Nutrient influx, Water quality and growth performance of Nile tilapia fry fed Recycled Food Waste Based Diet in a Closed Recirculating Fish Culture System

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Abstract

The nutrient influx, water quality and growth performance of Nile tilapia Oreochromis niloticus fed recycled food waste-based diets was studied in a closed recirculation system for 11 weeks, during which no water renewal was carried out in the system. Fish (mean weight 1.2 ± 0.11 g) were fed with different levels of recycled waste materials supplemented with lysine (lys) and methionine (met), and were designated as D1 (0% FIW+SSW), D2 (57.90 % FIW+SSW + lys and met) and D3 (54.84% FIW+SSW only). The results showed that fish fed D2 diet had a higher (P<0.05) growth performance and feed efficiency than those fed the other diets. The biofilter used in the recirculation system effectively converted ammonia to nitrate, as such toxic ammonia and nitrite were negligible. However inorganic phosphorus was similar among the treatments. The water coloration and turbidity were higher in the tanks of fish fed D3 diet. The results of this study revealed that inclusion of recycled food waste materials had less negative impact on the water quality in a closed recirculating fish culture system.

Key words: *Food industry waste; Growth performance; Recycled food waste; Soy sauce waste; Tilapia fry.*
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1. Introduction.

The huge expansion experienced by the aquaculture sector is faced with the challenges of sustainability and growing pressure to operate under strict environmental safety standards. This pressure is created by the growing population, thereby increasing the competition for both water and land. To meet these challenges, different aquaculture systems have been developed to maintain high biological carrying capacity in relatively little space (Quillere, *et. al*., 1993; Twarowska, *et. al.*, 1997). The closed recirculation fish culture system provides an alternative means of intensive production that reduces environmental impacts and allows greater control of factors that might affect growth. It also allows direct treatment of wastes and its re-use, the ability to limit the release of antibiotics to the external environment, and control of pH, temperature and chemistry. Such fine manipulations of the culture environment help to optimize fish growth. However, one of the major problems with the closed recirculation system is related to the rate of accumulation of suspended solids, particularly very fine particles. The presence and accumulation of particulate wastes in closed recirculation system (faeces, uneaten feed, and bacteria flocs) will impact negatively on the water quality by affecting the performance of the water treatment units. Hence water quality parameters are the major concerns in closed recirculation systems because relatively small concentrations of some of these parameters can be lethal (Eddy, *et. al*., 1983, Chen, *et. al.*, 2006). One of the ways to reduce this problem may be the reduction of nutrients in aquaculture effluent.

Aquaculture intensification has resulted in culturing activities with a higher degree of feed input and waste output per rearing system. This waste output has adverse effect on the quality of the rearing water system, since the enriching nutrients originate from the feed added to the rearing system (Hardy and Gatlin III, 2002). It has been well established that nitrogen is associated with the most expensive component of feed. Regardless of species, protein is the most important dietary nutrient in all fish diets; and fishmeal remains one of the major sources of dietary protein (Kaushik, 1990; Lu, *et. al.*, 2009).

Formulation of fish feeds containing high levels of conventional and non-conventional alternative protein source has become a major focus in aquaculture nutrition research. This has become imperative because of the ever-increasing cost and uncertain availability of fishmeal. Hence conventional plant proteins (soybean, rapeseed, etc.) appear to be the most suitable alternatives for fishmeal in fish diets. However, their scarcity as well as competition from other sectors like livestock, human consumption and industrial use makes their cost too high, thereby placing them far beyond the reach of average farmers or aqua feed producers (Fasakin, 1999). In Japan, there is a basic law establishing a recycling-based society, with emphasis on the minimization of associated biodegradable waste by-products by recycling them for effective reuse. With recent developments in technology, a concerted effort to recycle and reuse food waste is increasing.

The partial or complete replacement of fishmeal (FM) with alternative plant or animal proteins generally results in an imbalance of amino acids, which can affect fish growth and protein utilization efficiency, resulting in higher nitrogen (N) loading into the culture medium (Rumsey, 1993, Sugiura, 1998). Improvement of fish feed formulations through partial replacement of FM with ingredients low in phosphorus (P) is essential for lowering P loading (Takeuchi, *et. al.*, 1993; Cho and Bareau, 2001; Sugiura, 1998). Hence, optimal replacement of FM without negative effect on the fish growth and culture medium is highly desirable to fish nutritionists.

Tilapia are known as “aquatic chicken” because of their fast growth, good quality flesh, disease resistance, adaptability to a wide range of environmental conditions, ability to grow and reproduce in captivity, and feed on low trophic levels. Thus, they have become an excellent choice for aquaculture, especially in tropical and subtropical environments (El-Sayed, 2006; Gonzales, *et. al.*, 2007). Because of the importance of these species, it is critical that feeds are both economically and environmentally sustainable in any fish production culture.

