## Effects of supplementary nutrient in an aquaponic system for production of ornamental red tilapia (*Oreochromis* Sp.) and lettuce (*Lactuca sativa* var *longifolia*)

Rafiee Gh.R.<sup>1\*</sup>; Ros Saad Ch.<sup>2</sup>; Kamarudin M.S.<sup>2</sup>; Ismail M.R.<sup>2</sup>; Sijam K.<sup>2</sup>

Received: July 2018

Accepted: November 2018

#### Abstract

Effects of supplementary nutrient in the production of red tilapia (Oreochromis sp) and Lettuce (Lactuca sativa var longifolia) evaluated in a representative water recirculating aquaculture system. The nutrient solution supplemented was 25% level of (L25) nutrient solution as medium used for aquaponic production of lettuce in the NFT system (based on cooper's formula). Thus, a completely randomized experimental design conducted with two treatments in triplicates (Pl<sub>25</sub> and PL<sub>0</sub>). Six black rectangular tanks (114 x 86 x 100cm) used as fish culture tanks and each one equipped with three hydroponic troughs. Each tank filled with 640 L of water and aerated continuously with two circular air stones (3 L min<sup>-1</sup>) during the experiment. The system was efficiently able to remove high rate of total ammonia -nitrogen (TAN) excreted by fish during the experiment. The fish attained marketable size (200g) during a 110 –day period. Nutrient supply had not significant effects (p>0.05) on growth of fish during experimental period. The yields (Biomass/tank) of fish in treatments PL25 and Plo were 9.97 and 9.26 kg / tank, respectively. Three times cultivation and harvest of lettuce carried out during the experimental period. At the first harvest, the yield (mean wet weight) of lettuce showed significant (p < 0.05) differences between treatments, 1437g and 85 g in treatments PL25 and Pl<sub>0</sub>, respectively. In the second and third lettuce harvests, the yield of lettuce did not show any significant differences (p>0.05) and averaged 2112 and 1419 (Second harvest) and 1173 and 807 (Third harvest) for treatments PL25 and PL0, respectively. It was recorded that red tilapia could tolerate 25% of nutrient solution, used for aquaponic production of lettuce, and introduction of nutrient solution to the culture system is necessary to get higher yield of lettuce at initiation of culture system.

**Keywords:** Nutrient solution, Aquaponic system, Lettuce, Red tilapia, Nitrification, De-nitrification.

<sup>1-</sup>Department of Fishery and Environmental Sciences, Faculty of Natural Resources, University of Tehran, P. O. Box: 31585-4314, Karaj, Iran

<sup>2-</sup>Department of Agrotechnology, Faculty of Agriculture, Universiti Putra Malaysia, Serdang, Selangor, Malaysia

<sup>&</sup>lt;sup>\*</sup>Corresponding author's Email: ghrafiee@ut.ac.ir

### Introduction

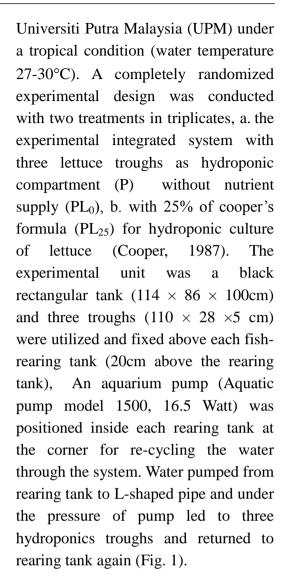
Water re-circulating systems initiated knowledge of fish based on requirements, physical and biological factors involving in the production of fish in the culture system. The potential of all physical and biological aspects and system performance in the removal of accumulated N-compounds, determine the capacity of each culture system. Total ammonia nitrogen (TAN), produced by fish, is the main pollutant which must be removed from the culture system then fecal matter and remained food that all being derived due to feeding of the fish (Rafiee and Saad, 2005). During three last decades, various biological methods utilized to treat water for reuse in intensified fish culture system (Spotte, 1970; Rackocy, 1989; Rafiee and Saad, 2005; Roosta and Hamidpour, 2011). In general, ammonia used by bacteria and it converts to nitrite, then to nitrate (Nitrification). In another process, converted under anaerobic nitrate condition to free nitrogen (N2) that the process called De-nitrification. In a recirculating aquaculture system, by using bacterial bio-filters, the toxicity of ammonia removes but whenever the low water exchange rates and high feeding rates is aimed, for higher production of fish, the accumulation of organic matter and consequent mineralized nutrients productions would be greater than nutrient removal by bacterial and physical filters. Therefore, these matters have high potential influence fish the to

