

Assessment of the Optimal Replacement Levels of Maize with Water Lettuce Leaf (*Pistia Stratiotes*) based Diets for *Clarias gariepinus*

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Abstract:

A 56 days-feeding trial was carried out to investigate the effects of Water Lettuce Leaf Meal as a partial replacement for Yellow Maize Meal in the practical diets for the freshwater mud catfish (*Clarias gariepinus*) juveniles. Five diets in which WLLM replaced YMM at 0%, 25%, 50%, 75% and 100%; designated as Diet 1 (control), Diet 2, Diet 3, Diet 4 and Diet 5 respectively. The replacement levels were prepared and fed to *Clarias gariepinus* juveniles. At the end of the feeding experiment, the replacement effects on the growth and nutrient utilization indices; weight gain (WG), specific growth rate (SGR), protein efficiency ratio (PER), Food conversion ratio (FCR), percentage survival rate (PSR) and total fish production (TFP) were determined. The treatment fed with control diet 1 WLLM (0%) gave growth and nutrient utilization results as expected. Weight gain (70.60 g) in WLLM (Diet 1) was significantly higher ($p < 0.05$) than 38.32 g for WLLM (Diet 5), but not significantly higher than 69.70 g for WLLM (50%). Also, SGR 3.10 g/day (diet 3) was significantly higher than other diets except control diet 1 (0%) WLLM. Likewise, FCR observed in the group fed control diet 1 (2.45) was not significantly better than diet 2 (2.69) and diet 3 (2.53). Empirically, there was no significant difference ($p > 0.05$) between the feed efficiency of practical diets 1, 2 and 3; PSR in the group fed 50% WLLM (86.67) was significantly different than values observed in groups fed other diets. Hence, made diet 3 (50% WLLM) having incredible total fish production value of 7.49 kg/m³.

Keywords: Maize; Growth response; Freshwaters; Water lettuce leaves meal; Biological value; Net energy

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Introduction

Pistia is a genus of aquatic plant in the arum family Araceae with *Pistia stratiotes* as the only species often called water cabbage, Nile cabbage or shell flower (Quattrocchi, 2000). It was earlier discovered in Africa from Nile near Lake Victoria as a free floating aquatic plant found growing abundantly in Tropical and sub-tropical region of the world and detests frost in the temperate during winter. Water cabbage is a perennial monocotyledon with thick - soft leaves that form a rosette and one of the least favoured floating macrophytes for herbivores (Singh, 1977). This plant inhabits lakes, ponds ditches, canals etc. with economic importance as a vegetable in India and serves as a breeding and nursery ground for mosquitoes of the genus *mansonia* (Vershney and Singh, 1976; Park, 2007). It contains needful amount of macro and micro nutrients capable of meeting fish growth and development, such as protein, carbohydrate, minerals, vitamins, fat content. Water lettuce is used for feeding Chinese carps in china and its usage has not been profitably realized as feed ingredients in developing countries where ripple effects of exogenous factors lead to food and nutrition insecurity.

Maize (*zea mays*) is the most widely grown grain crop throughout the Americas with 332 million metric tons grown annually on the United State alone (FAO, 2009). Maize is used by agricultural bodies and research institute such as FAO and SIRO, the grain is an excellent high- energy animal feed and highly digestible. Hence, use as the basis for high energy rations for fish production, fattening cattle and lambs. It contains less fibre, less biological value and fewer minerals than most other cereal grains. The family *Clariidae* is divided into two genera- *Clarias* and *Heterobranchus*; *Clarias gariepinus* is a member of the gnathostomatus chordate. At various geographical locations, it bears different names for example; it is called *C. lazera* in the northern and central Africa, *C. senegalensis*, in East Africa, *C. mossambicus*. In West Africa and in South Africa, it is called *C. gariepinus*. They are distributed all over the world but primarily, indigenous catfish occurring in fresh water throughout Nigeria (Viveen et al., 1985).