In our previous studies we demonstrated the suitability of recycled food waste materials from food industry waste (FIW) and soy sauce waste (SSW) in the practical diet of Nile tilapia fry and inclusion of these recycled food waste up to 58% enriched with lysine (lys), methionine (met) and arginine (arg) was also achieved (Bake, *et. al.*, 2010). Thus, the main objective of this study was to investigate the nutrient influx, water quality and growth performance of Nile tilapia fry fed recycled food waste based diet in a closed recirculating fish culture system.

2. Materials and methods

2.1 Culture Facilities:

The experiment was carried out at the Laboratory of Fish Culture, Tokyo University of Marine Science and Technology, Japan using replicated closed recirculating aquaculture systems. The system consists of a 30-liter rectangular fiberglass aquarium, which is the culturing chamber, a sedimentary chamber for solid waste collection and the bio-filtration chamber, which harbors the bacterial community for nitrification. The culturing chamber was completely closed to the atmosphere and was equipped with a slide glass cover to the aquarium and can be opened during water addition to compensate for evaporation and also during other aquaculture rudimentary work. Water temperature was maintained at 28±0.5 ^{o}C using electric heaters placed in each tank. The aquariums were provided with continuous aeration through an air compressor and illuminated by overhead fluorescent lights to maintain a constant photoperiod of 12 h light and 12 h dark cycle (8:00-20:00) throughout the study. The sediment tank used was the TUF (Tokyo University of Fisheries) column system according to Satoh, *et. al*. (1992).

This solid waste collector was attached to each aquarium and then connected to the bio-filter chamber. The solid waste was usually collected from the sediment tank once a week. The EMEH 2213330 model bio-filter tank was used. Inside the bio-filter tank is the synthetic filter media, which were the bacterial carriers for nitrification; coral sand were placed on top of the filter media to buffer the pH of the water system, and foam layer was placed between the coral sand and the water outlet of the pump of EMEH cylinder. Each system had an independent recirculating system and the water flow was maintained at 900 ml/min for each aquarium. Bacterial community for nitrification process was established in bio-filter tanks two weeks before the commencement of the feeding trial. After the establishment of the bacterial community in the bio-filter tanks, the systems were cleaned up and 33 liters of fresh dechlorinated tap water was supplied to each system without any water exchange throughout the experimental period. Distilled water was used for compensation of the loss due to evaporation or sampling for water quality analysis.

2.2 Diet formulation and preparation

Ingredients and diet formulation

Soy sauce waste: Soy sauce waste (SSW) was produced by Yamasa Corporation and processed by Nippon Formula Feed Mfg. Co. Ltd. After the fermentation of soybeans and soy sauce extraction, the residual cake, which is a waste product was collected, dried and recycled by dehydration to reduce the moisture to a low level. Crude protein and lipid contents of SSW were 26.13% and 11.93%, respectively.

Food industry waste: The food industry waste (FIW) used in this study was obtained from Nippon Formula Feed Mfg. Co. Ltd. It includes leftover food from convenience stores, food waste residues discharged during processing, hotel waste, restaurant cooking waste, tofu waste and bread production waste. Fry-cooking the waste with vegetable oil at a very low pressure processed the FIW and an initial temperature of between 80 - 100^{o}C maintained for 1 hour and later increased to 100 - 110^{o}C for about 30 minutes after which the product was allowed to cool off before grinding it into a powdered form. Crude protein and lipid contents of FIW were 19.60% and 11.34%, respectively.

Fishmeal: The fishmeal (FM) used in this experiment was obtained from Nippon Formula Feed Mfg. Co. Ltd. The crude protein and lipid contents of FM were 63.50% and 11.79%, respectively.

Experimental diets

Based on the nutritional requirements of tilapia (NRC 1993), three isonitrogenous and iso-lipidic diets were formulated at 35% protein and 9.5% lipid, containing different amount of recycled food waste materials supplemented with lysine (lys) and methionine (met) and were designated as D1 (0% FIW+SSW), D2 (57.90 % FIW+SSW + lys and met) and D3 (54.84% FIW+SSW only).

