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Effects of stocking density and water flow rate on performance, water quality and economic benefits of African catfish larvae (*Clarias gariepinus* Burchell, 1822) in the aquaponic system integrated with *Azolla* fern

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ABSTRACT

The effect of stocking on fish performance is known is most systems, and for many fish. Meanwhile water flow rate remains an important factor in recirculating aquaculture systems. However less information is still available on the influence of aquaculture management practices on the performance of aquaponics. This study determined the effect of stocking density and water flow rate on growth performance of African catfish larvae (*Clarias gariepinus*) and *Azolla* spp. in catfish-*Azolla* aquaponics system. The 4 × 4 factorial experiment consisted of four larval stocking densities (1500 larvae/m^3, 3000 larvae/m^3, 4500 larvae/m^3 and 6000 larvae/m^3) and four water flow rates (0.25 L/min, 0.5 L/min, 0.75 L/min and 1 L/min) executed in triplicate. The stocking density and water flow rate had an interactive influence ($P < 0.05$) on the concentration of DO, TAN, and NO_3^-. The stocking density and water flow rate had an interactive influence ($P < 0.05$) on the final mean weight, SGR, and survival of *C. gariepinus* larvae, but not for FCR and yield ($P > 0.05$). The highest fish weight was achieved under combination of stocking density 3245 larvae/m^3 and water flow rate of 0.87 L/min, but growth will still be optimized upto a stocking density of 4850 larvae/m^3 but at higher combinations of water flow rates (> 0.875 L/min). The stocking density and water flow rate had an interactive influence ($P < 0.05$) on chlorophyll *a* concentration, but not fresh weight, dry weight and yield ($P > 0.05$). Enterprise budgets for aquaponic with different stocking densities and water flow rate for fish and *Azolla* production allowed all the treatments to post positive returns to risk and were viable investments. Cash flow projections were made over a 10 year period for each of the units with the net present value (NPV) and internal rate of return (IRR) revealed that sales of larval fish and *Azolla* sp. at stocking density of 1500 larvae/m^3 were not profitable. However, all the other stocking densities were profitable except at 3000 larvae/m^3 and water flow rate of 0.25 L/min. Although at all stocking density the revenue were able to cover the variable costs, this study recommends stocking at 3000 to 4500 larvae/m^3 and water flow rate of 0.75 L/min and 1.00 L/min.

1. Introduction

The rearing of fish and plants simultaneously, is regarded as an age-old polyculture tradition dating back to over 2000 years ago in several parts of tropical Asia (Somerville et al., 2014). Later improvements in the mid-1970s led to the development of aquaponics, which typically integrates tank-based animal aquaculture, usually in recirculating aquaculture systems (RAS), and conventional hydroponics (Baganz et al., 2022; Kralik et al., 2022). Fish is then raised in a volume of water that has been treated through sedimentation and biofiltration components. Sedimentation and biofiltration increase the concentration of non-toxic nutrients and organic matter, which are then channeled into the secondary tank containing economically important plants (mostly crops) (Colt et al., 2022; König et al., 2018). The most common recirculating aquaponic systems for plant production employ either a media filled raised bed, nutrient film technique (NFT), or floating raft system for the plant growing area (Rakocy et al., 2016; Wallace-Springer et al., 2022). Through advancements, over the past three decades, there has been a wide variety of system designs, protocols, plants, and aquatic animals reared (Colt et al., 2022; Palm et al., 2018).

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Fish species that can be cultured in traditional culture systems and/or adaptable to crowding conditions have been reared in aquaponics (Pérez-Urrestarazu et al., 2019). Although there are diverse species of fish cultured under both small-scale and commercial large-scale aquaponics system (Alarcón-Silvas et al., 2021; Fischer et al., 2021), most of the data available for fish performance in the aquaponics are based on tilapia such as Blue tilapia (*Oreochromis aureus*), Nile tilapia (*Oreochromis niloticus*), hybrid tilapia (*Oreochromis mossambicus* × *Oreochromis niloticus*), Nile tilapia 'red strain' hybrid (*Oreochromis niloticus* × *O. aureus*) and red tilapia (*Oreochromis* sp.) (Al-Tawaha et al., 2017; Barbosa et al., 2022; Hundley et al., 2018; Nakphet et al., 2017). There are also reports of catfish performance in the aquaponic system (Baßmann et al., 2017; Baßmann et al., 2020; Diatin et al., 2021; Oladimeji et al., 2020).

A wide variety of plants have been cultivated with diverse species of fish in aquaponics. These include lettuce (Ani et al., 2022; Cohen et al., 2018), tomatoes (Datta et al., 2018; Schmautz et al., 2016; Suhl et al., 2016), basil (Ferrarezi and Bailey, 2019; Stathopoulou et al., 2018), strawberries (Abbey et al., 2019; Curry et al., 2017), cucumber (Estrada-Perez et al., 2018), pumpkins (Oladimeji et al., 2018) and other herbaceous plants (Espinosa Moya et al., 2016; Knaus et al., 2018). However, there have been fewer attempts at using aquatic plants in aquaponics. The use of duckweed *Azolla* fern as a protein ingredient in fish feed has been previously reported (Mandal et al., 2010). The *Azolla* spp. occurring in irrigated rice fields is a prolific breeder (Oyange et al., 2019) with a high relative growth rate when grown individually in open waters (Kösesakal and Yıldız, 2019). Interest in *Azolla* as a protein ingredient is due to the high protein concentration of its biomass, reported between 200 and 400 g/kg of the dry weight (DW) (Lumsangkul et al., 2022; Nasir et al., 2022; Rahmah et al., 2022). *Azolla* also contains 79–160 g/kg lipids, which may contribute to the gross energy of whole *Azolla* feed while also containing up to 50 g/kg (poly)phenolic compounds (Brouwer et al., 2019). However, few research undertakings have evaluated its performance in an aquaponics system.

An understanding of the economic benefits of aquaponics in the past were based primarily on hypothetical situations (Engle, 2016). However, in recent years, there is increasing information highlighting the practical economics analysis of aquaponics (Lobillo-Eguíbar et al., 2020; Pinho et al., 2022; Rizal et al., 2018; Zappernick et al., 2022). Generally, aquaponics is more economically viable in areas with limited land and water resources (Tokunaga et al., 2015). The key economic factors to be considered in setting up an aquaponic system as a business include the total capital outlay, operating costs of the system, and market price (Quagrainie et al., 2018). Against this backdrop, there remains a question of whether aquaponics operations will or, at least, could be an economically viable enterprise. More important, is how management practices within the aquaponics will influence the economic viability of the enterprise.

Stocking density is an important factor that affects fish growth, biomass, feed utilization, gross fish yield, and economic returns (Watts et al., 2016). The water flow rate is a significant factor in the recirculating aquaponics system, that may be responsible for regulating the inflow and outflow of substances within the system and hence may have a direct effect on the performance of cultured organisms (Diem et al., 2017; Shete et al., 2016). Indeed, the correlation of water flow rate with fish growth performance, survival, and production in the aquaponic system has been previously established (Delaide et al., 2017; Endut et al., 2009; Hussain et al., 2015; Nuwansi et al., 2016). Nevertheless, knowledge of the combined effect of stocking density and water flow rate that optimize both fish and vegetable performance without compromising the water quality and the economic returns remains limited in aquaponics. Therefore the objective of the current study was to determine the effects of stocking density and water flow rate on the growth performance, water quality, and economic benefit of African Catfish (*Clarias gariepinus*)-lettuce (*Azolla* spp.) aquaponics.

2. Materials and methods

2.1. Aquaponic design and operation

The 48 aquaponics used in this study (Supplementary Fig. A.1) were similar in design. Each consisted of a water reservoir (0.5 m^3), a rearing tank measuring 0.054 m^3 (0.6 m × 0.3 m × 0.3 m), a sedimentation tank (0.1 m^3), a biofiltration tank (0.1 m^3) and a hydroponic tank (0.25 m^2), with a sump (0.1 m^3) to pump water back to the reservoirs. Water flow was independent of the water reservoir to the aquaponic unit to the sedimentation tank, to the biofiltration tank, then to the hydroponic unit that flows back to the sump, and then pumped back to the reservoir.

2.2. Experimental fish and Azolla spp.

Eight *Clarias gariepinus* broostocks (mean weight 800 ± 56 g) were selected from broodstock ponds at Masinga Fish farm and induced to breed using ovaprim at 0.5 ml/kg of fish. After ovulation, the fish were artificially stripped, eggs obtained, fertilized (Okomoda et al., 2018), and incubated in an aerated glass incubator of capacity 0.1215 m^3 (0.45 m × 0.9 m × 0.3 m^3). After three days, the hatchlings were fed for 5 days to satiation with *Artemia* nauplii at intervals of 3 h, after which they were fed with commercial diet (Aller Aqua, 45% crude protein) until the end of the experiment.

Using a scoop net, the *Azolla* spp. were harvested from four rice fields in Mwea Irrigation Scheme, Kenya (Supplementary Fig. A.2). Mwea is located approximately 100 km northeast of the Kenyan Capital Nairobi (Latitude 0°41′S and Longitude 37°20′ E) at an elevation of 1160 m above sea level. The local climate is tropical with two rainy seasons, a long rainy season from March to May and a short rainy season in October and November (Narita et al., 2020). The plants were transferred to the laboratory and cultivated on styrofoam rafts floating on the surface of the hydroponic troughs and transferred to the hydroponic system.

2.3. Experimental setup (fish stocking density and water flow rates) experiment

The setup consisted of a 4 × 4 factorial experiment comprising four larval stocking densities (1500 larvae/m^3, 3000 larvae/m^3, 4500 larvae/m^3, and 6000 larvae/m^3) and four water flow rates (0.25 L/min, 0.5 L/min, 0.75 L/min, and 1.0 L/min respectively) as factors. All the treatments were executed in triplicate. A total of 9720 11-day-old hatchery-raised African catfish larvae (*Clarias gariepinus*) were distributed randomly into the 48 aquaponic tanks at the respective stocking densities and water flow rates. Feeding began a day after stocking the fry using 0.3 mm commercial pellet feeds (Aller Aqua, 45% crude protein). The fry were fed four times daily (morning: 08:00 and 12:00 h and evening: 16:00 and 20:00 h) at 5% body weight for 30 days. Gravity-assisted water flow was maintained from the water reservoirs through the aquaponic, and hydroponic system to the sump where it was pumped back into the reservoir to create a recirculation flow.

2.4. Monitoring water quality

For this study, water samples were obtained from the RAS rearing tanks (although water quality was also monitored in the sedimentation/biofiltration tanks, hydroponic unit, and sump). Water quality analyzed included dissolved oxygen (DO) concentration, pH, and alkalinity, measured in situ using calibrated JENWAY 3405 electronic probes (Barloworld Scientific Ltd., Essex, UK), with independent probes for each variable. Samples for nutrient analyses were taken using a 3 L Van Dorn bottle. Before water sampling, bottles were pre-soaked in 0.5 ml of nitric and sulphuric acid solutions of 1:1 volume ratio, washed in 2 L of tap water, and rinsed three times in ultra-pure water and dried before the fieldwork. Water was filtered using 0.45 − μm cellulose acetate filters and the filtrates were used to determine concentrations of Total

Ammonia Nitrogen (TAN), NO_2-N, and NO_3−N by spectrophotometry following standard methods (Federation Water Environmental and Association, 2005).

