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Sustainable Production of Fish *Oreochromis mossambicus* and Green Gram *Vigna radiata* in Recirculating Aquaponics System

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Abstract: Aquaponics fits into the integrated agri-aquaculture systems (IAAS). However, IAAS applies many different aquatic animal and plant production technologies in many contexts, whereas aquaponics is far more tightly associated with integrating tank-based fish culture technologies (e.g. recirculating aquaculture systems- RAS) with aquatic or hydroponic plant culture technologies. The green gram was cultured to test their yield efficiency in three different hydroponic subsystems viz., Media Bed System (MBS), Floating Raft System (FRS) and Nutrient Film Technique (NFT) whereas the freshwater fish *Oreochromis mossambicus* was cultured in Aquaponic system. The nutrients were supplied from fish culture tank while plant stripped nutrients from the wastewater before it was return to the fish. Fish *O. mossambicus* biomass was gains well and green gram *Vigna radiata* yield were good. The biomass gain and yield of both the crops was noticed in the following order: MBS>FRS>NFT with the significant difference was observed between all treatments. The NFT treatment was significantly lesser than the other two treatments in the case of (nitrate removal) whereas in other test treatments temperature, pH, and dissolved oxygen was shown no significant differences. The overall results conclude that the MBS aquaponics is more suitable in yielding fish biomass and plant yield when compared to FRS and NFT aquaponics system.

Keywords: Aquaponics, *Oreochromis mossambicus*, *Vigna radiata*, MBS, FRS, NFT

I. INTRODUCTION

Aquaponics is currently making waves as an alternative system for organic farming. Aquaponics is cooperation between plants and fish and the term originates from the two words aquaculture (The growing of fish in a closed environment) and hydroponics (the growing of plants usually in a soil-less environment). In the aquaponics the nutrient-rich water from raising fish provides a natural fertilizer for the plants and the plants purify the water for the fish. Aquaponics uses 90% less water than conventional agriculture. Being a closed system it does not waste water due to use of harmful fertilizer. The basic idea behind aquaponics waste serves as fertilizer for plants. Fish eat food which you provide and produce waste. The water pump carries water with fish feed waste. In the hydroponic pebbles they convert that waste (ammonia+nitrites) to fertilizer (nitrates). The plant feed on nitrates and purify the water. The clean water will reach back to the fishes.

The growing food insecurity, uncontrollable rising of food prices, water scarcity and poverty, especially in developing countries, coupled with concerns of climate pattern, have resulted significant global challenge. To overcome these issues, agriculture-based sustainable farming was suggested to produce food by conserving water and recycling nutrients, and converting wastes water into high-value resources. The global population has been growing rapidly, with a matching increase in global food demand, especially for high-quality protein, which has been driving the development of several agribusiness sectors such as aquaculture. However, large volumes of water are required to produce these animal proteins. The industrial scale traditional practices of aquaculture and agriculture and recent population growth have caused a serious water crisis. In order to avoid further crises related to the use of natural resources, new approaches and technologies are needed in agriculture aiming to achieve greater productivity with minimal environmental impact, when compared to conventional systems. In this context, aquaponics is emerging as an alternative method of food production. Aquaponics is an innovative smart and sustainable production system for integrating aquaculture (fish culture) with hydroponic (vegetable crops) that can play a crucial role in the future of environmental and socio-economic sustainability in rural and urban areas of India.

Green gram or mung bean (*Vigna radiata* L.) is one of the most important food legumes in India. It is the third most important pulse crop of India. Green gram is a rich source of protein (24%) and also contributes carbohydrates (60%), fat (1.5%), amino acids, vitamins and minerals etc. Area under green gram in India is 3.80 million hectares with an annual production of 1.1 million tonnes. In Tamil Nadu, the area under green gram is 0.13 million hectares with an annual production of 458.8 tonnes.

The average productivity of green gram over globe is $577 \text{ kg}\cdot\text{ha}^{-1}$ and in India it is $426 \text{ kg}\cdot\text{ha}^{-1}$, which is considered to be low. The low productivity of green gram is due to the cultivation of this crop in marginal and sub-marginal lands with poor management practices.