production and environmental quality problems adversely. Another way for preventing the accumulative frequency of nutrients throughout a fish culture system is to link the plant for assimilating the major components produced in intensified fish culture systems, i. e., ammonia, nitrite, nitrate, then, other nutrients (macro and microelements) which are responsible for eutrophication of aquatic environments. In this respect, many research initiated in both warm (Rakocy, 1994; Recovery research system) and moderate climates (Lewis et al., 1978; Pierce, 1980; Sutton et al., 1982; Rackocy, 2000; Seawright, 1993) to utilize waste- water as solution nutrients for vegetable growth. As alkalinity declines in recirculating system as bicarbonate is consumed and acid is formed during nitrification, liming agents [CaCO<sub>3</sub>,  $CaMg(CO_3)$  2, CaO,  $Ca(OH)_2$ ] are added to maintain pH at 7.5 for integrated plant-fish culture (Rackocy and Hargreave, 1993). Nutrient accumulation increases the conductivity of culture water as well. As a result, however, nutrient accumulation is one of the critical point but to maintain the condition at steady state and desirable for fish and plant is more important. Because, it cannot be easy and the main goal, since dynamic of nutrients in integrated system are complex and some variables subjected to mineral manipulation. Therefore, the ability to control the nutrient composition of fish and plant in culture water is limited, especially at the initiation of culture system while small amount of feed added to the system. Some studies indicated that from fish culture system. insufficient quantities of essential plant nutrients obtained (MCMurtry et al., 1997; Parker et al., 1990; Rackocy et al., 1993; Cerozi and Fitzsimmons, 2017). Thus, balancing the profile of nutrient in water at the tolerating ratios or levels for fish, plant and bacteria and achieving high production of fish and plant is so important. In this regards, the aim of this study was to investigate the effect of input-nutrient on fish and vegetable growth in a recirculating aquaculture system in order to using it for ornamental fish culture.

#### Materials and methods

#### System and experimental design

This study was conducted at the Aquatic Resources Technology Laboratory, Institute of Bioscience,



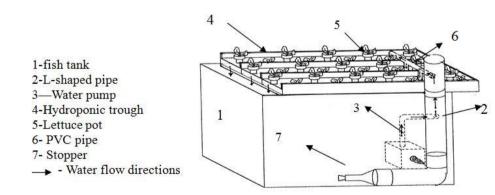


Figure 1: Schematic diagram of the system: integrated fish and plant co- culture with use of L-shaped pipe.

## Water supply

Two days before acclimating the fish, each fish tank filled with 640 L of tap water and aerated continuously with two circular air stones (3 L min<sup>-1</sup>). Water supply had following characteristics: age tap water: pH=8.1; EC= 0.24m mhos; N=1.1mg L<sup>-1</sup>; Mg=1.71 mg L<sup>-1</sup>. P=0.00031 mg L<sup>-1</sup>; Fe=0.0001 mg L<sup>-1</sup>; K=2.4 mg L<sup>-1</sup>; Mn =0.003 mg L<sup>-1</sup>; Zn=0.01 mg L<sup>-1</sup>; Ca=23.2 mg L<sup>-1</sup>.

The evapotranspiration and leaking loss of water compensated with tap water during the experiment. Prior to initiation the experiment, the fish acclimatized the system for one week. The EC of water in treatments with nutrient supply adjusted at  $0.65 \pm 0.02$ mmhos cm<sup>-1</sup>, using supplementary nutrient. Two stock solutions were prepared, using below mineral salts:

Stock solution A: Ca (NO3) 2, 3009 g; Fe -EDTA, 23g in 15 liter of tap water Stock solution B: KNO3, 1700g; MgSO4, 1539g; H2KPO4, 789g; MnSO4, 18.3g; HBO3, 5.1g; ZnSO4, 1.3g in 15 liter of tap water. 800 ml from each stock solution taken and added in each experimental tank to get 25% of nutrient solution, based on hydroponic production of lettuce in NFT system.