2.3 Fish and Feeding

The tilapia, *O. niloticus* fry (average weight: 1.21 ± 0.27g) used for this experiment were obtained from pure–bred tilapia broodstock in the Fish Culture Laboratory of the Tokyo University of Marine Science and Technology Japan. Twenty fish were stocked in each 30-liter aquariums and fed for 11 weeks. Two replicates of each treatment were reared for each of the 3 diets and were hand fed 4 times daily at 80% of Japanese Fisheries Agency Feeding rate standard at 7:00, 10:00, 14:00, and 17:00. Feeding rates were subsequently adjusted according to their growth rate per aquarium. Fish were denied feed 24 h prior to sampling. 10 fish were randomly sampled and weights were measured using a digital electronic weighing balance (AW220; Shimadzu Corporation, Kyoto, Japan) on a weekly basis. Total length and standard length were measured using digimatic calipers (CD-20CP; Mitutoyo Corporation, Tokyo, Japan).

2.4 Water sampling and Analytical procedure.

Water quality was measured throughout the experimental period. To determine the quality of the rearing water, samples were collected from each aquarium before feeding using clean, plastic bottles. Water temperature and dissolved Oxygen in the system were monitored daily in the morning by Dissolved oxygen (DO) meter (HQ 30d, HACH Company, Colorado, USA). Water samples were taken once a week from the surface water near the center of each aquarium for subsequent analysis for pH, ammonia-nitrogen (NH_4-N), nitrite–nitrogen (NO_2-N), nitrate–nitrogen (NO_3-N), phosphorus, coloration and turbidity. Concentration of these elements in rearing water was analyzed with reagents of water analysis (HACH Company, Colorado, USA) using the following methods for each element, ammonia nitrogen: Nessler method, nitrite nitrogen: Diazotization method, nitrate nitrogen: Cadmium reduction method and Phosphorus: Ascorbic acid method and analyzing them under ultra violet and visible light spectrophotometer (UV-1200, Shimadzu Corporation, Kyoto, Japan). Data collected from NH_4-N, NO_2-N and NO_3-N were used to determine the total dissolved nitrogen. (APHA, AWWA and WEF 2005). pH reading was taken using electronic pH meter (HM-5BS model. TOA electronics Ltd. Japan). Coloration and turbidity were measured according to JIS standard.

2.5 Sample collection and Biochemical analyses

The experimental diets, fish body samples, solid waste materials, the sludge and the rearing water were subject to chemical analysis at the end of the experiment. Fish were weighed individually at the beginning of the experiment and subsequently once a week using an electronic balance (EB-3200D, Shimadzu Corporation, Kyoto, Japan). The average of total weight of the fish in each tank was used as a unit of observation for analysis. Upon termination of the experiment, 10 fish from each aquarium were randomly selected for the chemical analyses of the whole body. Whole body samples were pooled from 5 fish per aquarium and minced by a centrifugal mill (Retsch ZM 200, Germany) fitted with a 0.25-mm screen. The homogenate was collected and kept at $-20^{O}C$ until analysis. Proximate analyses were done according to the methods detailed in Takeuchi (1988) and Folch *et. al.* (1957).

2.6 Evaluation of growth parameters.

Growth Rate, Food Efficiency and Sedimentation of each element in tanks: substance balance (culture water, sedimentation, fish, and bubble-separated substances).

Specific growth rate of wet body weight (SGR %) = $(\ln W_t - \ln W_0) / t \times 100$.

Feed efficiency (FE %) = $(Wt - W_0) / TFI \times 100$.

where:
Wt = Final body weight of the fish at the end of the experiment (g)
W_0 = Initial body weight (g)
TFI = total feed in take (g)
t = Time (days)

2.7 Statistical analyses.

Data were analyzed via one-way analysis of variance (ANOVA) using Statistica 6.0 (Stat-Soft, Inc., USA). Differences between treatments were compared by

Tukey's test. Level of significance was tested at $p<0.05$.

3. Results

3.1 Chemical analysis of the experimental diets

All the formulated diets used in the present study were iso-nitrogenous and iso-lipidic. Tables 1and 2 shows the feed formulation, cost of production, proximate composition and mineral content of the formulated experimental diets. D3 had the lowest cost of production (¥8.96/100g) while D1 had the highest cost of production (¥25.56/100g). The proximate composition of the experimental diets were similar, however, the moisture content was slightly lower in D1 than in the others treatments. Furthermore there was no much differences in the mineral contents of the experimental diets except for zinc that was higher in D1 than the other treatments.