Tilapia is an inexpensive, mild-flavored fish. It is the fourth most commonly consumed type of fish food. Many people love tilapia because it is relatively affordable. It is an ideal fish for farming because it doesn't mind being crowded, grows quickly and consumes a cheap vegetarian diet. These qualities translate to a relatively inexpensive product compared to other types of fish food. Tilapia is a pretty impressive source of protein. Even more impressive is the amount of vitamins and minerals in this fish. Tilapia is rich in niacin, vitamin B₁₂, phosphorus, selenium and potassium. In this line, the present study is made attempt on the culture of commercially important fish Tilapia (*Oreochromis mossambicus*) and green gram *Vigna radiata* combined in aquaponics system.

II. MATERIALS AND METHODS

A. Preparation of Plant Seed

The Green gram (*Vigna radiata*) plant were selected for the experiment. The green gram seeds were soaked for one day and then producing sprouts of 2cm length and they were planted in the hydroponic system.

B. Fish Origin and Holding

Oreochromis mossambicus were obtained from the Private Hatchery located at Kerala. The fishes were stocked in the designated tanks at the Zoology Lab., Nirmala College, Coimbatore. The stocking size of the fish was ranged from 50 to 65 grams. The fishes were kept in a cement tank for 15 days for acclimatization. The fishes were fed with pellet feed during acclimatization.

C. Experimental Design of Aquaponic System

The experimental aquaponic system consist of 15 fishes which were introduced into the system. The each aquaponic system consist of fish holding aquaponic tank, associated bio filter and aquaponic sub system. The glass aquarium fish tank with capacity of 20 liter was used presently to rear the fish. The fish tank contains airlift pipe (to the biofilter) and submersible water pump (to the hydroponic bed). The fish tank was covered with nylon net to control the evaporation and prevent fish jumping from the tank. Each tank contains 3 in one biofilter. The submersible water pump sat inside the fish tank sinking into the water. The waste water from fish rearing tank was supplied to the hydroponic bed where green gram was growing and water from plant bed was return to fish rearing tank. Each tank and bio filter unit had associated hydroponic plant growth component which was configured depending on the test experiment. The hydroponic component was rectangular in shape for media based system and for NFT (Nutrient film technique) system the PVC pipe lines were connected and placed above the aquaponic system. In the RAFT system the Thermocol were placed over the fish tank with the holes attached with the PVC holders. The submersible water pump in the fish tank continuously delivered the water to the hydroponic component. The water from hydroponic component returned to the fish tank with 20 mm drain pipe. The tap water was used to rear the fish and culture the agriculture plant.

D. Experimental Methodology

The experiment was designed to compare the three hydroponic growing sub-systems within the aquaponic unit(s). These three hydroponic subsystem consist of gravel bed culture (the hydroponic component was filled with river gravel with a rate of the return water flow set to produce a water level in the hydroponic component that completely saturate the gravel media) floating or raft culture contain raft made by the thermocol sheet with 2 cm width with 6 holes attached with PVC holder for plant support with floated on top of the water and NFT culture made up of 3 feet PVC pipes with holes on the top supported with PVC holders.

Four separate experiments, each with 3 replicates were tested as follows: 1. Control: Soil with green gram plant. 2. Media Bed: Fish in tank, plants and gravel in the hydroponic component. 3. Floating: Fish in tank, plants in the floating "raft" hydroponic component. 4. NFT: Fish in tank, plants in the NFT hydroponic component. At the initiation of the experiment, systems were flushed and refilled with fresh water. Fishes were stocked at each system for the experiment (the initial fish biomass were recorded).

E. Sample Analysis

The amount of fish feed (g), air and water temperature (°C), water replaced per tank (to adjust the evapotranspiration), sodium bicarbonate were added (to adjust the pH between 6.8 to 7.00) added directly to the fish rearing compartment, conductivity and dissolved oxygen (mg/l) were recorded. Fish rearing tanks water were sampled for ammonia (mg/l), nitrite (mg/l) and nitrate (mg/l). All samples were taken from the fish rearing compartment of the aquaponics units.

The amount of water replaced per tank to be determined by re-filling the tank to a pre-measured 20 l mark and recording the amount of water added. The temperature, pH and electrical conductivity were determined using multi parameter analysis Kit (Hanna multi parameter). Dissolved oxygen, hardness, alkalinity, ammonia, nitrite, and nitrate were analyzed by standard methods [1]. The leaf length and length of the whole plant were measured by using long scale and thread.