L-shaped pipe utilized in all systems as a settling chamber of fecal mat (5 inches diameter, 0.6-meter horizontal length and 1.5-meter vertical length). Each pipe positioned in parallel beside the rearing tank. A valve conducted to the end of horizontal part of the pipe for possible cleaning.

## Feed and feeding

The fish fed a floating pellet diet, containing 32% Protein (min), 6% Fat (max), 6% Fiber (max) and 11% moisture (max) that ordered from Car-gill Company, for grower of red tilapia.

## Production of lettuce seedlings

experiment Two weeks prior to commencement, for production of lettuce seedling, seeds of lettuce sowed in some sheets of sponge ( $40 \times 60$  cm) already cut to small pieces (3 ×3cm). seeds irrigated The daily until germinating, each newly seedling transferred to a small perforated plastic cups and transferred to hydroponics troughs.

## Protocol

The experiment run on March 5, and terminated on 25 August (110 Days). At the day of experiment commencement, the 45 cups, each comprising a oneweek aged seedling of lettuce, placed into the holes of polystyrene sheets that already fixed inside all NFT troughs. Three times planting and harvesting of lettuce were carried out during the experiment (averagely, with a five-week period). Each rearing tank stocked with 75 red tilapia juveniles with the average and standard length of 5.62±3.75 g and  $5.73 \pm 1.41$  cm, respectively. The fish fed twice a day ad libitum at 09.00 h and at 15.00h.

#### Water quality monitoring

Dissolved oxygen (DO) and water temperature (T) in fish tanks measured twice a week by YSI Model 57. Electroconductivity (EC) was determined twice a week using EC meter model HANA instrument conductivity meter HI 8033. The pH was determined using Orion model 410A pH meter. Total ammonia (NH<sub>3</sub>+NH<sup>+</sup><sub>4</sub>) and Nitrite-Nitrogen (NO2<sup>+</sup>) were measured (after dilution of the samples). Nitrate (NO<sub>3</sub><sup>+</sup>) was measured weekly (AHPA, 1980).

#### Data analysis

Data of variables subjected to pairedcomparison t-test analysis, using the software of SPSS (version 10.0).

#### Results

#### Fish production

The fish growth was not significantly different (p>0.05) between treatments at the end of experimental period. The average biomass of fish was 9.97±2.41 and 9.26±1.34 kg in treatment PL<sub>25</sub> and PL<sub>0</sub>, respectively at the end of experimental period (Table 1; Fig. 2). The rate of survival in treatment PL<sub>25</sub> was significantly lower (p < 0.05) than treatments PL<sub>0</sub> (Recorded plus rates of jumping) (Table 1). The SFG and FCR significantly rates were different (p < 0.05) between treatments at the end of experimental period.

# Table 1: Mean (mean ± S.D.) of fish biomass (FB), individual fish weight (IFT), specific growth rate (SGR), Feed Conversion Ratio (FCR), survival (Sur) between treatments at the end of experimental period.

-	Treatments	FB (kg)	IFT (g)	Sur (%)	SGR (g)	FCR
_	Treatments	8/			(8)	
	$PL_{25}$	$9.97 \pm 2.41^{a}$	185±23 <sup>a</sup>	73±25.43 <sup>b</sup>	1.61±0.20 <sup>a</sup>	1.25±0.15 <sup>b</sup>
	$PL_0$	9.26±1.34 <sup>a</sup>	211±14 <sup>a</sup>	$58{\pm}08.57^{\ a}$	1.84±0.12 <sup>b</sup>	$0.88{\pm}0.07^{\ a}$

The same subscript letters in a column are not significantly different at the 0.05 levels.