Table 1. Formulation of the experimental diets for *O. niloticus* fry (g/kg)

Ingredients	D1	D2	D3
Fishmeal [*1]	55.12	25.00	32.90
Soy sauce waste	0.00	28.95	27.42
Food industry waste	0.00	28.95	27.42
α Starch	16.37	4.00	4.50
Vitamin premix [*2]	2.00	2.00	2.00
P-Free Mineral mixture [*3]	4.00	3.00	3.00
$Ca(HPO_4)2.H_2O$	3.00	2.76	2.76
Soybean oil	3.10	0.00	0.00
Lysine	0.00	1.17	0.00
Methionine	0.00	1.17	0.00
Cellulose	16.41	0.00	0.00
Total	100.00	100.00	100.00
**Cost of each diet/100g	¥25.56	¥9.96	¥8.96
*1 Fishmeal: Anchovy fish meal from Chile			

*Composition (mg/100g); Thiamin HCl 6, riboflavin 10, pyridoxine HCl 4, cynaocobalamin 0.01, ascorbic acid 500, niacin, 40, Ca-pentothenate 10, inisitol 200, biotin,0.6, folic acid, 1.5, p-aminobenzoic acid 5, vitamink3 5, vitamina A acetate 4000IU, vitamin D3 400IU; *3 Composition (g/100g); NaCl 5.0, $MgSO_4.7H_2O$ 74.5, $FeC_6H_5O_7.nH_2O$ 12.5; trace element mixture[4]5.0 cellulose 3.0; [*]4 Composition (mg/g): $ZnSo_4.7H_20$ 353, $MnSO_4.5H_20$ 162; $CuS0_4.5H_20$ 31, $AlCl_2.6H_20$, $Kl0_2$ 3 Cellulose 440. **Cost of feeding stuffs at the prevalence market prices in Japan (January 2010): IUSD=¥91.00

Table 2. Proximate and mineral composition of the experimental diet

Diet code	D1	D2	D3
Moisture (%)	3.0	5.5	4.8
Crude Protein	35.1	37.8	36.9
Crude lipid	9.8	9.3	9.4
Ash	12.9	13.0	14.6
Energy (Kcal/g)	19.2	18.5	18.5
N (mg/g)	57.9	64.1	62.0
P (mg/g)	19.3	17.1	17.3
Ca (mg/g)	28.9	29.4	32.0
Mg (mg/g)	3.2	2.1	2.5
Na (mg/g)	17.5	26.0	24.3
K (mg/g)	19.7	25.6	19.1
Fe (µg/g)	1454.9	1112.1	1223.1
Zn (µg/g)	261.8	184.6	200.0
Mn (µg/g)	87.4	72.4	74.5
Cu (µg/g)	38.4	59.6	59.3

3.2 Growth and feed conversion

It was observed that there was no feed rejection by the fish fed the experimental diets, and they vigorously ingested the experimental diets. The change in weight per week and growth performance data of Nile tilapia fry fed the experimental diets for 11 weeks are summarized in Fig. 1 and Table 3. Table 3 shows that fish fed D2 had the highest values of all growth performances and was significantly different from the other treatments (P<0.05). On the other hand, fish fed D1 had the lowest final body weight (22.99±0.05g) however it was not significantly different from D3 (24.14±0.16g) (P>0.05). The SGR, FE and TFI of the tilapias fed experimental diets followed the same pattern as the final body weight. Survival of the fish fed experimental diets ranged from 97.5 – 100%. Survival rate of D2 was significantly higher than the others (P<0.05).

3.3 Chemical composition of the tilapia fry

Proximate composition, and mineral content of the whole fish body, which were sampled at the start and the end of the feeding trial are given in Table 4. Although the highest moisture content was observed in the fish fed D3 and the lowest value in D1, these values were not significantly different among the treatments. The carcass protein of the fish fed experimental diets does not differ significantly among the treatments. D2 had significantly higher body lipid than both D1 and D3. There were no significant differences among the treatments for the ash content. Except for P, Ca, Na Mn and Cu, initial body mineral contents were higher in comparism with mineral content levels of the experimental fish.

Table 3. Growth performances of *O. niloticus* fry fed experimental diets for 11 weeks in a closed recirculating system

Diet code	Av. Body weight (g)		Survival rate (%)	Specific growth rate (%)	Feed Efficiency (%)	Total feed consumed (g/fish)	Biomass conversion (%)
	Initial	Final					
D_1	1.21±0.02^a	22.99±0.24^{bc}	97.50±3.54^b	3.79±0.06^b	125.95±4.48^b	17.30±0.42^b	33.29±0.51^b
D_2	1.21±0.03^a	28.61±0.16^a	100.00±0.00^a	4.11±0.01^a	138.02±2.29^a	19.85±0.21^a	39.38±0.87^a
D_3	1.21±0.04^a	24.14±0.16^b	97.50±3.54^b	3.85±0.06^b	129.20±2.42^b	17.75±0.21^b	33.67±0.41^b

Fig. 1. Growth of *O. niloticus* fry fed the experimental diets for 11 weeks in a closed recirculation system.