F. Statistical Analysis

The data were subjected to analyses using statistical package SPSS version 16 in which one-way ANOVA and Duncan's Multiple Range Test had been performed at a significance level of ($P < 0.05$) at 95% confidence limit to know the significant difference between the treatments and control means for different variables.

III. RESULTS AND DISCUSSION

A. Water Quality Variables And Nutrient Dynamics

The water temperature in the fish rearing tanks during the study was varied from 23 to 28.5°C, with no marked variation among the experiments and control. Mean dissolved oxygen in all experiments and control varied significantly ($P \leq 0.05$). Dissolved oxygen was higher in E1 (6.79 ± 0.21) whereas the lowest dissolved oxygen concentration was noticed in E3 (5.77 ± 0.28).

Table 1: Water quality and nutrient dynamics in aquaponics and control system

Variables	E1	E2	E3
Temperature (°C)	27.08	27.05	27.07
Salinity (ppt)	1.60 ± 0.08	1.61 ± 0.08	1.60 ± 0.08
pH	7.33 ± 0.02	7.32 ± 0.01	7.33 ± 0.01
Dissolve oxygen (mg/l)	6.79 ± 0.21	6.11 ± 0.32	5.77 ± 0.28
Hardness (mg/l)	347.95 ± 3.52	347.86 ± 3.52	348.71 ± 3.75
Alkalinity	239.71 ± 5.51	238.62 ± 6.25	237.29 ± 5.77

Water quality variables such as temperature, pH, dissolved oxygen in all the experiments and control were within the ranges suitable for rearing Tilapia. Total alkalinity and total hardness were higher in the treatments. But all the values were within the desired levels.

Table 2: Physico-chemical variables and nutrient dynamics at aquaponics and control system

Variables (mg/l)	E1	E2	E3
Ammonia	0.10 ± 0.02	0.072 ± 0.01	0.05 ± 0.02
Nitrite- N	0.056 ± 0.01	0.060 ± 0.01	0.047 ± 0.01
Nitrate -N	9.93 ± 0.91	4.25 ± 0.89	3.24 ± 0.59
Phosphate	1.03 ± 0.28	0.71 ± 0.14	0.58 ± 0.11

Nitrite-N was significantly different among experiments. Less nitrite was noticed in integrated systems (E3) because plants and bacteria removed ammonia efficiently. Nitrate-N levels were significantly different among experiments. Experiment (E1) showed higher nitrate levels compared to treatments with plants (E2 and E3) (Table 2). This showed green gram plants effectively removed nitrates.

Table: 3 Growth performance of *O. mossambicus* in aquaponics and control system

Variables	E1	E2	E3
Initial weight (g)	4.24	4.23	4.21
Initial length(cm)	4.66	4.56	4.46
Final weight (g)	6.81	5.69	5.39
Final length(cm)	6.86	6.10	5.83
% weight gain	37.03%	34.52%	28.03%

The body weight of the Tilapia during the time of harvest among all experiments varied significantly ($P \leq 0.05$). The highest growth was observed in E1 (6.81 g) followed by E2 (5.69g), while E 3 (5.39 g) had significantly lower growth (Table 3).

Table 4: Growth performance of *V. radiata* in aquaponics and control system

Variables	Soil	E1	E2	E3
Initial height (cm) (7 th day)	18.8	19.9	18.6	17.3
Final height (cm) (21 st day)	28.9	31.2	25.7	23.4
Leaf length initial (cm) (7 th day)	2.3	2.5	2.10	2.2
Leaf length final (cm) (21 st day)	3.7	3.9	2.7	2.6
Percentage of height gain	53.73	56.79	38.18	35.27

Table 5: Qualitative analysis of phytochemicals in *V. radiata*

S. No.	Phytochemicals	Water Samples	Soil Samples
1.	Protein	+	+
2.	Phenolic compounds	+	+
3.	Terpenoids	+	-
4.	Flavonoid	-	+
5.	Steroid	-	-

The results of the water quality parameters were within the appropriate range of fish culture throughout the experiment. There were no significant differences noticed in water quality parameters of fish culture tank. It is reported that Tilapia can survive at pH range of 4-11 but fish can grow faster in water that is neutral or slightly alkaline pH [2].