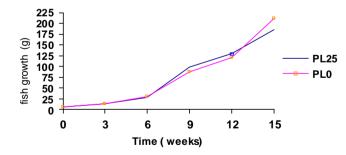


Figure 2: Changes in red tilapia growth between treatments during the experimental period.

#### Vegetable production

The lettuce seedling were cultivated and harvested three times during the experimental period. In the first time, the seedlings replaced through the hydroponic troughs five days after running the systems and harvested thirty days later. The growth rate of seedlings in treatments with supplementary nutrient solution (PL<sub>25</sub>) was significantly higher (p < 0.05) than treatment without nutrient solution  $(PL_0)$ . 1437±339 and 85±94. respectively. In second and third harvest of lettuce, the yield of lettuce did not show any significant differences (p > 0.05) between treatments. These rates in treatments PL<sub>25</sub> and PL<sub>0</sub> were averagely 2112±697 and1685±192 respectively for first and 1173±402, and 807±180g, respectively for third harvest (Fig. 3).

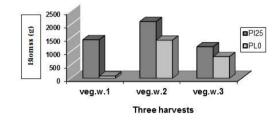


Figure 3: Mean yields of lettuce gained from three times planting and harvesting during the experimental period (veg. w. 1, 2, 3 = biomass of lettuce in first, second and third harvests in different treatments).

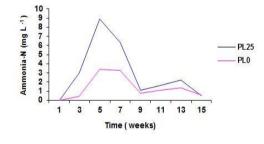


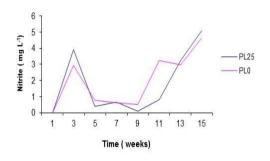
Figure 4: The changes of TA concentration in different treatments (in rearing fish tanks) during experiment.

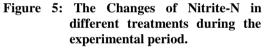
## Water quality parameters Total ammonia (TA)

TA concentration in treatments continuously increased and after five weeks reached to more than 10 mg  $L^{-1}$  (PL<sub>25</sub>). Afterward the concentration of

TA continuously decreased and one week prior to termination of the experiment got to the  $0.32 \text{ mg L}^{-1}$ . Nitrite (NO<sub>2</sub><sup>+</sup>): The concentration of nitrite did not show any significant

differences (p > 0.05)between treatments. After two weeks. the concentration of nitrite increased up to  $3 \text{ mg L}^{-1}$  in both treatments. Within the last two weeks, an increase in nitrite both concentrations recorded in treatments and reached to  $4 \text{ mg } \text{L}^{-1}$  (Fig. 4).





*Nitrate*  $(NO_3^+)$ 

With using nutrient solution in treatment  $PL_{25}$  at the commencement of experiment, concentration of nitrate increased up to 75 mg L<sup>-1</sup>. After on smart depletion of nitrate occurred in treatments PL25. The nitrate concentration ranged 0.023 to 6.39 mg L<sup>-1</sup> at the end of experiment (Fig. 5).

## Dissolved oxygen (DO), Temperature (T), pH and EC

DO did not show considerable depletion in rearing tanks and at the bottom of L shaped pipe. DO ranged 6 -  $8.5 \text{ mg L}^{-1}$  in rearing fish tanks. Water

temperature was between 27-30°C during the experimental period. The EC rates in different time intervals (in tanks) showed significant rearing 9<sup>th</sup> weeks. differences (p < 0.05) by afterward did not show any significant differences (p > 0.05)between treatments. The EC of water constantly increased in both treatments and reached up to 0.86 and 0.85 mmhos cm<sup>-</sup> <sup>1</sup> in PL 25, PL0, respectively (Fig. 6).

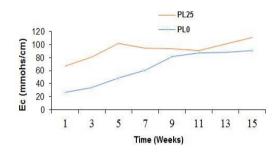


Figure 6: Average changes of EC in different treatments during the experimental period.

At the start of experiment, pH were adjusted at 7.8 (mixed aged tap and well water, prior to adding nutrient). When nutrient added to treatments PL<sub>25</sub>. It came down and reached to below 7. The sharp depletion of pH recorded in all treatments during the experiment and continuously showed a decrease. After 5 weeks pH reached to below six in both treatments, after that slowly increased and got to 6.5 in 8<sup>th</sup> weeks. Afterward had fluctuation and ranged from 5.4 to 7.1 by the end of the experiment (Fig. 7).