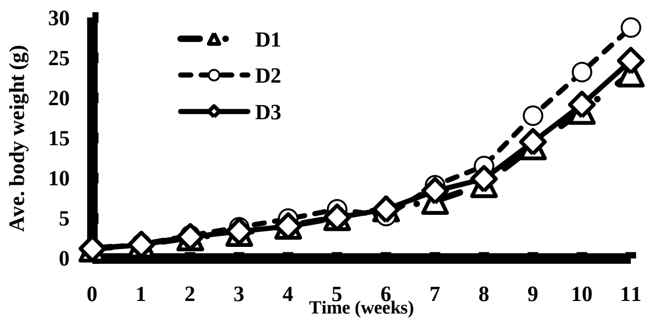

Table 4. Proximate and mineral composition of whole tilapia fed experimental diet

	Initial	Final		
		D1	D2	D3
Moisture (%)	77.9	74.5	73.2	75.2
Crude Protein	14.8	14.2	14.3	14.2
Crude lipid	3.6	4.6	5.0	4.4
Ash	3.4	5.8	5.7	5.8
N(mg/g)	107.3	88.9	85.3	91.7
P (mg/g)	29.6	37.3	33.6	35.1
Ca (mg/g)	45.2	68.5	64.2	61.0
Mg (mg/g)	0.3	0.3	0.2	0.2
Na (mg/g)	14.8	17.3	16.6	16.7
K (mg/g)	27.6	27.0	27.3	21.7
Fe (μg/g)	335.7	176.8	196.5	179.1
Zn (μg/g)	242.9	125.8	130.6	133.9
Mn (μg/g)	11.8	42.6	44.4	42.1
Cu (μg/g)	26.5	85.9	91.8	91.7

Fig. 2. Water quality variations in the culture system of Nile tilapia fed experimental diets for 11 weeks.

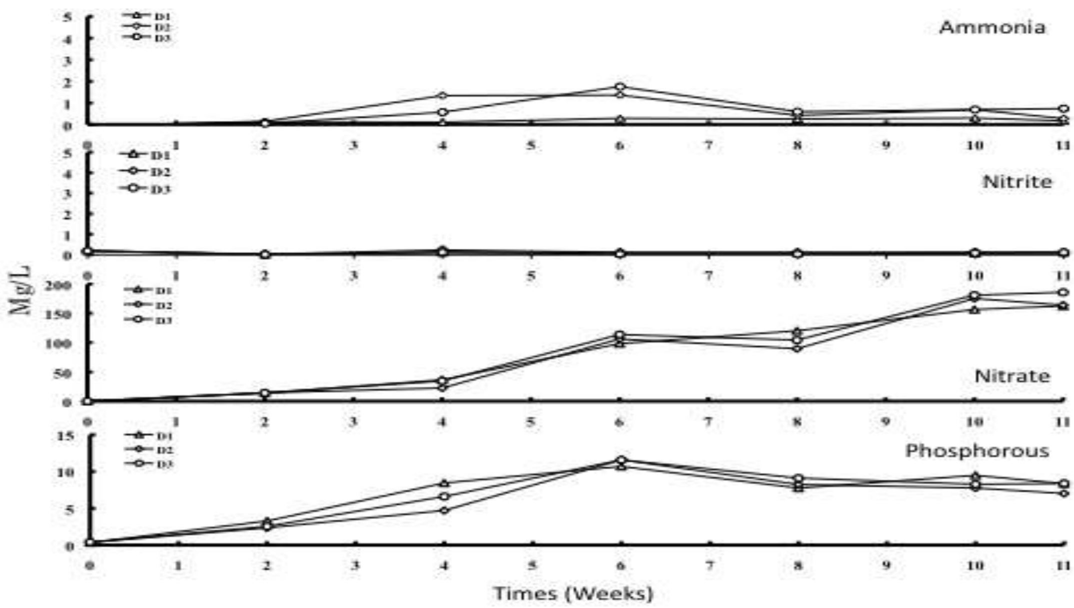

Fig. 3. Water quality variations in the culture system of Nile tilapia fed experimental diets for 11 weeks.

