reacting to starvation-induced stress, the stress hormones adrenaline, noradrenaline, and cortisol recruited and increased glucose to fulfill the energy need—this is one reason for an increase in the blood plasma glucose level in on-day feeding/one-day fasting treatment [58].

In light of the high cost of fish feed and running an aquaponic system, mainly *O. niloticus* culture, the objective of this study was to answer one question: if *O. niloticus* is to experience optimal growth, is it the case that the recommended quantity of feed and its nutritional composition must take into account rearing in the aquaponics system that is based on feeding/fasting regimes? The findings revealed that there are significant differences in the cost of tilapia feed, as the cost of feed decreases with the increase in feeding days (Table 3). These findings are in line with [59], who demonstrated that feeding and fasting schedules are a feasible and practical programs for lowering the production cost and improving the economic efficiency of Nile tilapia production.

4.3. Spinach Productivity

One of the most popular green vegetables in the *Chenopodiaceae* family, spinach *(S. oleracea)* is grown all over the world. Because of the useful qualities of its nutrients and non-essential chemical compounds, including vitamins and minerals, as well as its phytochemicals and bioactives, spinach is known to have significant health-promoting effects [60,61].

In aquaponics, plant productivity is indirectly related to fish feeding regimes, water quality, and microbial activity. The availability of nutrients in the solution within the aquaponic system is impacted by the feeding rate. Plant nutrition is more or less efficiently achieved by increasing or decreasing fish feeding [62], because the water's nitrate content is available for a longer period during the day. The findings of the current study showed that feeding and fasting regimes in aquaponics affected the productivity of spinach in terms of height, leaf count, diameter, and weight (Table 4). The maximum yield was noted in the treatment of daily feeding and the lowest in one-day feeding/one-day fasting. The lowest growth in one-day feeding/one-day fasting was due to insufficient nutrients, which reduce crop quality and yields [63]. The results showed a significant difference in the spinach yield from the second to the fourth harvest, as the lack of tilapia feeding days led to a lack of plant nutrients (Figure 3). The one-day feeding/one-day fasting treatment had the greatest decrease in spinach harvests. On the other hand, the abundance of nutrients was not significantly affected between the daily feeding and three-day feeding/one-day fasting regimes in spinach harvests [64]. Although the abundance of nutrients for the plant decreased when less fish were fed, no statistical differences were observed in the nutritional compounds of spinach leaves in nitrogen, phosphorus, calcium, copper, and zinc, while the results indicated a significant decrease in potassium, magnesium, and sulphur between the treatments (Table 5). It is clear that the decrease in plant nutrients had not reached the point where deficiency symptoms appear on the leaves [65–67]. The chlorophyll and carotenoid content in spinach leaves was also affected by the plant's nutrient abundance. The results showed that the levels of chlorophyll and carotenoid were the least significant in the one-day feeding/one-day fasting treatment (Table 6). No statistical differences were observed between daily feeding and three-day feeding/one-day fasting treatments. Also, no symptoms of pigment deficiency were observed in spinach leaves in all treatments. According to [68], chlorophyll "a" and "b", of spinach cultivated hydroponically were 1.37 ± 0.03 mg/g final weight and 0.68 ± 0.00 mg/g final weight, respectively.

5. Conclusions

Feed consumption is the principal factor influencing productivity and cost in aquaculture, as it plays a vital role in optimising production efficiency. The findings of the study indicate that the implementation of a feeding schedule consisting of three days of feeding followed by one day of fasting, using a nutritionally balanced feed containing 35% protein, in an aquaponic system based on the nutrient film technique (NFT), results in the greatest levels of productivity and profitability for Nile tilapia fingerlings and spinach, when compared to alternative treatment methods.

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