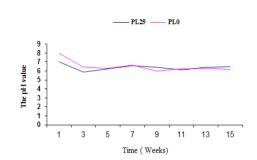


Figure 7: The pH changes in different treatments during the experimental period.

#### Discussion

In this study, with use of simple feasibilities, a new design for production of fish in integrated water re-circulating fish culture system with use of supplementary nutrient introduced. The system demonstrated could successfully that remove increasingly of Naccumulation compounds through experimental period. The result showed that seven weeks after starting the experiment, systems were able to hold the of total concentration ammonia-N below the 48h LC<sub>50</sub> 2.5 mg  $L^{-1}$  reported for Sarothrodon aurea (Redner and Stilkney, 1979). The concentration of nitrite also averaged 3 mg  $L^{-1}$  (Fig. 4) at the end of experiment that it was less than the threshold already reported in catfish, 96h mean tolerance of 7.5 mg  $L^{-1}$  (Sutton and Lewis, 1982). The concentration of nitrate in treatments with nutrient supply was 74 mg  $L^{-1}$  at the start of experiment, afterward, smart depletion of nitrate occurred in particular in treatments PL<sub>25</sub>. Nitrate depletion probably was due to activities of plants and bacteria in assimilation of nitrate. Existence of nitrite in rearing

tanks also emphasized that the nitrification de-nitrification and activities occurred by bacteria during the experiment; However, at the start of experiment there was not anv concentration of ammonia and nitrite in water supply. At the first of the experiment, lack of nutrient was evidence in treatments without nutrient supply because of unsuitable growth of vegetable. When water EC approached to the level of 0.33 mmhos (after 40 days), moderately growth of lettuce occurred in comparison with the nutrient supply system, meaning that Ec for introduction standard of vegetable in integrated system could be 0.33 mmhos. however profile of nutrient must be considered for standardizing the Ec. Concentration of nutrient in water did not reach the levels commonly utilized in commercial production of vegetable in hydroponic system, large amount of feed suggested approximately 8-10 kg/m<sup>3</sup> must be used to get suitable medium for culture of most vegetable in hydroponic system (Rackocy et al.,, 1993). It was resulted excretion of fish that and biodegradation of organic to inorganic matter just provide moderate nutrients required for vegetable (lettuce) culture, emphasizing the effect of nutrient solution either on growth of plant or further removal of N-compounds. Foliar use of nutrient demonstrated as one of practical method to improve nutrient deficiency in hydroponic part of integrated system (Roosta and Hamidpour, 2011). Such a system even

emphasis on permanent production of leafy vegetables for local and civil regions due to balancing the nutrient in different aquaponics design (Kyaw and Andrew, 2017). The growth of fish was not significantly different between treatments. it accentuated that supplementary nutrient had not significantly negative effect on growth of fish. The fish reached to marketable size (200 g) within 110 days. In a natural condition at 23°C, tilapia (Sarothrodon nilocticus) reaches to 200 g weight within six months (Naegel, 1977). However, it has been reported that tilapia shows maturity after six months of age, but during this experiment some fish started displaying mating behaviors and color changes when attained 100 g weight (after two months) and showed territorial operations and attacked to each other, this condition can be related to welfare condition of fish in the culture system. One of the most important points of this experiment was sharp depletion of pH, from about 8 below 6 during the first four weeks of experiment and below seven for next period, indicating reactions of De-nitrification by bacteria. Depletion of pH had also observed already by other experimenter (Naegal, 1977; Rakocy et al., 1995; Rakocy, 1989). Acidic conditions not only have an important role in neutralizing the toxicity of high concentration of ammonia in rearing tank but also affecting solubility of some elements such as Fe (ferrous), Zn (Zinc) and

Phosphorus (P) which can be more available to plant.