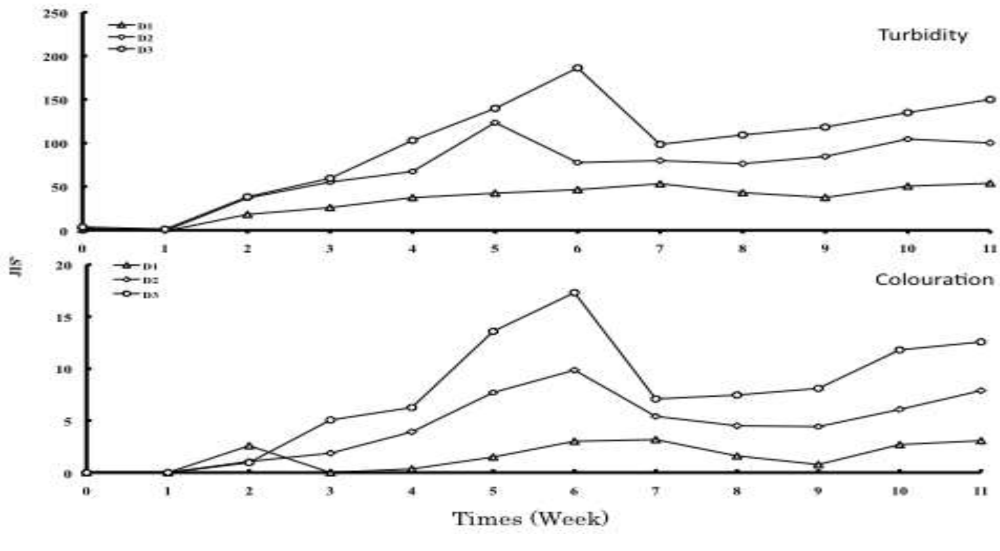

Fig. 4. pH variations in the culture system of Nile tilapia fed experimental diets for 11 weeks

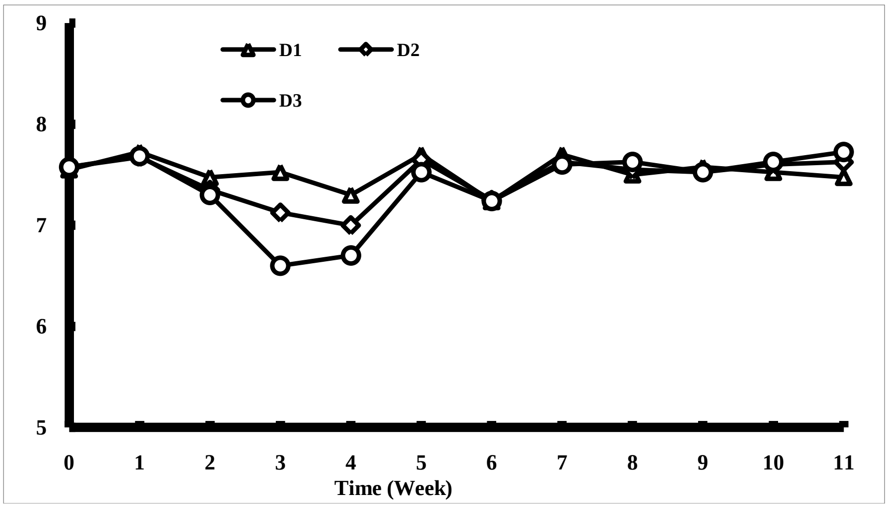

Table. 5. Final concentration of the mineral elements in the rearing water.

	D1		D2		D3	
	Initial	Final	Initial	Final	Initial	Final
TIN (mg/L)	0.19	162.87	0.19	164.44	0.19	186.74
P (mg/L)	0.33	8.81	0.37	6.85	0.39	8.62
Ca (mg/L)	40.78	111.49	40.78	47.58	40.78	78.15
Mg (mg/L)	5.34	15.74	5.56	8.89	5.50	11.16
Na (mg/L)	75.90	71.85	70.43	59.63	75.89	64.51
K (mg/L)	42.64	64.07	54.17	66.51	49.43	73.63
Fe (µg/L)	100.64	3.82	89.66	42.93	90.09	27.07
Zn (µg/L)	33.73	21.85	31.47	24.31	32.37	28.80
Mn (µg/L)	0.00	0.00	0.00	0.00	0.00	0.00
Cu (µg/L)	160.05	45.00	175.84	24.18	117.94	0.00

4. Discussion

Growth parameters serve as indicators of fish's ability to utilize and retain nutrients from a given diet. The growth performance of the Nile tilapias fed experimental diets in this study were comparable to our previous results obtained from flow throw system (Bake, *et. al.,* 2010). The closed recirculating system used in this study despite not sophisticated, proved effective throughout the rearing period. The growth performances of the fish depicted that Nile tilapias have the ability to thrive in water containing high concentrations of nitrate. This agrees with the reports of Endo, *et. al.* (1999, 2000). The better growth performance of the fish fed D2 may be attributed to the essential amino acid (EAA) supplementation of the diet, This agrees with our previous work that supplementation of recycled waste material with EAA can improve the growth performance of Tilapia by augmenting for the deficient EAA in the recycled waste materials (Bake *et. al.*, 2010). This is also evident in the total feed consumed per fish and the final weight. All the fish in this study showed normal growth with high FE. The FE was above 100% for all the experimental fish, indicating the diets were of high quality and the fish adequately utilized the nutrients.