In the present study the recorded range of pH was 7-7.33 which found suitable for Tilapia growth. In aquaponics system the bacteria such as Nitrosomonas and Nitrobacter was taking care of water quality. Therefore, the pH was noticed within 7.2 to 8.2 whereas nitrification is inhibited below the value of pH range 5 [3]. Therefore the pH range in the present study was within the range of nitrification. Water quality variables were within the ranges which was suitable for Tilapia culture. The physico-chemical characteristics such as temperature, pH, dissolved oxygen in all the experiments and control were found within the ranges which was suitable for rearing Tilapia. Total alkalinity and total hardness were higher in the experiments and control groups, but all the values were within the desired levels.

Adequate aeration is essential in aquaponics system for proper growth and survival of the fishes. The DO concentration in the present study was found to vary from 5.77 to 6.99 ppm which was within the optimum range of Tilapia culture. The fish requires 5ppm DO for optimal growth, and if the concentration falls 2.5ppm the significant growth retardation occurred [4]. In the present study the DO concentration was decreased after 2 weeks from the starting of the experiment this might be due to microbial community and root respiration. The nitrifying bacteria growing on the root system could have contributed to oxygen uptake [5].

Tilapia has high tolerance of temperature with the optimal growth at 25 to 30°C nitrifying bacteria can operate between 7 to 35°C temperatures with the optimum of 25°C (Wortman and Wheaton, 1991). During the experimental period the average water temperature was 21.1to 32.6°C which has been reported that as optimum range for tilapia growth and yield [6].

The recorded ammonia was ranged from 0.05 to 0.027 mg/l. The suggested amount of ammonia recirculating aquaculture should be less than 1.00 mg/l [7]. Authors [8] reported that 86 to 98% of ammonium removal of ammonium nitrogen (NH_4) constructed wet land system receiving aquaculture wastewater. Ammonium is major source of inorganic nitrogen taken up by roots of higher plants [9]. There may be a less reliance on nitrification for ammonia removal when sufficient plants are present in the aquaponics system. Nitrite-N was significantly different among treatments. Less nitrite was observed in integrated systems (E3) because plants and bacteria removed ammonia efficiently.

Authors [10] reported that plant roots are more competitive for ammonium than oxidizing bacterial species (*Nitrosomonas europaea*). Increasing the nitrite content in water exerts more stress on the fish and leads to growth suppression, tissue damage, and mortality [11].

Nitrate-N is relatively harmless, and it is the preferred form of nitrogen for growing higher plants [12]. Nitrate-N is not generally of great concern to target species such as Koi Carp, and aquatic species can tolerate extremely high nitrate-N concentration that is more than 25 mg/L. Authors [13] found many fish produced in aquaculture systems can tolerate nitrate concentrations exceeding 25 mg/L. Authors [14] recommended $\text{NO}_3\text{-N}$ concentrations should not exceed 50 mg/L in waters used for culturing of fish and shellfish. In this study, $\text{NO}_3\text{-N}$ concentrations were in the favorable ranges in all experiments. In this study, the percentage of nitrate removal in aquaponics systems were also studied. Highest removal was observed in E2 followed by E1 and E3. Authors [15] found the same results of ammonia production, and subsequent nitrification was clearly increasing with higher stocking densities. Authors [16] reported 90.9% removal of nitrate using gravel bed as media in hydroponics. Net nutrient accumulation within the entire aquaponics system may be another criterion used to assess the suitability or efficiency of the integrated hydroponic subsystem.

In the present study media based hydroponic replicates removed more nitrate from the fish culture. Final net nitrate concentrations (for the aquaponics system) followed < Media Bed System < Floating Raft System < Nutrient Film Technique. The three different experiments contain final nitrate removal significantly lower in NFT replicates, suggesting that plants in NFT replicates were approximately 20% less efficient at nitrate-nitrogen removal than the other two experiments. The lower nitrate removal experiment of the NFT due to those plants in NFT experiments root-water contact area of less than 50% typical in NFT system [17]. Plants in both gravel and floating treatments had 100% of root area inundated providing more opportunity to assimilate nitrate from the respective water culture. Author [18] also found that NFT culture was less efficient in terms of nutrient removal than medial bed culture. In this test system 31% of nitrate was removed when using gravel bed and 20% of nitrates were removed using an NFT system. Previous study supports a similar conclusion of the present study where the media bed hydroponics result efficient than NFT in removing nitrate from fish culture water. The mean phosphate concentration varied significantly ($P \leq 0.05$) among all experiments. The highest phosphate concentration was noticed in E1 (1.03 ± 0.21), followed by E2 (0.71 ± 0.14), E3 (0.58 ± 0.11) (Table 2). Phosphate ($\text{PO}_4\text{-P}$) levels were significantly different among the experiments, and levels were lower in the experiments E2, and E3 compared to E1 because of the phosphate utilization by green gram plants.