Depletion of N-compounds in all treatments accentuated that the lettuce and L- shaped pipe chamber efficiently operated as de-nitrification parts. Low concentrations of ammonia. nitrite and nitrate and floating of fecal mat above the vertical part of the pipe indicated the ability of horizontal part of Lshaped pipe in de-nitrification 1993). (Seawright, Since. denitrification occurs under anaerobic condition, high accumulation of waste fish inside the horizontal compartment of the L-shaped pipe probably prepared anaerobic condition for activity of denitrificant bacteria. In fact, It can be said that this part of system operated similar to both nitrification and denitrification bio-filters. These compartments separately utilized by some scientists for production of fish and vegetable throughout water recirculating system (Naegal, 1977: Sutton, 1982; Lewis, 1978; Pierce, 1980; Rakocy, 1984; Wren, 1984; Zweig, 1986; McMurtry et al., 1990; Rafiee et al., 2005).

Comparing the result of this experiment with other works, showed that the efficiency of present system (PL25 and PL0) for high production of fish and vegetable and maintenance the water quality parameters at optimum ranges. If the land area surrounded for production of fish and vegetable considered, the usage of less space (1 m<sup>3</sup> area) and high production of fish vegetable within 15 weeks and

dominated in this study. Considering the fish yield of 31.5 kg/m<sup>2</sup>, for cat fish (Lewis *et al.*, 1978), 9.20 kg/m<sup>2</sup> for Nile tilapia (Watten and Budch, 1984) and 13.61-16.41 kg/m<sup>2</sup> for present system. system The present comprised vegetable plant and nutrient use, PL25, showed capable for production of fish and vegetable in urban and local regions in tropical condition, especially for ornamental plant and fish culture. Determining the roles of biological and physical compartments separately in assimilation of nutrient required for future studies. Since, nutrient supply affects de-nitrification and nitrification bacteria (Austin and Austin, 1989). The impact of nutrient on activity of bacteria and biodegradation of organic to inorganic matter also require further studies.

## References

- APHA (American Public Health<br/>Association) American Water Works<br/>Association, and Pollution Control<br/>Federal (16th ed.), APHA,<br/>Washington, DC (1980), p. 1268
- Austin, B. and Austin, D.A., 1989. Bacterial Fish Pathogen Diseases in farmed and Wild Fish. Chi Chester: Ellis Harwood.
- Cerozi, B.S. and Fitzsimmons, K., 2017. Phosphorus dynamics modeling and mass balance in an aquaponics system. *Journal of Agricultural Systems*, 153, 94-100.
- Cooper, A., 1979. The ABC of NFT Grower book, London, 181P.

- Kyaw, T.Y. and Andrew, K. Ng., 2017. Smart aquaponics system for urban farming. *Journal of Energy Procedia*, 143, 342-347.
- Lewis, W.M., Yoop, J.H., Schramm, H.L. and Brandenburg, A.M., 1978. Use hydroponics of to maintain quality water of recirculated water in a fish culture **Transactions** ofsystem. the American Fisheries Society, 107(1), 92-99.
- Mackay, K.P., and Tover, W., 1981. An ecological approaches to water re-circulating for salmonids: preliminary experience. Bioengineering symp. For fish culture (FCS publication No 1), 249-258.
- MacMurtry, M.R., Nelson, P.V., Sanders, D.C. and Hodges, L., 1990. Sand culture of vegetables using recirculated aquacultural effluents. *Journal of Applied Agricultural Research*, 5(4), 280-284.
- Naegel, L.C.A., 1977. Combined production of fish and plants in recirculating water. *Aquaculture*, 10, 17-24.
- Parker. D., Anouti. A. and Dickenson, G., 1990. Experimental integrated results fish/plant production system. Environ. Res. Report ERL 90-34 Lab. no. (unpubl.), 12P.
- Pierce,B.,1980.WaterreusesaquaculturesystemsintwogreenhousesinnorthernVermont.

Proceedings of the World Mariculture Society, 11, 18-127.