Ammonia nitrogen and nitrite-N remained below maximum tolerable levels for Nile tilapia. NH_3-N values, recorded in this study, were lower than the one reported by Leclerq and Hopkins (1985) and were much less than the lethal levels of 2.4

mg/L (48h-LC50) reported by Redner and Stickney (1979). Ammonia-N is toxic to commercially cultured fish at concentrations above 1.5 mg (Daud *et. al*., 1988). Nitrate-nitrogen (NO_2-N) and nitrate-nitrogen (NO_3-N) are the products of ammonia oxidation; furthermore aquatic species can tolerate extremely high (greater than 100 mg/L) concentrations of NO_3-N (Ebeling, *et. al*., 2002). However, the toxicity threshold depends strongly on the species, size, fine solids, refractory organics, surface-active compounds, metals, and nitrate (Colt, 2006). In the context of this study the bacteria community in the biofilter tanks contributed to the increase of nitrate nitrogen in the rearing water of all experimental tanks by the efficient conversion of ammonia-nitrogen in the rearing tanks thus, the system tend to have the ability to support intensive aquaculture without much adverse effect on the environment. The gradual change in the color of the rearing water may be attributed to the color of the diets; however, the coloration of the rearing water did not tend to have any negative effect on the fish.

The pH is an important water quality parameter in recirculating systems as toxicity of other compounds to fish, especially ammonia and chlorine, are affected by pH. Lawson (1995) reported that as pH decreases, ammonia is converted into a less toxic ammonium form; therefore, the increase in pH will lead to the accumulation of ammonia in the system. High pH values tend to facilitate the solubilization of ammonia, heavy metals and salts. Low pH levels tend to increase carbon dioxide and carbonic acid concentrations. Lethal effects of pH on aquatic life occur below pH 4.5 and above pH 9.5. In this study the pH concentration in all the treatments did not drop below 6.0. This suggests that the coral sand were very effective to buffer the acid with nitrate production.

The most cost–effective diet in terms of production cost is D3. However with the inclusion of limited quantity of lysine and methionine, D2 gave a better growth performance than D1 and D3, hence in a long term cost analysis of the diets containing recycled food waste and a limited quantity of lysine and methionine D2 could be more profitable in term of yield. The reduction in the per kilogram feed cost can facilitate the use of recycled food wastes in the diets. Moreover the fact that recycled food wastes can be available throughout the season, and no competition for humans use compared to the other conventional alternative proteins appears to be an additional advantage.

In conclusion this study reveals that a combination of recycled food waste supplemented with EAA gave a higher growth performance and lower negative impact on the water quality parameters and the system, than the other tested diets thus recycled food waste can be used as ingredient in the diet of Nile tilapia with less Nutrient influx in a simple closed recirculation systems. However there is still a need to evaluate the use of this waste in a closed recirculation system on a longer term.

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References

APHA (American Public Health Association), (2005). American Waterworks Association, and Pollution Control Federal 16th ed., APHA, Washington, DC, pp. 1268.

Bake, G. G., M. Endo, A. Akimoto and T. Takeuchi (2010). Effect of reducing fishmeal content by using recycled food waste materials supplemented with lysine, methionine and arginine for a practical diet of Nile tilapia Oreochromis niloticus fry. Fish. Sci. (In press).

Chen, S., Ling, J., Blancheton, J., (2006). Nitrification kinetics of biofilm as affected by water quality factors. Aquacultural Engineering. 34, 179-197.

Colt, J., (2006). Water quality requirements for reuse systems. Aquacultural Engineering, 34, 143–156.

Cho C.Y. and Bureau D.P. (2001): A review of diet formulation strategies and feeding systems to reduce excretory and feed wastes in aquaculture. Aquacult. Res. 32: 349–360

Daud, S.K.; Hasbollah, D. and Law, A.T. (1988). Effect of unionized ammonia on red Oreochromis mossambicus/O. niloticus hybrid) fry. Pages 411- 413 In Pullin, R.S.V.

Ebeling, J.M. and Summerfelt, S.T, (2002). Performance evaluation of a full-scale intensive recirculating aquaculture system's waste discharge treatment system. In: Rakestraw, T.T, Douglas, L.S. and Flick, G.J.(Eds), The Fourth International Conference on Recirculating Aquaculture, Virginia Polytechnic Institute and State University, Blacksburg, VA, pp. 506-515.

Eddy, F., Kunzlik P. and Bath, R. (1983). Uptake and loss of nitrite from the blood of Rainbow trout, Salmo gairdneri Richardson, and Atlantic salmon, Salmo salar L. in fresh water and dilute sea water. J. Fish Bio. 23: 105-116.

El-Sayed, A.-F. M. (2006). Tilapia Culture. CAB International, Wallingford, UK, 277 p.