2.5. Fish and plant growth performance parameters

In estimating the fish growth and survival for each experiment, 30 fish from each tank were sampled weekly and weighed to calculate the individual mean weight. Growth in weight of the fish was expressed as the specific growth rate (SGR, %/day) using the formula SGR (%/day) = $(e^g - 1)100$ where $g = (\ln(W_2) - (\ln(W_1))(t_2\text{-}t_1)^{-1}$ and W_2 and W_1 are weights on day t_2 and t_1 respectively. Mortalities were determined at each sampling date by counting the remaining fish in the tanks and percent survival was calculated as: the number of fish remaining in the tanks/Number of the stocked fry*100.

At the end of the trials, the fish were harvested using repeated netting, and the total yield was determined by weighing in a beam balance. Daily feed ration was calculated from the average individual wet weight determined by weighing two groups of five fish and their dry matter content estimated from a relationship between wet weight and dry weight. The calculation of the Food conversion ratio (FCR) was based on measurements of the dry matter content at the start and the end of the experiment defined as the total food ration (dry matter) per unit of dry fish weight and was calculated following earlier protocols (Verreth and Den Bieman, 1987).

Plant fresh weight was determined by weighing at least five plants using a Bonvoisin Lab scale 1000 g × 0.01 g high precision electronic analytical balance (London, UK). The plants were dried in an oven to a constant weight and dry weight was determined which was converted to yield in kg/m^3. Chlorophyll-*a* concentration was determined using the portable chlorophyll (Chl) meter (CL-01, Hansatech) (Cassol et al., 2008). Proximate analysis was done for the plant samples to measure the moisture content, total ash, ether extract, and crude fiber using standard laboratory methods (Horwitz et al., 1970; Thiex, 2009). The protein content was determined by measuring nitrogen (N × 6.25) levels using the Kjeldahl methods (Marcó et al., 2002). The Nitrogen Free Extracts (NFE) was determined as (g/kg): 1000 − (moisture content + crude protein + crude lipid + ash + fiber). Gross energy was calculated using conversion factors for protein, lipids, and carbohydrates (Sales, 2009).

Mineral element analysis in plant followed standard protocols (AOAC, 2005). Approximately 5 g of each sample (as wet weight basis) were placed in a Teflon digestion vessel and double acid digested with nitric acid (HNO_3) and perchloric acid ($HCLO_4$). Seven mineral elements (sodium Na, potassium K, calcium Ca, magnesium Mg, iron Fe, and zinc Zn) were determined using the Atomic Absorption Spectrophotometer model AA-6300 (Shimadzu, Japan). Phosphorus was determined colorimetrically by Spectronic 20 (Gallenkamp, UK) using the phosphovanado molybdate method.

2.6. Economic analysis of catfish-Azolla spp. aquaponics

All costs for the system construction and management were recorded. The total operation costs and cost of assets during the study was noted together with the overall yields of fish and *Azolla* sp. An enterprise budget was used to determine the revenue, costs and returns of the aquaponics system under different stocking densities and water flow rates. The profitability of the enterprise was analyzed using the net returns above variable costs. The breakeven price was calculated using the formula

$$\text{Breakeven price} = \frac{\text{Fixed cost per unit}}{1 - (\text{Variable cost per unit/Selling Price per unit})}$$

The underlining assumptions factored in the economic analysis were:

(i) Land was already in existence an thus not factored in the capital cost.
(ii) Some associated risks with aquaponic operations which include but not limited to; significant higher cost of operation, component failures and reliable supply of inputs were mitigated during the course of this study.
(iii) Three financial scenarios were proposed based on revenue and funding models.

Scenario 1: The setup and working capital was raised by equity and not by term loan. The revenue model was derived based on the minimum market price for plant products.

Scenario 2: Similar financing structure with scenario 1. However, the revenue model is based on the maximum realistic market price for plant products.

Scenario 3: The setup and working capital raised by bank term loan, while the revenue model is similar to Scenario 2.

The NPV was determined by calculating the difference between the present value of cash inflows and the present value of cash outflows over a period of time. This was done by discounting the cash flow to the present time using a discounted rate. For the NPV of an investment to be accepted, the value should be positive and rejected if the NPV is negative (Ross et al., 2008). NPV was used to determine the IRR. The NPV formula is as follows:

$$\text{NPV} = (\text{Cash flows})/(1+\text{r})^{\text{i}}$$

where I = Initial Investment (Total setup cost), Cash flows = Cash flows in the time period, r = Discount rate, and i = time period (10 years).

The IRR is a discount rate that makes the NPV of all the cash flows from a particular project to be equal to zero. Due to the nature of the IRR formula, however, it cannot be calculated analytically but either through trial-and-error or using software programmed to calculate IRR. The higher the IRR, the more profitable the venture. The IRR calculations was as follow:

$$0 = NPV = \sum_{n=0}^{N} \frac{CF_n}{(1+IRR)^n}$$

where: CFn = Net cash inflow during the period n, n = Each period, N = Holding period, NPV = Net Present Value, and IRR = Internal Rate of Return.

2.7. Statistical analysis

Data analyses were performed using STATISTICA 6.0. The dependent variables were fish and plant performance data which was ratio type. Fish performance data included mean weight, weight gain (%), specific growth rate (SGR), Food Conversion Ratio (FCR), survival and yield (kg/m^3). Plant growth performance metrics were measured in terms of plant fresh weight (g/plant wet wt) and plant dry weight (g/plant dry wt), chlorophyll *a* concentration and yield (kg/m^3). Fish and plant growth performance parameters per treatment were computed as means ± Standard Error. The independent variables (factors) was stocking density with four levels (1500 larvae/m^3, 3000 larvae/m^3, 4500 larvae/m^3 and 6000 larvae/m^3) and water flow rates with four levels (0.25 L/min, 0.5 L/min, 0.75 L/min and 1.0 L/min). The independent variables were quantitative measurable, and were maintained unchanged throughout the experimental period. Thus the entire experiment was 4 × 4 factorial experiment, with 4 × 4 = 16 treatments. These variables were assumed to be continuous as there could be possible infinite combination of treatment for the independent variables than the selected levels, thus the appropriate design is one that would optimized the interaction between the two sets of independent variables (Yossa and Verdegem, 2015). The relationship between larval growth under varying stocking densities and water flow rates were analyzed using response surface regression of the advanced regression model protocol (Quinn and Keough, 2002; Gotelli and Ellison, 2004). In the design a polynomial second order model of the form: $y = \beta_0 + \sum_{i=1}^{k}\beta_i x_i + \sum_{i=1}^{k}\beta_{ii}x_i^2 + \sum_{i=1}^{n}\sum_{j=1}^{m}\beta_{ij}x_i x_j + \ldots\varepsilon$.

The maximum combination of stocking density and water flow rate that optimize fish growth parameters was analyzed using orthogonal polynomial contrast procedure.

3. Results

3.1. Water quality changes

Water quality parameters in the aquaponics stocked with *Clarias gariepinus* larvae at varying stocking densities and water flow rate are shown in Table 1. The stocking density and water flow rate had an interactive influence ($P < 0.05$) on the concentration of DO, TAN, and NO_3^- (Table 1, Fig. 1). The DO levels suitable for fish growth (>4.5 mg/L) was achieved at stocking densities between 1500 and 6000 larvae/m^3 and flow rates above 0.65 L/min, however the stocking density and water flow that ensured maximal DO (>6.5 mg/L) occurred at any combination of stocking density from 1525 to 3000 larvae/m^3 and water flow rates ranging from 0.68 to 1.0 L/min. Minimum concentration of TAN (<0.34 mg/L) in the culture units would be achieved by any combinations of stocking density ranging from 1450 to 4250 larvae/m^3 and water flow rates ranging from 0.75 to 1.0 L/min. At concentration combining stocking density 4600 to 6000 larvae/m^3 and water flow rate of 0.25 to 0.42, TAN will be at highest level in the culture units (>0.54 mg/L). The minimum concentration of NO_3^- (0.16 mg/L) occurred in stocking density 1500 to 3550 larvae/m^3 and water flow rates 0.72 to 1.0 L/min. However high and detrimental values of TAN (>0.36 mg/L) was obtained at any combination of stocking 5650 to 6000 mg/L and water flow rate from 0.25 to 0.48 L/min. The concentration of NO_2-significantly ($P < 0.05$) increased with increasing density but was not affected the interaction between stocking density and water flow rate ($P > 0.05$). There were no significant interaction ($P > 0.05$) and main effect of pH, and alkalinity with variation in stocking density or water flow rate ($P > 0.05$).

3.2. Fish performance and survival under varying density and flow rate

The larvae grew from 0.76 mg at stocking to highest mean weight of 11.1 ± 1.3 mg. The stocking density and water flow rate had an interactive influence ($P < 0.05$) on the final mean weight, SGR, and survival of *C. gariepinus* larvae, but not for FCR and yield ($P > 0.05$) (Table 2, Fig. 2). The highest fish weight was achieved under combination of stocking density 3245 larvae/m^3 and water flow rate of 0.87 L/min (Fig. 2a). Nevertheless growth will still be optimized upto a stocking density of 4850 larvae/m^3 but at higher combinations of water flow rates (> 0.875 L/min). The decline observed in the growth of fish at higher density treatment regardless of water flow rates. Similarly, the SGR of fish was optimized by combination of stocking density 3240 larvae/m^3 and water flow rate of 0.88 L/min (Fig. 3b). Although survival was optimized by any combination of stocking density ranging from 1500 to 3345 larvae/m^3 and flow rate 0.72 to 1.0 L/min, the best combination that ensured maximum survival occurred at stocking density 2850 larvae/m^3 and water flow rate of 0.82 L/min.

3.3. Azolla fern performance under varying density and flow rate

The fresh weight, dry weight and yield of *Azolla* spp. during the production cycle in hydroponics differing in stocking densities and water flow rates are shown in Table 3 (Fig. 3). Fresh weight ranged from 106.9 kg in treatment stocked at 1500 larvae/m^3 and flow rate of 1.0 L/min, to the highest value of 333.1 kg in treatment stocked at 6000 larvae/m^3 and flow rate of 0.25 L/min. The yield of *Azolla* spp. ranged form 1.22 to 4.2 kg/m^3 dry wt. Generally, the stocking density and water flow rate had an interactive influence ($P < 0.05$) on chlorophyll *a* concentration (Fig. 3b), but not fresh weight, dry weight and yield ($P > 0.05$) (Table 3, Fig. 3). The dry weight and yield were influenced by stocking density where they increased with increasing stocking density.

The mineral composition of *Azolla* sp. as a feed ingredient is presented in Table 4. Whereas stocking density significantly influenced ($P < 0.05$) the concentration of all the mineral elements in aquaponics, there were no interactive effects between stocking density and water

Table 1

Water quality parameters (means ± SD) under varying stocking densities and water flow rates in a *C. gariepinus-Azolla* fern integrated aquaponic system.