- Rafiee G.R., Saad, C.R., Kamarudin,
  M.S., Sijam, Ismail, K. and Yusop,
  M.R., 2002. A simple technical feasibility for production of fish and vegetable in an integrated recirculating system, Proceeding of Second International Conference on Sustainable Agriculture for Food, Energy and Industry, Beijing, 8-12 September 2002.
- Rafiee G.R. and Saad, C.R., 2005. Nutrient cycle and sludge production during different stage of red tilapia (*Oreochromis* sp.) growth in a recirculating aquaculture system. *Journal of Aquaculture*, 244,109-118.
- Rakocy, J.E., 1989. Vegetable hydroponics and fish culture a productive interface. *World Aquaculture*, 20 (3), 42-47.
- Rakocy, J.E., Hargreaves, J.A. and Bailey, D.S., 1989. Effects of hydroponic vegetable production on water quality in a closed recirculating system. *Journal of Scientia Horticulture World Aquaculture Society*, (64A), 201.
- Rakocy, J.E., 1989. Hydroponic lettuce production in a recirculating fish culture system. Univ., pp. 4-10. Virgin Islands Agriculture Exp. Station, Island Perspect 3.
- Rakocy, J.E., 1994. Waste water management in integrated recirculating system. PP 75-80. University of the Virgin Islands Agricultural Experiment Station, RR

2, Box 10,000 Kingshil, VI 00850, U,A. Bull. Nalt. Res. Inst. Agric, Suppl. 1.

- Rakocy, J.E., Hargreaves, J.A. and Bailey, D.S., 1993. Nutrient accumulation in a recirculating aquaculture system integrated with hydroponic vegetable production. 112-136. in: Wang, J.K. (Ed.), Techniques for Modern Aquaculture. Proceedings of a Conference, 21-23 June 1993, Spokane, WA.
- Rakocy, J.E., 1995. The roles of plant crop production in aqua cultural waste water, Aqua cultural Engineering and waste management, proceedings from the aquaculture Expo VIII and Aquaculture in the Mid-Atlantic Conference, Washington, D.C. June 24-28, 1995.
- Rakocy, J.E., 2000. Integrating tilapia culture with vegetable hydroponics in recirculating systems, Journal of World. *Aquaculture society*, 1(1), 163-184.
- Render, B.D. and Stickney, R.R., 1979. Acclimation to ammonia by *Tilapia aurea. Transactions of the American Fisheries Society*, 108(4), 383-388.
- Roosta, H.R. and Hamidpour, M., 2011. Effect of foliar application of some macro and micro –nutrients on tomato plant in aquaponic and hydroponic systems. *Journal of Scientia Horticulturae*, 129, 396-402.
- Seawright, D.E., 1993. A method for investigating nutrient dynamics in integrated aquaculture, hydroponics

systems. In: Wang, J.K. (Ed.), Techniques for Modern Aquaculture. Proceedings of a Conference, 21-23 June (1993), Spokane, WA, pp.137-147.

- **Spotte, S.H., 1970.** Fish and invertebrate culture. Water management in closed system. 145 pp. Wiley-Inter science, New York.
- Sutton, R.J. and Lewis, W.M., 1982. Further observations on a fish production system that incorporates hydroponically grown plants. *Prog. Fish-Cult.*, 44(1), 55-59.
- Watten, B.J. and Busch, R.L., 1984. Tropical production of Tilapia, *Sarothrodon aurea*, and Tomato, *Lycopersicon esculentum*, in small scale re-circulating water system. *Aquaculture*, 41, 271-283.
- Wren, S.W., 1984. Comparison of hydroponic crop production techniques in a recirculating fish culture system. M.S. thesis, Texas A&M University, College Station, TX, P: 66.Zar J. H. 1996. Biostatistical analysis, 662P. 3<sup>rd</sup> ed. Prentice Hall, New Jersey.
- Zimmerman, R., Iturriaga, R. and Kiel, U.K., 1978. Simultaneous determination of the total number of aquatic bacteria and the number thereof involved in respiration *American Society for Microbiology*, 36(6), 926-935.
- Zweig, R.D., 1986. An integrated fish culture hydroponic vegetable production system. pp: 34-40. Aquaculture. Magazin., May/June.