Fish fed with 2% body weight in all experiments. Similarly [19] reported 5 kg of *Oreochromis niloticus* fed 2% biomass per day throughout the culture period of lettuce. Feed Conversion Ratio (FCR) varied significantly in all the experiments ($P \leq 0.05$). The FCR (food conversion ratio) was better in E1 compared to all other experiments because of weight gain of fish attained high in E1. The present result is supported by the findings of Bernier and Peter (2001). Reduced food intake levels and/or disruption of the feeding behavior is a common feature of the behavioral response to stress in fish. Declining fish growth rate and feed utilization with increasing levels of stocking densities was observed by [20]. Authors [16] compared three different types of hydroponic subsystems (gravel bed, floating raft, and Nutrient Film Technique) and recommended gravel bed or floating raft hydroponic subsystem in an aquaponics system. Therefore, a gravel bed hydroponic subsystem was used in this study for growing green gram. Growth of green gram in terms of height gain, and leaf length was higher in E1 followed by E2 and E3.

Plants have basic organic process importance by their content of macromolecule, sugar, and oil, of minerals, vitamins and water for growth and development in man and animals. Primary metabolism is important for growth and development of plants include the common amino acids, sugars, protein, pyrimidines, and purines of nucleic acid, chlorophyll etc. secondary metabolism in plants plays a major role in its environment. Attractions of pollinators, natural defense system against predators and diseases, etc. are examples of the roles of secondary metabolites [21]. The secondary metabolites formed also are an important trait for our food [taste, colour, scent etc.] and ornamental plants. Numerous plants secondary metabolites such as flavonoids, alkaloids, tannins, saponins, steroids, anthocynins, terpenoids, rotenoids etc. have found commercial application as drug, dye, flavor, fragrance, insecticide etc. such as fine chemicals are extracted and purified from plant materials [22].

Globally, research into biologically active natural products from plants has attracted many natural product chemists. The natural products play an important role in the field of new drug research and development because of their low toxicity, easy availability and low cost. The latest research observed that the bioactive and antioxidant, potentials of these plants are attributed to presence of polyphenols, flavonoids, alkaloids, terpenoids, vitamins and so [23]. The phytochemical screening of green gram plants showed that the presence of proteins, phenolic compounds, terpenoids present in under water growing plants whereas the flavonoids and steroids are absent. The soil plant contains proteins, phenols and flavonoids whereas the terpenoids and steroids were absent.

The present study comparing Media Bed, Floating and NFT hydroponic sub system for green gram, nitrate and phosphate removal, water use and buffering capacity showed that the NFT hydroponic subsystem was significantly less efficient than either Media bed or Floating raft hydroponic culture technologies. The least result for NFT might be due to probably low levels of root contact with water compared to Media Bed and Floating raft hydroponic systems [17]. Growth of tilapia over the time frame of experiment (21 days) was good and not affected by the any system.

IV. CONCLUSION

The present study conclude that the aquaponics system can capable of producing fish Tilapia and agriculture food crop green gram *Vigna radiata* in the integrated way with utilizing small land area and water. Further it is inferred that this aquaponics system completely organic and totally sustainable. Therefore, the integrated recirculating aquaponics system should be established wherever less land and water is available for the sustainable production of fish and agriculture food crops for the livelihood development and employment generation among the rural people.

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Plate 1a: 7 Day Grown *V. radiata* (Media Bed System)

Plate 1b: 21 Day Grown *V. radiata* (Media Bed System)

Plate 2a: 7 Day Grown *V. radiata* [RAFT]

Plate 2b: 21 Day Grown *V. radiata* [RAFT]

Plate 3a: 7 Day Grown *V. radiata* [NFT]

Plate 3b: 21 Day Grown Plant [NFT]

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