Stocking density (larvae/m^3)	Water flow rate (L/min)	Water quality parameters					
		DO (mg/L)	pH	Alkalinity (mg/L $CaCO_3$)	TAN (mg/L)	NO_3-N (mg/L)	NO_2-N (mg/L)
1500	0.25	5.1 ± 0.6	7.2 ± 0.4	80.2 ± 9.2	0.46 ± 0.08	0.19 ± 0.04	0.024 ± 0.002
	0.50	5.7 ± 0.5	7.2 ± 0.2	82.4 ± 10.1	0.42 ± 0.07	0.19 ± 0.03	0.025 ± 0.004
	0.75	6.4 ± 0.4	7.3 ± 0.3	75.1 ± 7.5	0.39 ± 0.19	0.17 ± 0.05	0.023 ± 0.003
	1.00	6.9 ± 0.4	7.2 ± 0.4	76.3 ± 6.9	0.33 ± 0.19	0.15 ± 0.05	0.021 ± 0.003
3000	0.25	5.0 ± 0.3	6.8 ± 0.4	67.8 ± 6.3	0.50 ± 0.08	0.25 ± 0.04	0.045 ± 0.003
	0.50	5.1 ± 0.4	6.9 ± 0.2	82.9 ± 4.5	0.47 ± 0.07	0.22 ± 0.03	0.042 ± 0.005
	0.75	5.3 ± 0.7	6.9 ± 0.3	75.3 ± 4.4	0.42 ± 0.19	0.19 ± 0.05	0.044 ± 0.006
	1.00	5.7 ± 0.3	6.9 ± 0.4	76.2 ± 5.9	0.37 ± 0.19	0.16 ± 0.05	0.038 ± 0.004
4500	0.25	4.5 ± 0.5	6.5 ± 0.4	80.2 ± 9.2	0.53 ± 0.08	0.29 ± 0.04	0.045 ± 0.003
	0.50	4.7 ± 0.3	6.6 ± 0.2	82.4 ± 10.1	0.46 ± 0.07	0.28 ± 0.03	0.043 ± 0.006
	0.75	5.4 ± 0.6	6.8 ± 0.3	75.1 ± 7.5	0.42 ± 0.19	0.23 ± 0.05	0.043 ± 0.005
	1.00	5.8 ± 0.4	6.9 ± 0.4	80.2 ± 9.2	0.40 ± 0.19	0.19 ± 0.05	0.039 ± 0.005
6000	0.25	4.1 ± 0.6	6.4 ± 0.4	82.4 ± 10.1	0.57 ± 0.08	0.33 ± 0.04	0.042 ± 0.005
	0.50	4.4 ± 0.5	6.8 ± 0.2	75.1 ± 7.5	0.51 ± 0.07	0.29 ± 0.03	0.044 ± 0.006
	0.75	4.9 ± 0.4	6.9 ± 0.3	76.3 ± 6.9	0.45 ± 0.19	0.27 ± 0.05	0.042 ± 0.007
	1.00	5.4 ± 0.5	7.1 ± 0.4	76.2 ± 5.9	0.41 ± 0.19	0.22 ± 0.05	0.040 ± 0.009

	P values					
	DO (mg/L)	pH	Alkalinity (mg/L $CaCO_3$)	TAN (mg/L)	NO_3-N (mg/L)	NO_2-N (mg/L)
SD linear effects	0.0002	0.0563	0.9334	0.0003	<0.0001	<0.0001
SD quadratic effect	0.1616	0.2114	0.8451	0.5662	0.0032	0.0002
WFR linear effect	0.0001	0.3122	0.5582	<0.0001	<0.0001	0.5227
WFR quadratic effect	0.1317	0.0982	0.7871	0.1342	0.1345	0.3162
SD × WFR effect	0.0012	0.4936	0.8339	0.04664	0.0042	0.4639

SD: Stocking density (larvae/m^3); WFR: Water flow rate (L/min).

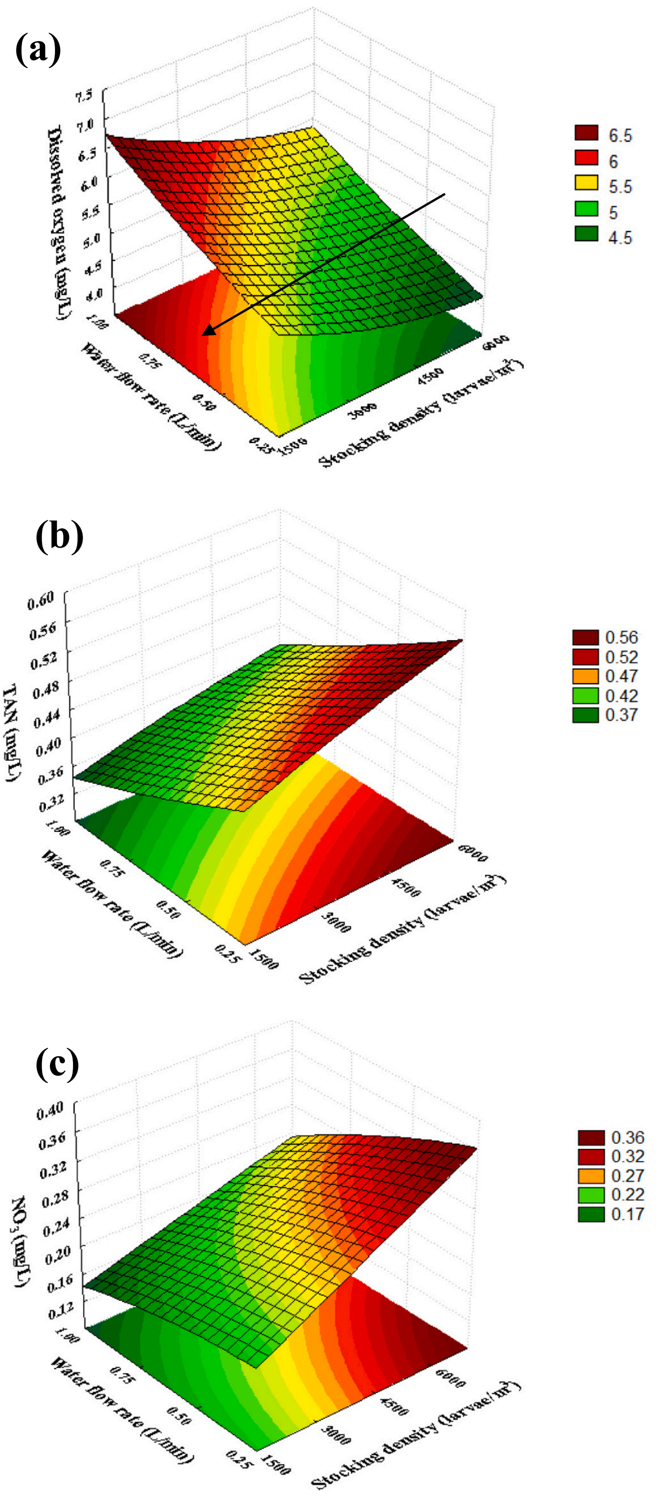

Fig. 1. Dissolved oxygen DO (a), Total Ammonia Nitrogen (TAN) (b) and Nitrites (NO$_3^-$) (c) concentrations under varying stocking densities and water flow rates in a *C. gariepinus-Azolla* fern integrated aquaponic system. Panel a, b and c: Interaction effect between stocking density and water flow rate ($P < 0.05$).

flow on any mineral element ($P > 0.05$). The minerals showed decreasing concentrations at higher fish stocking densities. The proximate composition of *Azolla* sp. as a feed ingredient is presented in Table 5. Except for NFE, there were no interactive effects between stocking density and water flow on any proximate composition of the *Azolla* sp. ($P > 0.05$) Crude protein levels and crude ash were affected by

Table 2
Clarias gariepinus larvae growth parameters and yield under different stocking densities and water flow rate in aquaponics (data are presented as means ± SE).

Parameter	Stocking densities (larvae/m^3)															
	1500				3000				4500				6000			
	Water flow rates															
	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0
Mean weight (mg)	4.5 ± 0.5	6.9 ± 0.8	10.1 ± 0.2	11.1 ± 0.3	3.9 ± 0.5	5.5 ± 0.7	9.0 ± 0.1	8.9 ± 0.1	3.7 ± 0.4	5.5 ± 0.7	6.0 ± 0.7	7.5 ± 0.9	3.2 ± 0.4	3.4 ± 0.4	4.2 ± 0.5	4.4 ± 0.5
Weight gain (%)	481.8 ± 2.5	826.6 ± 4.2	1231.6 ± 5.8	1346.8 ± 4.3	417.9 ± 4.5	628.9 ± 6.2	1079.7 ± 5.2	1091.3 ± 3.4	385.5 ± 5.1	619.5 ± 4.4	695.3 ± 3.5	904.0 ± 9.0	303.9 ± 11.3	346.8 ± 5.6	445.5 ± 8.9	478.9 ± 4.5
SGR	2.95 ±	3.68 ±	4.31 ±	4.47 ±	2.73 ±	3.30 ±	4.11 ±	4.10 ±	2.63 ±	3.14 ±	3.45 ±	3.82 ±	2.32 ±	2.51 ±	2.84 ±	2.92 ±
FCR	1.45 ±	1.41 ±	0.83 ±	0.83 ±	1.68 ±	1.61 ±	0.98 ±	1.02 ±	1.59 ±	1.55 ±	1.34 ±	1.23 ±	1.90 ±	1.91 ±	1.28 ±	1.19 ±
Survival (%)	75.1	92.4	98.2	99.2	68.7	81.1	95.4	94.3	67.7	81.2	87.5	95.4	65.9	63.7	73.7	73.2
Fish yield	60.9 ± 10.5	75.1 ± 11.9	80.1 ± 12.3	80.0 ± 16.7	60.9 ± 14.5	150.1 ± 16.5	238.6 ± 10.4	321.4 ± 12.4	165.4 ± 15.6	197.1 ± 12.6	213.1 ± 13.4	232.1 ± 16.1	61.1 ± 18.2	149.7 ± 19.5	238.6 ± 22.3	321.4 ± 23.4

	P values				
	Mean weight	SGR	FCR	Survival (%)	Fish yield
SD linear effects	0.0342	<0.0001	0.0035	0.0001	0.021
SD quadratic effect	0.1729	0.0324	0.7844	0.1387	0.3221
WFR linear effect	0.0021	<0.0001	0.0012	<0.0001	0.0421
WFR quadratic effect	0.052	0.0209	0.049	0.8241	0.3212
SD × WFR effect	0.0213	0.02721	0.2142	0.0462	0.3364

SD: Stocking density (larvae/m^3); WFR: Water flow rate (L/min). SGR: Specific growth rate (%/day), FCR: Food conversion ratio.