Dietary completion and technological treatment of plant feedstuffs could increase their uses in fish feeds and may reduce reliance on conventional feedstuffs (Mbonge, 2007). Although, large amounts of plant feedstuffs were used as animal husbandry feeds. Relatively, few have been used in fish feeds, because fish required high levels of dietary protein to energy with low level of fiber (Shiau and Peng, 1993). Numerous earlier works reported possible use of plant proteins in fish feeds including (Soltan, 2002; Soltan, 2005). The extent of feedstuffs utilization of plant origin depends on availability, acceptability by fish, processing method and nutritive value. Comprehensively, understanding fish nutrition is a prerequisite for successful fish culture (Adedokun et al., 2016). Culturally, the soaring cost and demand for qualitative fish feeds from conventional feed resources necessitates fish culturists of the developing countries to incorporate cheap and locally available ingredients in fish feeds. As the net income realized from the enterprise increases, the long practicing fish farmers would remain in the business due to cheaper fish feed. This, also

enhance quality and quantity of feed given the fish for improved output and productivity (Oluwasola and Ige, 2015). Recently the utilization of aquatic plants having high biological value are used to supplement fish feed has taken a new dimension for producing the much required animal protein at low cost (Lakshmanan et al., 1967). Therefore, water lettuce does not face the same competition as conventional feedstuff (maize). According to Rumsey (1993), increased use of well processed plant protein supplements in fish feed reduces the cost of fish meal. This paper reports the growth performance and nutrient utilization of African catfish fed water lettuce based diets.

Materials and Methods

Experimental fish and design

This experiment was carried out in the research and training fish farm centre of Oyo State College of Agriculture and Technology Igbora, Nigeria. The fish *Clarias gariepinus* were obtained in the College, acclimatized for three (3) days inside the bowls. The experiment was set up in a completely randomized design (CRD) with five treatments replicated three times each as diet I control (0%) WLLM, diet II (25%) WLLM, diet III (50%) WLLM, diet IV (75%) WLLM and diet V (100%) WLLM. The *Clarias gariepinus* of average weight of 14.77 ± 0.26 g were distributed randomly and accordingly to the treatments containing different inclusion levels of WLLM. Each bowl has stocking density of twenty (20) juveniles totaling three hundred (300) juveniles for Fifteen (15) plastic bowls and aeration of water was achieved using central air pump that was connected to each bowl through air stones. Fish were weighed using a portable electric balance of OHUAS-LS-400 g and the initial weighing was carried out after 3 days of acclimation.

Collection and preparation of water lettuce leaf meal

As part of management practices, weeding of Mature water lettuce plants was achieved by hand pulling, collected and brought to the feed mill of research and training fish farm centre in OYSCATECH where the leaves were removed and washed with clean water to remove the dirt and other debris, drained properly and dried at room temperature (25°C), they were ground with a domestic hammer mill to powdered form, packed and kept in air-tight polythene bags until needed (Plate 1).

Seasonal variation effects of water lettuce

The effects of this leaf throughout the year in the tropical region can be better put as follows:

Positive effects includes: It outcompetes algae for nutrients in the water thereby preventing massive algal blooms; provides food for fish, turtles and water fowls; lower water temperature.

Negative effects includes: It blocks view of water below; may take over the surface of the pond; it blocks air-water interface which reduces the amount of dissolved oxygen; it impairs smooth navigation in the larger pond

Generally during rainy season, the leaf becomes too massive,



Plate 1: Water lettuce plant.

die in cold areas and possibly polluting the water system. However, the dry season is noted for optimization of positive effects of the leaf.

Diets preparation and feeding trial

The fish diet ingredients were procured from Ibadan Oyo State. The diets used in the experiment were formulated using the Pearson square method. The diets were iso-nitrogenous (40% crude protein) and contained 0, 25, 50, 75 and 100% inclusion levels of water lettuce leaf meal. The 40% dietary protein level was chosen on the basis that *Clarias spp* strived well on protein level between 40-45%. The ingredient were grounded and mixed properly by hand to have the desirable mixture that could be easily handled during diet preparation (Fagbenro and Arowosoge, 1991). The percentage composition of the experimental diets is shown in **Table 1**. The diets were assayed for dry matter, crude protein, lipid

(ether extract) and ash according to standard procedures of AOAC 1999. Fish were fed twice a day between 7:00-9:00 am and 7:00-8:00 pm at 5% body weight. Pellets were spread by hand to air dry and feed given was adjusted weekly after weighing.

Measurement of growth parameters

The growth parameters according to the method described by Olvera-nova et al., (1990) were used as follows:

$$\text{Percentage weight gain} = \frac{\text{Weight gain}}{\text{Initial weight}} \times 100$$

$$\text{Specific growth rate (SGR)} = \frac{\ln W_f - \ln W_o}{\text{Time (days)}} \times 100$$

$$\text{Feed Conversion Rate (FCR)} = \frac{\text{Total weight of dry feed offered (g)}}{\text{Total live weight gained by fish (g)}}$$

Protein fed = Feed intake × % CP in diet

$$\text{Protein efficiency ratio} = \frac{\text{Weight gain}}{\text{Protein fed}}$$

$$\text{Percentage survival} = \frac{\text{Number of fish harvested}}{\text{Number of fish stocked}} \times 100$$

(Where, the natural Logarithm Wf and Wo are the final and initial weight respectively, T is the time of the experiment.