Endo M., Takeuchi, T., Yoshizaki, G., Toyobe M., Kanki , Suzuki K., Oguchi M. and Kibe S. (1999). Studies on the development of closed ecological recirculating aquaculture system (CERAS) IV. Along term feeding experiment with Oreochromis niloticus in the fish-rearing closed tank. CELSS J., 12: 17-24.

Endo M., Takeuchi, T., Yoshizaki, G., Satoh S., Omori K., Oguchi M. and Nakajima A. (2000). Studies on the development of closed ecological recirculating aquaculture system (CERAS) VI. Phosphorus form and mineral budgets in the fish- rearing closed tank during a long-term feeding experiment with tilapia. CELSS J., 13: 19-26.

Fasakin E. A., Balogun, A. M. and Fasuru B. E. (1999). Use of duckweed, Spirodela polyrrhiza L. Schleiden, as a protein, feedstuff in practical diets for tilapia, Oreochromis niloticus L. Aquacult. Res. 30: 313-318.

Folch J, Lee M, and Sloane-Stanley, G. H. (1957). A simple method for the isolation and purification of total lipids from animal tissues. J. Biol. Chem. 226: 497-509.

Hardy, R. W. and Gatlin, D. M. III (2002): Nutritional strategies to reduce nutrient losses in Intensive aquaculture. In: Cruz-Suárez LE,Ricque-Marie D, Tapia-Salazar M, Gaxiola-Cortés MG, simoes N (eds). Avances en Nutrición Acuícola VI Memorias del VI Simposium.Internacional de Nutrición Acuícola. Cancún, Quintana Roo, México. 23–34.

Kaushik, S. J. (1990). Use of alternative protein sources for the intensive rearing of carnivorous fish. In:Mediterranean Aquaculture (eds R. Flos, L. Tort and P. Torres), pp. 125–138. Ellis Horwood Limited

Redner, R. and Stickney, R. R. (1979). Acclimation to ammonia by Tilapia aurea. Tran Am. Fish Soc. 108:383-388.

Lawson, T. B. (1995). Fundamentals of Aquacultural Engineering. Chapman and Hall Publishers, NewYork.

Leclercq, D. I. and Hopkins, K.. (1985). Preliminary test of an aerated tank system for tilapia culture. Aquacult. Eng. 4:229–304.

Gonzales, M. J., Hutson, H. A., Rosinski, E. M., Wu, V. Y., Powless, F. T. and, Brown B. Paul (2007): Evaluation of Fish meal–free diets for first feeding Nile tilapia, Oreochromis niloticus. J. Appl. Aquaculture. 19(3): 89-99.

National Research Council, NRC (1993). Nutrient Requirements of Fish. National Academy Press, Washington, DC, 114pp.

Lu, J., Li, J., Furuya, Y., Yoshizaki, G., Sun, H., Endo, M., Haga, Y., Satoh, S. and Takeuchi T. (2009): Efficient productivity and lowered nitrogen and phosphorus discharge load from GH-transgenic tilapia (Oreochromis niloticus) under visual satiation feeding. Aquaculture. 293: 241-247.

Quillere, I., Marie, D., Roux, L., Gosse, F. and Morot-Guadry, J. F.(1993). An artificial productive ecosystem based on a fish/bacteria/plant association: 1. Design and management. Agric. Ecosyst. Environ. 47:13–30. Aquacult. Eng. 16: 133–147.

Rumsey, G. L. (1993). Fishmeal and alternate sources of protein in fish feeds. Fisheries 18: 14–19.

Satoh, S., C.Y. Cho, and T. Watanabe (1992) Effect of faecal retrieval timing on digestibility of nutrients in rainbow trout diet with the Guelph and TUF faeces collection systems. Nippon Suisan Gakkaishi, 58 : 1123–1127.

Sugiura, S. H., (1998). Development of low-pollution feeds for sustainable aquaculture. PhD Thesis, University of Washington, Seattle, WA.

Takeuchi, T., Hoshi, M., Satoh, S., Watanabe, T., Takashima, Y. and Kawamata, T. (1993). Effects of dietary digestible energy and available phosphorus contents on total amount of nitrogen excretion from carp. Suisan Zoshoku 41: 359–365.

Takeuchi, T. (1988). Laboratory work- chemical evaluation of dietary nutrients. In: Watanabe T (ed). Fish Nutrition and Marine Culture. Japan International Cooperation Agency, Tokyo. pp. 179-233.

Twarowska, J. G., Westerman, P. W. and Losordo, T. M. (1997). Water treatment and waste characterization evaluation of an intensive recirculating fish production system. Aquacult. Eng. 16: 133–147.