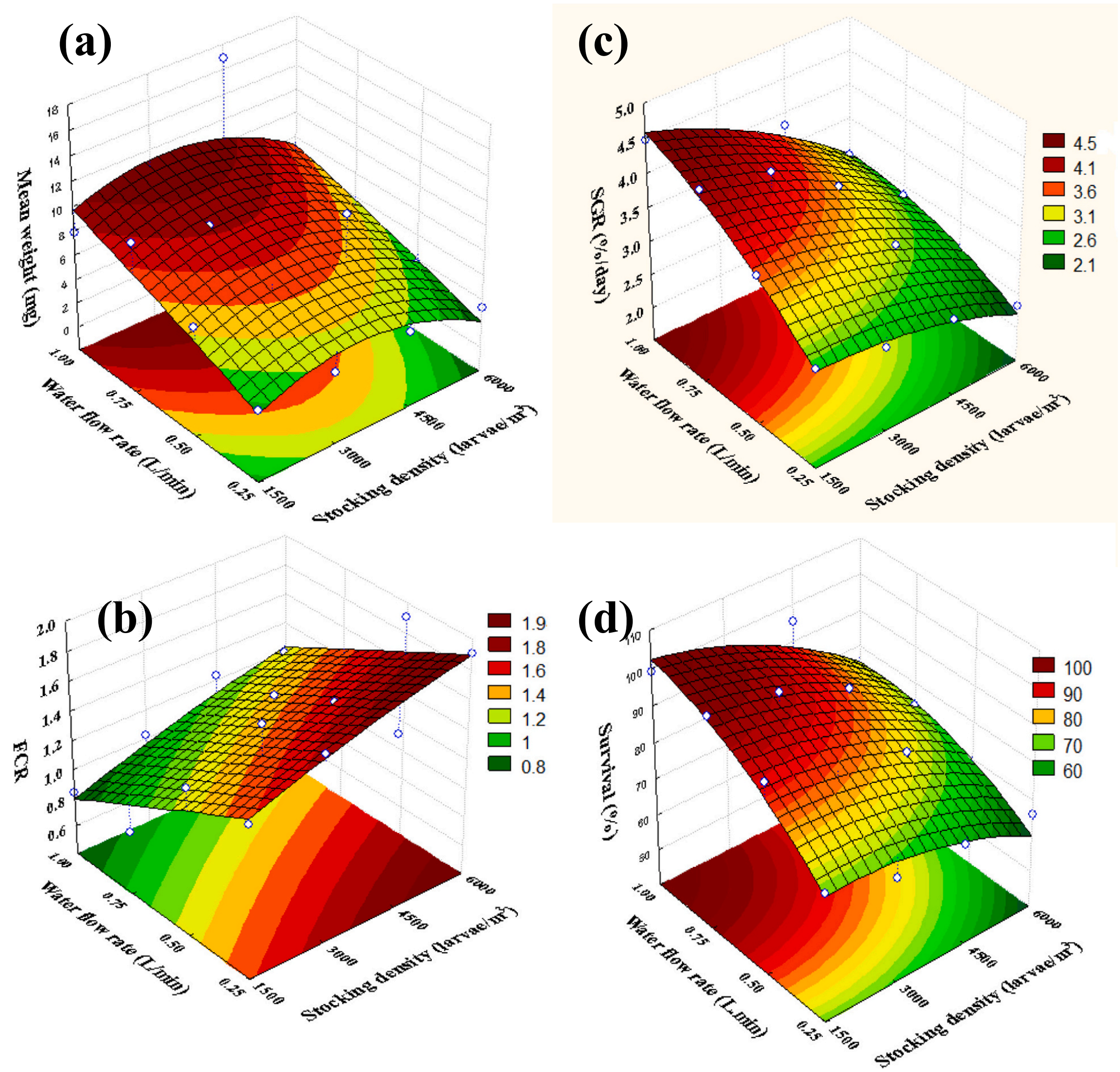

Fig. 2. Mean weight (a), specific growth rate (SGR) (b) Food Conversion Ratio (FCR) (c) and survival (d) of *C. gariepinus* larvae cultured under varying stocking densities and water flow rates in a *C. gariepinus-Azolla* fern integrated aquaponic system. Panel a, c and d: Interaction effect between stocking density and water flow rate ($P < 0.05$).

stocking density, where the protein levels increased with increasing stocking density.

3.4. Economic analysis of stocking density and flow rates of aquaponics system

An initial capital outlay required in financing the development and construction of a single unit of aquaponic was USD 406 (Supplementary Table A.1). Each unit of the model system was composed of a reservoir tank, rearing glass tanks fitted with airstones, tubing and filters, hydroponic unit and a sump. The table was subdivided into larvae and *Azolla* (hydroponic) components for analysis of the costs associated with each component. The African catfish larvae and *Azolla* sp. production components cost $ 34.10 and $ 6.50 respectively. Water flowed from the reservoir by gravity into larvae rearing glass tanks, biofilteration unit, the hydroponic units and sump.

Enterprise budgets for aquaponic farms with different stocking densities and water flow rates for fish and *Azolla* production is shown in Table 6. Only one manager was required for all farm sizes. One maintenance employee was needed at the farm. The maintenance employee's wage was equally divided between the enterprises. Two hired laborer was needed for the operation of the aquaponics system. Hired labor wages were $ 100.00 per month. The employees were needed to operate the hatchery and greenhouse facilities. Due to the high revenues gained by fingerlings and *Azolla* sp. sales, all variable and fixed costs were covered. All the treatments posted positive returns to risk and were viable investments. The break-even prices for variable costs were able to cover the cost of fish in the local market as they were below the sale price of Ksh 10 per case that the farm was able to receive in the market.

Cash flow projections were made over a 10 year cycle within a year for each of the units (Table 7). There was an initial cash outlay to purchase capital items for the construction of the facility. In the following years there were operating expenses and revenues generated from sales. Capital items with useful lives of <20 years were replaced at the end of their depreciation period and those costs were subtracted from revenues for that year. The net present value (NPV) and internal rate of return

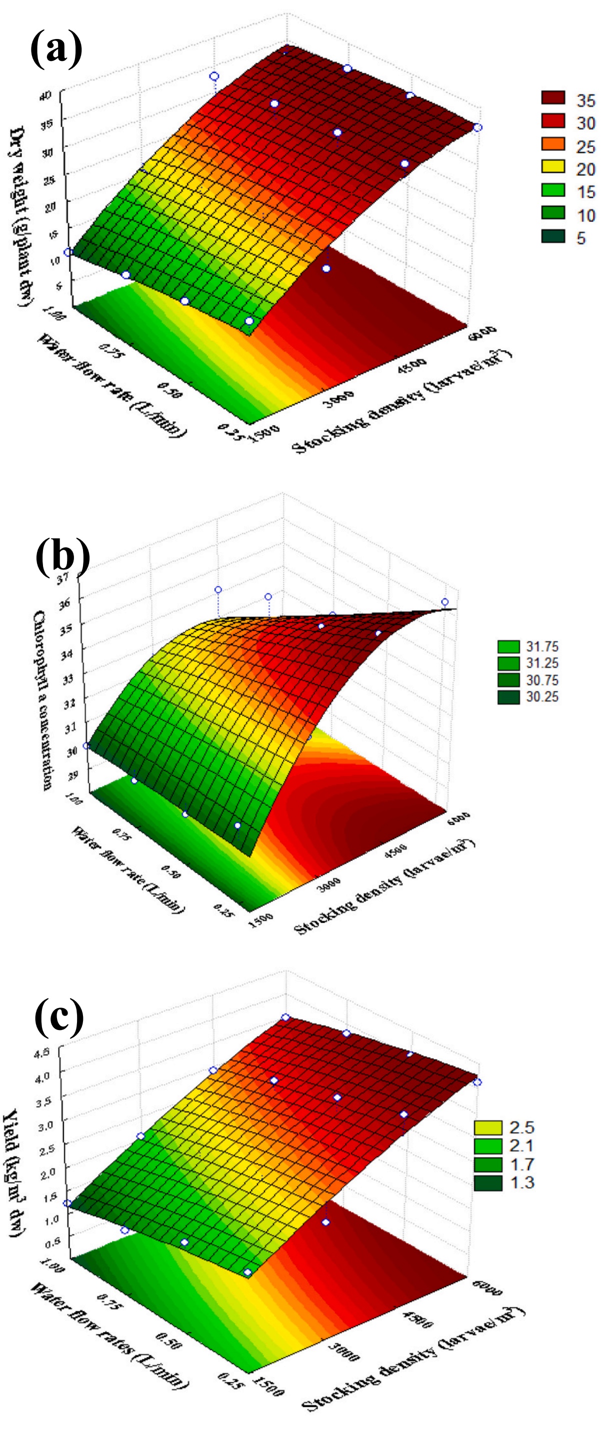

Fig. 3. Plant dry weight (a), chlorophyll *a* concentration (b), and yield (c) under varying stocking densities and water flow rates in a *C. gariepinus-Azolla* fern integrated aquaponic system. Panel b: Interaction effect between stocking density and water flow rate ($P < 0.05$).

(IRR) were calculated for each of the model unit. A discount rate of 18% was used to calculate the NPV. It was observed that sales of fish and *Azolla* sp. at stocking density of 1500 larvae m^{-3} were not profitable regardless of the water flow rates. However, all the other stocking densities were profitable except at 3000 larvae/m^3 and water flow rate of 0.25 L/min and increased with increased water flow rate and stocking density. Investors must determine their requirements for acceptable returns when choosing their farm size. If the IRR is too low, than other investment opportunities must be found.

Table 3
Azolla sp. growth parameters and yield under different stocking densities and water flow rate in aquaponics (data are presented as means ± SE).

Parameter	Stocking densities															
	1500				3000				4500				6000			
	Water flow rates (L/min)															
	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0
Plant fresh weight (g/plant wet wt)	173.1 ± 12.3	162.3 ± 10.5	114.3 ± 9.8	106.9 ± 9.23	203.2 ± 10.2	204.0 ± 11.5	199.3 ± 14.2	180.3 ± 13.7	321.5 ± 20.3	315.7 ± 18.9	307.3 ± 20.3	298.2 ± 24.5	333.1 ± 19.7	328.2 ± 30.3	319.4 ± 21.5	289.3 ± 23.4
Plant dry weight (g/plant dry wt)	19.0 ± 2.1	15.4 ± 2.3	13.2. ± 2.3	10.4 ± 1.7	22.3 ± 2.2	22.4 ± 2.4	21.9 ± 2.5	19.8 ± 2.3	35.3 ± 2.6	34.7 ± 3.6	33.8 ± 2.4	32.8 ± 2.9	36.6 ± 3.2	36.1 ± 3.7	35.1 ± 3.3	31.8 ± 4.2
Yields (kg/m^3 dry wt)	2.13 ± 0.17	1.96 ± 0.21	1.42 ± 0.22	1.22 ± 0.24	2.50 ± 0.24	2.49 ± 0.25	2.20 ± 0.24	2.07 ± 0.24	4.06 ± 0.31	3.71 ± 0.28	3.36 ± 0.34	2.91 ± 0.25	4.12 ± 0.32	3.99 ± 0.32	3.80 ± 0.40	3.48 ± 0.34
Leaf chlorophyll *a* concentration (μg/g)	31.2 ± 2.9	30.1 ± 2.7	30.0 ± 2.4	30.0 ± 2.2	33.4 ± 2.7	33.1 ± 3.2	33.0 ± 2.7	32.1 ± 2.3	36.2 ± 2.1	35.2 ± 3.2	35.1 ± 2.4	34.1 ± 2.8	36.3 ± 2.1	34.2 ± 2.4	33.2 ± 2.6	30.1 ± 3.1

	P values			
	Plant fresh weight	Plant dry weight	Plant yields	Leaf chlorophyll *a*
SD linear effects	<0.0001	<0.0001	<0.0001	<0.0001
SD quadratic effect	0.0391	0.0241	0.2542	0.003
WFR linear effect	0.03412	0.0312	0.0051	0.0007
WFR quadratic effect	0.4221	0.211	0.422	0.1421
SD × WFR effect	0.421	0.2375	0.311	0.0012

SD: Stocking density (larvae/m^3); WFR: Water flow rate (L/min).

Table 4
Mineral composition of *Azolla* spp. grown in hydroponics systems (g/ kg dw).

Parameter	Stocking densities															
	1500				3000				4500				6000			
	Water flow rates (L/min)															
	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0
N	28.2 ± 1.1	27.2 ± 0.9	26.2 ± 1.2	24.6 ± 0.6	29.4 ± 2.1	27.2 ± 1.9	28.2 ± 2.3	27.2 ± 2.2	32.3 ± 1.9	32.1 ± 2.4	31.2 ± 2.4	30.2 ± 2.8	34.2 ± 2.4	34.1 ± 2.1	33.1 ± 2.5	30.5 ± 2.2
P	7.2 ± 0.5	7.2 ± 0.3	7.0 ± 0.4	6.7 ± 0.3	7.4 ± 0.4	7.3 ± 0.5	7.3 ± 0.4	7.1 ± 0.5	7.7 ± 0.4	7.6 ± 0.5	7.3 ± 0.4	7.3 ± 0.3	8.2 ± 0.5	8.1 ± 0.4	7.8 ± 0.5	7.1 ± 0.4
K	72.3 ± 6.7	70.2 ± 6.2	70.1 ± 8.4	66.7 ± 5.4	74.5 ± 4.5	74.6 ± 5.3	74.1 ± 6.6	69.3 ± 6.7	75.5 ± 6.1	75.2 ± 6.9	74.6 ± 5.6	73.4 ± 5.2	76.5 ± 4.2	76.3 ± 7.2	76.1 ± 7.0	73.3 ± 6.3
Ca	11.3 ± 2.4	11.3 ± 1.5	11.7 ± 1.1	11.2 ± 0.9	11.1 ± 1.3	10.8 ± 0.9	10.5 ± 0.8	10.1 ± 0.9	9.5 ± 0.8	9.3 ± 0.9	9.4 ± 0.7	9.5 ± 0.7	9.1 ± 0.8	9.4 ± 0.8	9.1 ± 0.6	8.1 ± 0.6
Mg	7.5 ± 0.8	7.4 ± 0.7	7.5 ± 0.7	7.3 ± 0.5	7.3 ± 0.6	7.2 ± 0.5	7.3 ± 0.7	7.4 ± 0.5	7.1 ± 0.4	7.1 ± 0.5	7.0 ± 0.6	7.1 ± 0.4	6.9 ± 0.7	6.7 ± 0.8	6.8 ± 0.6	6.1 ± 0.5
Fe	0.14 ± 0.03	0.15 ± 0.03	0.13 ± 0.03	0.15 ± 0.04	0.13 ± 0.04	0.12 ± 0.05	0.13 ± 0.03	0.14 ± 0.04	0.11 ± 0.05	0.14 ± 0.06	0.13 ± 0.05	0.13 ± 0.03	0.09 ± 0.04	0.10 ± 0.02	0.08 ± 0.02	0.06 ± 0.01
Na	1.9 ± 0.2	2.2 ± 0.3	2.3 ± 0.3	2.4 ± 0.3	2.2 ± 0.3	2.3 ± 0.4	2.1 ± 0.2	2.3 ± 0.3	2.1 ± 0.3	2.0 ± 0.2	2.1 ± 0.2	1.9 ± 0.2	1.9 ± 0.2	1.8 ± 0.2	1.2 ± 0.2	1.1 ± 0.1

	P values						
	N	P	K	Ca	Mg	Fe	Na
SD linear effects	<0.0001	<0.0001	<0.0001	0.0002	0.0002	0.0018	<0.0001
WFR linear effect	0.9300	0.4002	0.0125	0.4812	0.0782	0.0445	0.0083
SD quadratic effect	0.0009	0.0001	0.0001	0.0321	0.1221	0.9321	0.1146
WFR quadratic effect	0.4623	0.1405	0.0122	0.2682	0.3211	0.6012	0.4726
SD × WFR effect	0.5124	0.1486	0.2211	0.3431	0.0927	0.2411	0.0601

4. Discussion

4.1. Water quality response to stocking density and water flow rates

Water quality is an important factor that influences the growth of organisms. With exception of few cases throughout the experiment, water quality across all the treatments were within the favorable range required for many species of fish even under high stocking density (Hasan et al., 2017). Although the analyzed water quality parameters were generally within the range of previous studies (Estim et al., 2019; Maucieri et al., 2018; Oladimeji et al., 2018), several water quality parameters were significantly influenced by stocking density and water flow rates. The dissolved oxygen ranging from 4 to 7 mg/l and was found within suitable ranges of *C. gariepinus* culture (Mallya, 2007). The reported tolerable levels of DO for *Clarias gariepinus* is 2.8 to 6.8 mg/L (Momoh and Solomon, 2017), a scheme that does not take into consideration the stocking density and water flow rate. At higher stocking densities, the concentration of DO was depleted probably due to high oxygen consumption by fish, higher organic matter accumulation, feed residues and feces of fish (Al Tawaha et al., 2021; Rayhan et al., 2018), which may potentially increase the microbial consumption of oxygen for their oxidation. The increased flow rate therefore appeared to increase the concentration of DO apparently through improved aeration. Increased DO levels owing to higher water exchange rates or aeration are in agreement with several studies in conventional fish rearing units (Boyd et al., 2018; Yi and Lin, 2001) and in aquaponics (Anando et al., 2022; Wu et al., 2018). Indeed it has been reported that regulation of water flow rates achieve optimal dissolved oxygen (DO) levels in the culture system and limit the accumulation of fish metabolites and other wastes (Schram et al., 2009). Therefore optimal combinations of stocking densities and water flow rate assured the highest levels of DO is encouraged.

The main sources of ammonium in culture systems are fish excretion and decomposition of feeds (Robles-Porchas et al., 2020). Therefore, at low stocking density, the concentration of ammonia is expected to be low in water. In this study, minimum concentration of TAN (<0.34 mg/L) in the culture units is achievable by combinations of low stocking density ranging from 1450 to 4250 larvae/m^3 and high water flow rates ranging from 0.75 to 1.0 L/min. This seems to suggest that concentration of TAN generally increased in tandem with increasing stocking density but may be modulated by high water flow rate similar to a previous study on African catfish (*Clarias gariepinus*) and water spinach (*Ipomoea aquatica*) aquaponic system (Endut et al., 2009). At higher water flow rates, there is more oxygenation in the water media that breakdown ammonia to less toxic forms thus reducing their bioaccumulation. At high stocking density of 4600 to 6000 larvae/m^3 and low and water flow rate of 0.25 to 0.42, there was high concentrations of TAN which pose threat to the culture of aquatic organism owing to its ability to diffuse across cell membranes (Franklin and Edward, 2019).

Nitrate-N is relatively nontoxic to fish and is not a considerable health hazard except at exceedingly high levels (Stone and Thomforde, 2004). The minimum concentration of NO_3^- (0.16 mg/L) occurred in stocking density 1500 to 3550 larvae/m^3 and water flow rates 0.72 to 1.0 L/min suggesting removal of NO_3^- from the water column. A number of mechanisms are responsible for the removal of NO_3-N from the culture units. In water, ammonium is converted rather rapidly to nitrite (NO_2) and nitrate (NO_3) by aerobic bacteria from the genera Nitrosomonas and Nitrobacter, through nitrification. Aeration is one of the mechanism that facilitates the process of nitrification (Åmand et al., 2013) and has been found to result in removal of ammonia, nitrate and nitrates in wastewater (Uggetti et al., 2016). There are two main mechanisms for the removal of NO_3-N, of which the first one is plant uptake from the growth medium and the second being microbial assimilation in the water column (Shete et al., 2016). The reduction in NO_3 at higher flow rate has also been shown. In this study there was a reduction of NO3-, at increased high water flow rates which concur with previous a study which showed that the removal percentage of nitrate-N increased with the increasing flow rates from 0.8 to 1.6 L/min and decreased with the increasing flow rates from 1.6 to 4.0 L/min (Endut et al., 2009). Although daily water flow rate may help in controlling the

Table 5
Proximate composition of *Azolla* spp. grown in the hydroponics systems (g/kg dw).

Stocking density (Larvae/m^3)	Water flow rate (L/min)	Crude protein	Crude lipids	Crude fiber	Crude ash	NFE
1500	0.25	28.2 ± 1.1	7.2 ± 0.6	71.3 ± 5.9	72.3 ± 6.7	72.3 ± 7.1
	0.50	27.2 ± 0.9	7.2 ± 0.5	70.2 ± 6.7	70.2 ± 6.1	70.2 ± 6.8
	0.75	26.2 ± 1.2	7.0 ± 0.5	72.1 ± 7.2	70.1 ± 5.7	70.1 ± 6.6
	1.00	24.6 ± 0.6	6.7 ± 0.7	69.7 ± 8.2	66.7 ± 5.2	66.7 ± 6.1
3000	0.25	29.4 ± 2.1	7.3 ± 0.5	71.5 ± 7.8	74.5 ± 5.7	74.5 ± 6.2
	0.50	27.2 ± 1.9	7.3 ± 0.6	72.6 ± 8.1	74.6 ± 5.6	74.6 ± 6.6
	0.75	28.2 ± 2.3	7.3 ± 0.8	71.1 ± 7.7	74.1 ± 5.4	74.1 ± 5.5
	1.00	27.2 ± 2.2	7.1 ± 0.6	69.3 ± 7.5	69.3 ± 5.1	69.3 ± 5.6
4500	0.25	32.3 ± 1.9	7.4 ± 0.6	72.5 ± 7.1	75.5 ± 5.6	75.5 ± 5.2
	0.50	32.1 ± 2.4	7.3 ± 0.7	71.2 ± 6.5	75.2 ± 4.4	75.2 ± 7.1
	0.75	31.2 ± 2.4	7.3 ± 0.7	69.6 ± 6.6	74.6 ± 5.0	74.6 ± 5.9
	1.00	30.2 ± 2.8	7.1 ± 0.5	70.4 ± 6.2	73.4 ± 5.3	72.4 ± 6.9
6000	0.25	34.2 ± 2.4	7.2 ± 0.7	70.5 ± 7.2	76.5 ± 5.2	72.5 ± 4.9
	0.50	34.1 ± 2.1	7.0 ± 0.4	71.3 ± 5.4	76.3 ± 6.1	72.3 ± 5.5
	0.75	33.1 ± 2.5	7.2 ± 0.5	70.1 ± 6.3	76.1 ± 6.2	72.1 ± 5.7
	1.00	30.5 ± 2.2	7.0 ± 0.5	71.3 ± 7.5	73.3 ± 5.2	72.3 ± 5.6

	P value				
	Crude protein	Crude lipids	Crude fiber	Crude ash	NFE
SD linear effects	<0.0001	0.2051	0.9064	<0.0001	0.0095
WFR linear effect	0.9341	0.0722	0.6858	0.1722	0.0007
SD quadratic effect	0.0009	0.0004	0.0811	0.0003	0.0002
WFR quadratic effect	0.4621	0.0192	0.6832	0.0122	0.0457
SD × WFR effect	0.8124	0.1425	0.5536	0.0851	0.0091

nitrate concentration, the levels at any combination of stocking 5650 to 6000 mg/L and water flow rate from 0.25 to 0.48 L/min in the current study was still found to be high and therefore highly should be reduced in the aquaponics.

4.2. Growth response of fish to stocking density and water flow rates

The mean weight of *Clarias gariepinus* larvae under different stocking densities and water flow rate ranging from 0.76 mg at stocking to highest mean weight of 11.1 ± 1.3 (approximately 1346.8 ± 4.3%) has been reported in many culture units (Kemigabo et al., 2019; Olurin and Oluwo, 2010; Onura et al., 2018). A general observation was that growth will still be optimized upto a stocking density of 4850 larvae/m^3 but at higher combinations of water flow rates ($>$ 0.875 L/min), suggesting that water flow rates exert much influence on fish weight. These studies compares well with those of (Endut et al., 2009) who obtained the second highest specific growth rate of fish with higher water flow rate of 0.8 L/min; however, maintaining the flow rate of 1.6 L/min gave the best production performance of fish. Meanwhile it has also been previously shown that maximum growth of goldfish was obtained in 12- and 24-h/day with improved water flow rates (Shete et al., 2013). A comparison of reciprocating flow versus constant flow in an integrated, gravel bed, aquaponic test system, failed to establish any significant difference between constant flow and reciprocating system considering the fish growth (Lennard and Leonard, 2004). The reduced fish weights at higher stocking density, may be attributed to competition for space and food at higher density as well as possible deterioration of water quality parameters, which would be tremendously improved through improved water flow. The decline observed in the growth of fish at higher density treatment regardless of water flow rates was associated with a poor quality of water especially lower DO and high ammonia concentration. In contrast, some authors have observed a reduction in growth rates at increasing density regardless of the water quality, which implies that physiological stress due to crowding and competition for feed and living space could inhibit the growth of fish (Maucieri et al., 2019).

The SGR values ranging from 2.6 to 4.5 is not far above the recommended value of 1.7 to 2.5 for culture of African catfish larvae in a several culture system (Endut et al., 2010; Nwipie et al., 2015) but were generally higher than in most studies. These high values may be related to the ideal growth conditions in aquaponics to sustain maximal growth for the 30 days trial period. The growth of larvae at SGR of fish was optimized by combination of stocking density 3240 larvae/m^3 and water flow rate of 0.88 L/min.

Survival was optimized by any combination of stocking density ranging from 1500 to 3345 larvae/m^3 and flow rate 0.72 to 1.0 L/min, the best combination that ensured maximum survival occurred at stocking density 2850 larvae/m^3 and water flow rate of 0.82 L/min of *Clarias gariepinus* under stocking density and flow rates also differed. At stocking density and water flow rate of 0.75 and 1.0 L/min, survical averaged 94–95% which was higher survival in comparison to flow rates of 0.25 L/min (68.7%) and 0.5 L/min (81.2%). At the highest stocking density of 6000 larvae/m^3, survival was poor at all the water flow rates. The reduced survival at high stocking density is due to deterioration of water quality parameters.

4.3. Azolla fern performance under varying density and flow rate

During the study, yield of *Azolla* spp. ranged form 1.22 to 4.2 kg/m^3 dry wt which is higher than those reported in other studies (Oyange et al., 2019). Based on the median range of production values reported it was estimated that 452,000 kg of plants was harvested in each aquaponics unit within 12 months. The study further established that production in terms of fresh weight, dry weight and yield of *Azolla* sp. increased with increasing fish stocking density but were not affected by water flow rates. The observed increase in plant production at high density may be related to high nutrient released from the recirculating units and availed to plants mainly in the form of NO_3-. Although a high stocking fish density was observed to result to lower growth rates of water spinach due to promotion of denitrification as discussed for other similar studies (Endut et al., 2010). It is surprising that water flow rate did not interact with stocking density in influencing plant production in which water flow rates alone affected growth of water spinach (*Ipomea aquatica*) in an aquaponic system (Endut et al., 2009). In a different study, comparing three water flow rates (3.2, 1.5, and 1.0 L/min) it was established that fish and spinach growth increased with increasing water flow rate from 1.0 L/min to 1.5 L/min (Hussain et al., 2014). Similarly, poor growth performance of spinach was established at reduced water circulation period (Shete et al., 2013). Perhaps it is possible to suggest that flow rate was effective in affecting plant growth due to optimal fish stocking densities during their growth trials.

Stocking density significantly influenced the concentration of all the mineral elements in aquaponics, but there were no interactive effects between stocking density and water flow on any mineral element. The minerals showed decreasing concentrations at higher fish stocking densities, which may be related to supply of nitrogenous substances

Table 6
Enterprise budgets for *Clarias gariepinus* larvae and *Azolla* sp. production component of a model aquaponic at different stocking densities and water flow rate.

Parameters	Stocking densities (larvae/m^3)															
	1500				3000				4500				6000			
	Water flow rates (L/min)															
	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0	0.25	0.5	0.75	1.0
Number of fish stocked	81	81	81	81	162	162	162	162	243	243	243	243	324	324	324	324
Survival (%)	75.1	92.4	98.2	99.2	68.7	81.2	95.4	94.3	67.7	81.2	87.5	95.4	65.9	63.7	73.8	73.2
Harvest weight (g/tank)	61	75	80	80	61	150	239	321	165	197	213	232	61	150	239	321
Total revenue from larvae	6.09	7.49	7.95	8.04	6.09	14.97	23.86	32.14	16.46	19.73	21.26	23.18	6.09	14.97	23.86	32.14
Revenue from *Azolla* spp./tank	3.63	3.41	2.47	2.14	4.35	4.30	3.80	3.57	7.01	6.41	5.82	5.03	7.13	6.90	6.56	6.02
Gross receipts/tank	9.72	10.90	10.42	10.17	10.43	19.27	27.67	35.71	23.46	26.14	27.07	28.22	13.21	21.87	30.43	38.16
Variable costs																
Cost of feeds per tank	0.11	0.12	0.15	0.17	0.13	0.15	0.18	0.21	0.16	0.18	0.22	0.25	0.19	0.23	0.27	0.31
Field labour	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06
Cost of equipment	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45
Electricity	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Miscellaneous	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Sub-total variable costs	0.91	0.92	0.95	0.97	0.93	0.95	0.98	1.01	0.96	0.98	1.02	1.05	0.99	1.03	1.07	1.11
Interest on operating cost	0.15	0.15	0.15	0.16	0.15	0.15	0.16	0.16	0.15	0.16	0.16	0.17	0.16	0.16	0.17	0.18
Total variable cost (TVC)	1.05	1.07	1.10	1.12	1.08	1.10	1.13	1.17	1.11	1.14	1.18	1.22	1.15	1.19	1.24	1.29
Fixed costs																
Fixed costs	2.24	2.24	2.24	2.24	2.24	2.24	2.24	2.24	2.24	2.24	2.24	2.24	2.24	2.24	2.24	2.24
Amortization	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Interest on fixed cost	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18
Total fixed cost	3.42	3.42	3.42	3.42	3.42	3.42	3.42	3.42	3.42	3.42	3.42	3.42	3.42	3.42	3.42	3.42
Total cost (TC)	4.47	4.49	4.52	4.54	4.50	4.52	4.55	4.59	4.53	4.56	4.60	4.64	4.57	4.61	4.66	4.71
Net returns above TVC	8.66	9.82	9.33	9.05	9.36	18.17	26.53	34.55	22.35	25.00	25.89	26.99	12.06	20.68	29.19	36.87
Net returns above TC	5.24	6.40	5.91	5.63	5.94	14.75	23.11	31.13	18.93	21.58	22.47	23.57	8.64	17.26	25.77	33.45
Break even price	0.06	0.05	0.04	0.04	0.06	0.02	0.01	0.01	0.02	0.02	0.02	0.01	0.06	0.02	0.01	0.01

Assumptions:
Budget assumptions: The interest rates on fixed cost = 18%.
Production assumption: Fingerlings will be marketed after 30 days of production.
Marketing assumption: There is demand for the fish and plants throughout the year.
Land assumption: Own the land where the aquaponic is practiced and that the space for aquaponics attracts no other economic use.

Table 7
Net present value (NPV) and internal rate of return (IRR) for the model aquaponics at different *Clarias gariepinus* larvae stocking densities and water flow rates for 30 days period.

Stocking density (Larvae/m^3)	Water flow rate (L/min)	NPV (18%)	IRR
1500	0.25	−12,061.54	16.2
	0.50	−5545.21	16.7
	0.75	−7610.62	17.2
	1.00	−9697.95	17.8
3000	0.25	−8026.15	16.7
	0.50	31,586.51	21.9
	0.75	63,570.00	25.8
	1.00	77,821.09	36.2
4500	0.25	4701.10	19.2
	0.50	75,887.43	29.8
	0.75	80,201.75	34.2
	1.00	87,050.74	39.6
6000	0.25	6534.69	19.5
	0.50	53,021.52	24.6.7
	0.75	98,915.12	40.4
	1.00	118,131.84	47.8

from the fish wastes as well as feeds. The nitrogenous content may be increased at increasing fish stocking density and water flow rate. Meanwhile Ca, Mg, Fe and Na showed decreasing concentrations at higher fish stocking densities. The hydroponically grown *Azolla* spp. supplied with the nutrient present in aquaculture water produced apparently normal, healthy looking plants, indicating that there were no major mineral deficiencies.

4.4. An economic analysis of catfish-Azolla in response to stocking density and water flow rates

In the current study the yields of both fish and plants were affected by stocking density and water flow rate and thus these factors were significant in affecting the economic benefits from aquaponics. Our estimated profitability is lower than those found in previous studies on large-scale aquaponic operations. This could be because our estimates are based on experimental operations rather than commercial cases. Commercial operations face many challenges in their day-to-day operations such as weather conditions and supply chain logistics issues. In locations where fresh produce are available at lower price, aquaponics farms may face more severe market competition. At the same time, their construction cost may be lower in the current study than in other developed countries where majority of the studies dwell. While we only studied small-scale operations, the results suggest potential for aquaponics to play a larger role in supplying both vegetables and larval fish in the local market.

Enterprise budgets for aquaponic farms with different stocking densities and water flow rates for fish and *Azolla* production indicated all the treatments posted positive returns to risk and were viable investments. The break-even prices for variable costs were able to cover the cost of larval fish in the local market as they were below the sale price of US$ 0.10 per case that the farm was able to receive in the market. Cash flow projections were made over a 10-year period for each of the units with the net present value (NPV) and internal rate of return (IRR) were calculated for each of the model farms. It was observed that sales of fish and *Azolla* sp. at stocking density of 1500 larvae/m^3 was largely not profitable regardless of the water flow rate. However, all the other stocking densities were profitable and increased with increased water flow rates and stocking density. Investors must determine their requirements for acceptable returns when choosing the stocking density to use and should factor the water flow rate.

5. Conclusions

In the model aquaponics system, the stocking density and water flow rate had an interactive influence ($P < 0.05$) on the concentration of DO, TAN, and NO_3^-. The stocking density and water flow rate had an interactive influence ($P < 0.05$) on the final mean weight, SGR, and survival of *C. gariepinus* larvae, but not for FCR and yield ($P > 0.05$). The highest fish weight was achieved under combination of stocking density 3245 larvae/m^3 and water flow rate of 0.87 L/min, but growth will still be optimized upto a stocking density of 4850 larvae/m^3 but at higher combinations of water flow rates (> 0.875 L/min). The stocking density and water flow rate had an interactive influence ($P < 0.05$) on chlorophyll *a* concentration (Fig. 3b), but not fresh weight, dry weight and yield ($P > 0.05$). Enterprise budgets for aquaponic with different stocking densities and water flow rate for fish and *Azolla* production allowed all the treatments to post positive returns to risk and were viable investments. The break-even prices for variable costs were able to cover the cost of fish in the local market as they were below the sale price of $ 0.10 per case that the farm was able to receive in the market. Cash flow projections were made over a 10-year period for each of the units with the net present value (NPV) and internal rate of return (IRR) suggest that sales of fish and *Azolla* sp. were profitable above stocking density of 3000 larvae/m^3 and water flow rates of 0.25 L/min and increased with increased water flow rate and stocking density.

Although at all stocking density the revenue were able to cover the variable costs, this study recommends stocking at 4500 larvae m^{-3} and water flow rates of 0.75 and 1.0 L/min since at higher density than that the growth of fish reduces. So far, only a few scientific studies have dealt with aquaponic cultivations, and none of them are directly comparable, owing to the fact that different systems, sizes and locations were used. Basic information on the most important factors influencing aquaponics is missing, preventing the transfer of results to other locations, systems and organisms. More detailed studies on fish and plant production within comparable design of aquaponic systems are therefore recommended.

CRediT authorship contribution statement

James Mugo-Bundi: Conceptualization, Methodology, Formal analysis. **Julius O. Manyala:** Data curation, Supervision, Writing – review & editing. **Mucai Muchiri:** Funding acquisition, Project administration, Supervision. **Geraldine Matolla:** Formal analysis, Writing – review & editing.

Declaration of Competing Interest

The authors declare no conflict of interest in the publication of this manuscript.

Data availability

Not applicable to this manuscript.

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Appendix A. Supplementary data

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References

Abbey, M., Anderson, N.O., Yue, C., Short, G., Schermann, M.A., Phelps, N., Venturelli, P., Vickers, Z.M., 2019. An analysis of strawberry (*Fragaria*×*ananassa*) productivity in northern latitudinal aquaponic growing conditions. J. Am. Pomol. Soc. 73 (1), 22–37.

Al Tawaha, A.R., Wahab, P.E.M., Jaafar, H.B., Zuan, A.T.K., Hassan, M.Z., 2021. Effects of fish stocking density on water quality, growth performance of tilapia and yield of butterhead Lettuce grown in decoupled recirculation aquaponic systems. J. Ecol. Eng. 22 (1).

Alarcón-Silvas, S., León-Cañedo, J., Fierro-Sañudo, J., Ramírez-Rochín, J., Fregoso-López, M., Frías-Espericueta, M., Osuna-Martínez, C., Páez-Osuna, F., 2021. Water quality, water usage, nutrient use efficiency and growth of shrimp *Litopenaeus vannamei* in an integrated aquaponic system with basil *Ocimum basilicum.* Aquaculture 543, 737023.

Al-Tawaha, A., Puteri Edaroyati, M., Siti Aishah, H., 2017. Requirements for Inserting Intercropping in Aquaponics System for Sustainability in Agricultural Production System.

Åmand, L., Olsson, G., Carlsson, B., 2013. Aeration control–a review. Water Sci. Technol. 67 (11), 2374–2398.

Anando, D.A., Andriani, Y., Hamdani, H., Zahidah, Z., 2022. Effect of oxygen supply on productivity in aquaponics system. World Sci. News 171, 82–96.

Ani, J.S., Manyala, J.O., Masese, F.O., Fitzsimmons, K., 2022. Effect of stocking density on growth performance of monosex Nile Tilapia (*Oreochromis niloticus*) in the aquaponic system integrated with lettuce (*Lactuca sativa*). Aquac. Fish. 7 (3), 328–335.

AOAC, 2005. Official Methods of Analysis, 18th ed. Association of Official AnalyticalChemists, Washington, DC.

Baganz, G.F., Junge, R., Portella, M.C., Goddek, S., Keesman, K.J., Baganz, D., Staaks, G., Shaw, C., Lohrberg, F., Kloas, W., 2022. The aquaponic principle—it is all about coupling. Rev. Aquac. 14 (1), 252–264.

Barbosa, P.T.L., Povh, J.A., Farias, K.N.N., da Silva, T.V., Teodoro, G.C., Ribeiro, J.S., Stringhetta, G.R., dos Santos Fernandes, C.E., Corrêa Filho, R.A.C., 2022. Nile tilapia production in polyculture with freshwater shrimp using an aquaponic system and biofloc technology. Aquaculture 551, 737916.

Baßmann, B., Brenner, M., Palm, H.W., 2017. Stress and welfare of African catfish (*Clarias gariepinus* Burchell, 1822) in a coupled aquaponic system. Water 9 (7), 504.

Baßmann, B., Harbach, H., Weißbach, S., Palm, H.W., 2020. Effect of plant density in coupled aquaponics on the welfare status of African catfish, *Clarias gariepinus*. J. World Aquacult. Soc. 51 (1), 183–199.

Boyd, C.E., Torrans, E.L., Tucker, C.S., 2018. Dissolved oxygen and aeration in ictalurid catfish aquaculture. J. World Aquacult. Soc. 49 (1), 7–70.

Brouwer, P., Nierop, K.G., Huijgen, W.J., Schluepmann, H., 2019. Aquatic weeds as novel protein sources: alkaline extraction of tannin-rich *Azolla*. Biotechnol. Rep. 24, e00368.

Cassol, D., De Silva, F., Falqueto, A., Bacarin, M., 2008. An evaluation of non-destructive methods to estimate total chlorophyll content. Photosynthetica 46 (4), 634–636.

Cohen, A., Malone, S., Morris, Z., Weissburg, M., Bras, B., 2018. Combined fish and lettuce cultivation: an aquaponics life cycle assessment. Proc. CIRP 69, 551–556.

Colt, J., Schuur, A.M., Weaver, D., Semmens, K., 2022. Engineering design of aquaponics systems. Rev. Fish. Sci. Aquac. 30 (1), 33–80.

Curry, C., Krosch, C., Riera Vila, I., 2017. Solving iron Deficiency with Chelated iron Applications in Strawberry-Koi Aquaponics Systems.

Datta, S., Mahapatra, B., Bhakta, J., Bag, S., Lahiri, S., Mandal, R., Jana, B., 2018. Aquaponics: A green and sustainable eco-tech for environmental cum economic benefits through integration of fish and edible crop cultivation. In: Wastewater Management Through Aquaculture. Springer, pp. 207–224.

Delaide, B., Delhaye, G., Dermience, M., Gott, J., Soyeurt, H., Jijakli, M.H., 2017. Plant and fish production performance, nutrient mass balances, energy and water use of the PAFF Box, a small-scale aquaponic system. Aquac. Eng. 78, 130–139.

Diatin, I., Shafruddin, D., Hude, N., Sholihah, M.a., Mutsmir, I., 2021. Production performance and financial feasibility analysis of farming catfish (*Clarias gariepinus*) utilizing water exchange system, aquaponic, and biofloc technology. J. Saudi Soc. Agric. Sci. 20 (5), 344–351.

Diem, T.N.T., Konnerup, D., Brix, H., 2017. Effects of recirculation rates on water quality and *Oreochromis niloticus* growth in aquaponic systems. Aquac. Eng. 78, 95–104.

Endut, A., Jusoh, A., Ali, N., Wan Nik, W., Hassan, A., 2009. Effect of flow rate on water quality parameters and plant growth of water spinach (*Ipomoea aquatica*) in an aquaponic recirculating system. Desalin. Water Treat. 5 (1–3), 19–28.

Endut, A., Jusoh, A., Ali, N., Nik, W.W., Hassan, A., 2010. A study on the optimal hydraulic loading rate and plant ratios in recirculation aquaponic system. Bioresour. Technol. 101 (5), 1511–1517.

Engle, C.R., 2016. Economics of Aquaponics. Oklahoma Cooperative Extension Service.

Espinosa Moya, E.A., Angel Sahagún, C.A., Mendoza Carrillo, J.M., Albertos Alpuche, P. J., Álvarez-González, C.A., Martínez-Yáñez, R., 2016. Herbaceous plants as part of biological filter for aquaponics system. Aquac. Res. 47 (6), 1716–1726.

Estim, A., Saufie, S., Mustafa, S., 2019. Water quality remediation using aquaponics sub-systems as biological and mechanical filters in aquaculture. J. Water Process Eng. 30, 100566.

Estrada-Perez, N., Hernandez-Llamas, A., MJ Ruiz-Velazco, J., Zavala-Leal, I., Romero-Bañuelos, C.A., Cruz-Crespo, E., Juárez-Rossete, C., Domínguez-Ojeda, D., Campos-Mendoza, A., 2018. Stochastic modelling of aquaponic production of tilapia (*Oreochromis niloticus*) with lettuce (*Lactuca sativa*) and cucumber (*Cucumis sativus*). Aquac. Res. 49 (12), 3723–3734.

Federation Water Environmental, Association, A, 2005. Standard Methods for the Examination of Water and Wastewater. American Public Health Association (APHA), Washington, DC, USA, 21.

Ferrarezi, R.S., Bailey, D.S., 2019. Basil performance evaluation in aquaponics. HortTechnology 29 (1), 85–93.

Fischer, H., Romano, N., Jones, J., Howe, J., Renukdas, N., Sinha, A.K., 2021. Comparing water quality/bacterial composition and productivity of largemouth bass *Micropterus salmoides* juveniles in a recirculating aquaculture system versus aquaponics as well as plant growth/mineral composition with or without media. Aquaculture 538, 736554.

Franklin, D.A., Edward, L., 2019. Ammonia toxicity and adaptive response in marine fishes. Indian J. Geo-Mar. Sci. 48 (3), 273–279.

Gotelli, N.J., Ellison, A.M., 2004. A primer of ecological statistics, Vol. 1. Sinauer Associates, Sunderland, pp. 1–640.

Hasan, Z., Dhahiyat, Y., Andriani, Y., Zidni, I., 2017. Water quality improvement of Nile tilapia and catfish polyculture in aquaponics system. Nusantara Biosci. 9 (1), 83–85.

Horwitz, W., Chichilo, P., Reynolds, H., 1970. Official methods of analysis of the Association of Official Analytical Chemists. Official Methods of Analysis of the Association of Official Analytical Chemists.

Hundley, G.C., Navarro, F.K.S.P., Ribeiro Filho, O.P., Navarro, R.D., 2018. Integration of Nile tilapia (*Oreochromis niloticus* L.) production *Origanum majorana* L. and *Ocimum basilicum* L. using aquaponics technology. Acta Scientiarum. Technology 40, e35460.

Hussain, T., Verma, A., Tiwari, V., Prakash, C., Rathore, G., Shete, A., Nuwansi, K., 2014. Optimizing koi carp, *Cyprinus carpio* var. koi (Linnaeus, 1758), stocking density and nutrient recycling with spinach in an aquaponic system. J. World Aquacult. Soc. 45 (6), 652–661.

Hussain, T., Verma, A., Tiwari, V., Prakash, C., Rathore, G., Shete, A., Saharan, N., 2015. Effect of water flow rates on growth of *Cyprinus carpio* var. koi (*Cyprinus carpio* L., 1758) and spinach plant in aquaponic system. Aquac. Int. 23 (1), 369–384.

Kemigabo, C., Jere, L.W., Sikawa, D., Masembe, C., Kang'ombe, J., Abdel-Tawwab, M., 2019. Growth response of African catfish, *Clarias gariepinus* (B.), larvae and fingerlings fed protease-incorporated diets. J. Appl. Ichthyol. 35 (2), 480–487.

Knaus, U., Appelbaum, S., Palm, H.W., 2018. Significant factors affecting the economic sustainability of closed backyard aquaponics systems. Part IV: autumn herbs and polyponics. Aquac. Aquar. Conserv. Legis. 11 (6), 1760–1775.

König, B., Janker, J., Reinhardt, T., Villarroel, M., Junge, R., 2018. Analysis of aquaponics as an emerging technological innovation system. J. Clean. Prod. 180, 232–243.

Kösesakal, T., Yıldız, M., 2019. Growth performance and biochemical profile of *Azolla pinnata* and *Azolla caroliniana* grown under greenhouse conditions. Arch. Biol. Sci. 71 (3), 475–482.

Kralik, B., Weisstein, F., Meyer, J., Neves, K., Anderson, D., Kershaw, J., 2022. From water to table: A multidisciplinary approach comparing fish from aquaponics with traditional production methods. Aquaculture 552, 737953.

Lennard, W.A., Leonard, B.V., 2004. A comparison of reciprocating flow versus constant flow in an integrated, gravel bed, aquaponic test system. Aquac. Int. 12 (6), 539–553.

Lobillo-Eguíbar, J., Fernández-Cabanás, V.M., Bermejo, L.A., Pérez-Urrestarazu, L., 2020. Economic sustainability of small-scale aquaponic systems for food self-production. Agronomy 10 (10), 1468.

Lumsangkul, C., Linh, N.V., Chaiwan, F., Abdel-Tawwab, M., Dawood, M.A., Faggio, C., Jaturasitha, S., Van Doan, H., 2022. Dietary treatment of Nile tilapia (*Oreochromis niloticus*) with aquatic fern (*Azolla caroliniana*) improves growth performance, immunological response, and disease resistance against *Streptococcus agalactiae* cultured in bio-floc system. Aquac. Rep. 24, 101114.

Mallya, Y.J., 2007. The Effects of Dissolved Oxygen on Fish Growth in Aquaculture. The United Nations University Fisheries Training Programme. Final Project.

Mandal, R., Datta, A., Sarangi, N., Mukhopadhyay, P., 2010. Diversity of aquatic macrophytes as food and feed components to herbivorous fish- a review. Indian J. Fish. 57 (3), 65–73.

Marcó, A., Rubio, R., Compañó, R., Casals, I., 2002. Comparison of the Kjeldahl method and a combustion method for total nitrogen determination in animal feed. Talanta 57 (5), 1019–1026.

Maucieri, C., Nicoletto, C., Junge, R., Schmautz, Z., Sambo, P., Borin, M., 2018. Hydroponic systems and water management in aquaponics: a review. Ital. J. Agron. 13 (1/1012).

Maucieri, C., Nicoletto, C., Zanin, G., Birolo, M., Trocino, A., Sambo, P., Borin, M., Xiccato, G., 2019. Effect of stocking density of fish on water quality and growth performance of European carp and leafy vegetables in a low-tech aquaponic system. PLoS One 14 (5), e0217561.

Momoh, P., Solomon, R., 2017. Physiochemical parameters of catfish (*Clarias gariepinus*) fed a combination of rice bran and Irish potatoes. Direct Res. J. Vet. Med. Anim. Sci. 2 (2), 36–50.

Nakphet, S., Ritchie, R.J., Kiriratnikom, S., 2017. Aquatic plants for bioremediation in red hybrid tilapia (*Oreochromis niloticus*×*Oreochromis mossambicus*) recirculating aquaculture. Aquac. Int. 25 (2), 619–633.

Narita, D., Sato, I., Ogawada, D., Matsumura, A., 2020. Integrating economic measures of adaptation effectiveness into climate change interventions: A case study of irrigation development in Mwea, Kenya. PLoS One 15 (12), e0243779.

Nasir, N.A.N.M., Kamaruddin, S.A., Zakarya, I.A., Islam, A.K.M.A., 2022. Sustainable alternative animal feeds: recent advances and future perspective of using azolla as animal feed in livestock, poultry and fish nutrition. Sustain. Chem. Pharm. 25, 100581.

Nuwansi, K., Verma, A., Prakash, C., Tiwari, V., Chandrakant, M., Shete, A., Prabhath, G., 2016. Effect of water flow rate on polyculture of koi carp (*Cyprinus carpio* var. koi) and goldfish (*Carassius auratus*) with water spinach (*Ipomoea aquatica*) in recirculating aquaponic system. Aquac. Int. 24 (1), 385–393.

Nwipie, G., Erondu, E., Zabbey, N., 2015. Influence of stocking density on growth and survival of post fry of the african mud catfish, *Clarias gariepinus*. Fish. Aquac. J. 6, 116.

Okomoda, V.T., Koh, I.C.C., Shahreza, S.M., 2018. A simple technique for accurate estimation of fertilization rate with specific application to *Clarias gariepinus* (Burchell, 1822). Aquac. Res. 49 (2), 1116–1121.

Oladimeji, A., Olufeagba, S., Ayuba, V., Sololmon, S., Okomoda, V., 2018. Effects of Different Growth Media on Water Quality and Plant Yield in a Catfish-Pumpkin Aquaponics System. Journal of King Saud University-Science.

Oladimeji, S.A., Okomoda, V.T., Olufeagba, S.O., Solomon, S.G., Abol-Munafi, A.B., Alabi, K.I., Ikhwanuddin, M., Martins, C.O., Umaru, J., Hassan, A., 2020. Aquaponics production of catfish and pumpkin: comparison with conventional production systems. Food Sci. Nutr. 8 (5), 2307–2315.

Olurin, K., Oluwo, A., 2010. Growth and survival of African catfish (*Clarias gariepinus*) larvae fed decapsulated Artemia, live Daphnia, or commercial starter diet. Isr. J. Aquac. Bamidgeh 62 (1), 50–55.

Onura, C.N.a., Van den Broeck, W., Nevejan, N., Muendo, P., Van Stappen, G., 2018. Growth performance and intestinal morphology of African catfish (*Clarias gariepinus*, Burchell, 1822) larvae fed on live and dry feeds. Aquaculture 489, 70–79.

Oyange, W., Chemining'wa, G., Kanya, J., Njiruh, P., 2019. Azolla fern in Mwea irrigation scheme and its potential nitrogen contribution in paddy rice production. J. Agric. Sci. (Toronto) 11 (18), 30–44.

Palm, H.W., Knaus, U., Appelbaum, S., Goddek, S., Strauch, S.M., Vermeulen, T., Jijakli, M.H., Kotzen, B., 2018. Towards commercial aquaponics: a review of systems, designs, scales and nomenclature. Aquac. Int. 26 (3), 813–842.

Pérez-Urrestarazu, L., Lobillo-Eguíbar, J., Fernández-Cañero, R., Fernández-Cabanás, V.M., 2019. Suitability and optimization of FAO's small-scale aquaponics systems for joint production of lettuce (*Lactuca sativa*) and fish (*Carassius auratus*). Aquac. Eng. 85, 129–137.

Pinho, S.M., Flores, R.M.V., David, L.H., Emerenciano, M.G., Quagrainie, K.K., Portella, M.C., 2022. Economic comparison between conventional aquaponics and FLOCponics systems. Aquaculture 552, 737987.

Quagrainie, K.K., Flores, R.M.V., Kim, H.-J., McClain, V., 2018. Economic analysis of aquaponics and hydroponics production in the US Midwest. J. Appl. Aquac. 30 (1), 1–14.

Quinn, G.P., Keough, M.J., 2002. *Experimental design and data analysis for biologists*. Cambridge University Press.

Rahmah, S., Nasrah, U., Lim, L.-S., Ishak, S.D., Rozaini, M.Z.H., Liew, H.J., 2022. Aquaculture wastewater-raised *Azolla* as partial alternative dietary protein for Pangasius catfish. Environ. Res. 208, 112718.

Rakocy, J., Masser, M.P., Losordo, T., 2016. Recirculating Aquaculture Tank Production Systems: Aquaponics-Integrating Fish and Plant Culture.

Rayhan, M.Z., Rahman, M.A., Hossain, M.A., Akter, T., Akter, T., 2018. Effect of stocking density on growth performance of monosex tilapia (*Oreochromis niloticus*) with Indian spinach (*Basella alba*) in a recirculating aquaponic system. Int. J. Environ. Agric. Biotechnol. 3 (2), 239073.

Rizal, A., Dhahiyat, Y., Andriani, Y., Handaka, A., Sahidin, A., 2018. The Economic and Social Benefits of an Aquaponic System for the Integrated Production of Fish and Water Plants, IOP Conference Series: Earth and Environmental Science. IOP publishing, p. 012098.

Robles-Porchas, G.R., Gollas-Galván, T., Martínez-Porchas, M., Martínez-Cordova, L.R., Miranda-Baeza, A., Vargas-Albores, F., 2020. The nitrification process for nitrogen removal in biofloc system aquaculture. Rev. Aquac. 12 (4), 2228–2249.

Ross, S.A., Westerfield, R., Jordan, B.D., Biktimirov, E.N., 2008. Essentials of Corporate Finance. McGraw-Hill/Irwin.

Sales, J., 2009. Accuracy of the use of energy conversion factors for compound fish diets. Arch. Anim. Nutr. 63 (6), 491–509.

Schmautz, Z., Loeu, F., Liebisch, F., Graber, A., Mathis, A., Griessler Bulc, T., Junge, R., 2016. Tomato productivity and quality in aquaponics: comparison of three hydroponic methods. Water 8 (11), 533.

Schram, E., Verdegem, M., Widjaja, R., Kloet, C., Foss, A., Schelvis-Smit, R., Roth, B., Imsland, A., 2009. Impact of increased flow rate on specific growth rate of juvenile turbot (*Scophthalmus maximus*, Rafinesque 1810). Aquaculture 292 (1–2), 46–52.

Shete, A., Verma, A., Tandel, R., Prakash, C., Tiwari, V., Hussain, T., 2013. Optimization of water circulation period for the culture of goldfish with spinach in aquaponic system. J. Agric. Sci. 5 (4), 26.

Shete, A., Verma, A., Chadha, N., Prakash, C., Peter, R., Ahmad, I., Nuwansi, K., 2016. Optimization of hydraulic loading rate in aquaponic system with common carp (*Cyprinus carpio*) and Mint (*Mentha arvensis*). Aquac. Eng. 72, 53–57.

Somerville, C., Cohen, M., Pantanella, E., Stankus, A., Lovatelli, A., 2014. Small-scale aquaponic food production: integrated fish and plant farming. In: FAO Fisheries and Aquaculture Technical Paper (589), I.

Stathopoulou, P., Berillis, P., Levizou, E., Sakellariou-Makrantonaki, M., Kormas, A., Angelaki, A., Kapsis, P., Vlahos, N., Mente, E., 2018. Basil and Nile tilapia production in a small scale aquaponic system. Journal of FisheriesSciences.com 12 (4), 1–3.

Stone, N.M., Thomforde, H.K., 2004. Understanding Your Fish Pond Water Analysis Report. Cooperative Extension Program, University of Arkansas at Pine Bluff, US ….

Suhl, J., Dannehl, D., Kloas, W., Baganz, D., Jobs, S., Scheibe, G., Schmidt, U., 2016. Advanced aquaponics: evaluation of intensive tomato production in aquaponics vs. conventional hydroponics. Agric. Water Manag. 178, 335–344.

Thiex, N., 2009. Evaluation of analytical methods for the determination of moisture, crude protein, crude fat, and crude fiber in distillers dried grains with solubles. J. AOAC Int. 92 (1), 61–73.

Tokunaga, K., Tamaru, C., Ako, H., Leung, P., 2015. Economics of small-scale commercial aquaponics in Hawai'i. J. World Aquacult. Soc. 46 (1), 20–32.

Uggetti, E., Hughes-Riley, T., Morris, R.H., Newton, M.I., Trabi, C.L., Hawes, P., Puigagut, J., García, J., 2016. Intermittent aeration to improve wastewater treatment efficiency in pilot-scale constructed wetland. Sci. Total Environ. 559, 212–217.

Verreth, J., Den Bieman, H., 1987. Quantitative feed requirements of African catfish (Clarias gariepinus Burchell) larvae fed with decapsulated cysts of Artemia: I. The effect of temperature and feeding level. Aquaculture 63 (1–4), 251–267.

Wallace-Springer, N., Wells, D.E., Pickens, J.M., Ayipio, E., Kemble, J., 2022. Effects of hydraulic retention time of aquaculture effluent on nutrient film technique lettuce productivity. Agronomy 12 (10), 2570.

Watts, C., Bright, L.A., Coyle, S., Tidwell, J., 2016. Evaluation of stocking density during second-year growth of largemouth bass, Micropterus salmoides, raised indoors in a recirculating aquaculture system. J. World Aquacult. Soc. 47 (4), 538–543.

Wu, H., Zou, Y., Lv, J., Hu, Z., 2018. Impacts of aeration management and polylactic acid addition on dissolved organic matter characteristics in intensified aquaponic systems. Chemosphere 205, 579–586.

Yi, Y., Lin, C.K., 2001. Effects of biomass of caged Nile tilapia (*Oreochromis niloticus*) and aeration on the growth and yields in an integrated cage-cum-pond system. Aquaculture 195 (3–4), 253–267.

Yossa, R., Verdegem, M., 2015. Misuse of multiple comparison tests and underuse of contrast procedures in aquaculture publications. Aquaculture 437, 344–350.

Zappernick, N., Nedunuri, K., Islam, K., Khanal, S., Worley, T., Laki, S., Shah, A., 2022. Techno-economic analysis of a recirculating tilapia-lettuce aquaponics system. J. Clean. Prod. 365, 132753.