Statistical analysis of data

One-way analysis of variance (ANOVA) was carried out to determine the effect of diets on growth and nutrient utilization parameters using 15.0 versions of SPSS (1999) statistical package. The Duncan's Multiple Range Test was used to determine the differences between the treatment means. The alphabetical notation was used to mark the differences at significant level of an alpha 0.05 (Alatise et al., 2014).

Water quality management

Water quality was monitored every week throughout the

feeding experiments. Water temperature (°C) and hydrogen ion concentration (pH) were measured daily with a combined digital pen-type meter while the dissolved oxygen (mg/l) was measured using the Winkler's method and conductivity by a digital conductivity meter (APHA AWWA WPCF, 1999).

Results and Discussion

Proximate composition of (*Pistia stratiotes*) leaf meal and yellow maize meal

The ability of a feedstuff such as leaf meal in fish diets can be evaluated on the basis of its proximate chemical compositions. The proximate composition of water lettuce (*Pistia stratiotes*) leaf meal and yellow maize meal are presented in **Table 2**. The results showed that the aquatic plant leaf meal was found to be moderately lower in crude protein (9.04%), fat (1.16%) and Nitrogen Free Extract (49.55%) compared with the yellow maize meal which contained crude protein (11.83%), fat (5.47%) and Nitrogen Free Extract (66.08%). The results also showed that crude fibre (19.13%) and crude ash (21.12%) in water lettuce leaf meal were higher than the values obtained for yellow maize meal with crude

Table 1: Percentage composition of the experimental diets.

Feed Ingredients	Dietary Treatments				
	DT 1 (0% WLLM)	DT 2 (25% WLLM)	DT 3 (50% WLLM)	DT 4 (75% WLLM)	DT 5 (100% WLLM)
Water lettuce	0.00	5.25	10.50	15.75	21.00
Yellow maize	21.00	15.75	10.50	5.25	0.00
Danish Fish meal	23.50	23.50	23.50	23.50	23.50
Soya bean meal	23.50	23.50	23.50	23.50	23.50
Vitamin premix	2.00	2.00	2.00	2.00	2.00
Methionine	1.00	1.00	1.00	1.00	1.00
Lysine	1.00	1.00	1.00	1.00	1.00
Vegetable oil	2.00	2.00	2.00	2.00	2.00
Bone meal	2.00	2.00	2.00	2.00	2.00
Salt	0.50	0.50	0.50	0.50	0.50
Total	100	100	100	100	100

Table 2: Proximate composition (% dry weight) of water lettuce (*Pistia stratiotes*) leaf meal.

Chemical analysis	Water lettuce leaf meal	Yellow maize meal
Moisture content	6	5.52
Crude protein	9.04	11.83
Crude fat	1.16	5.47
Crude fibre	19.13	15.14
Ash	21.12	1.48
NFE*	49.55	66.08
Gross energy	265.2	386.92
Digestible energy	164.92	250.91

*Nitrogen Free Extract (NFE) is calculated by difference = 100 - (protein + lipid + fibre + ash); Gross energy = Caloric value of protein 5.65, NFE 4.1 and lipid 9.45 kcal/g (NRC, 1993); Digestible energy = Caloric value of protein 3.5, NFE 2.5 and lipid 8.1 kcal/g (Brett, 1973).

fibre (15.14%) and crude ash (1.48%) respectively. The moisture content of water lettuce leaf meal was higher (6.00%) than the yellow maize meal (5.52%). These values agree with the results reported by Rodríguez et al., (2000) for water lettuce leaf meals. The gross energy was higher in yellow maize meal (386.92 kcal/g) than the water lettuce leaf meal (265.20 kcal/g). The digestible energy was also higher in yellow maize meal (250.91 kcal/g) than the water lettuce leaf meal (164.92 kcal/g).

Proximate composition of water lettuce leaf meal based diets

The proximate composition of water lettuce leaf meal based diets is presented in Table 3. The results showed that diet 1 has the highest value of protein (41.56%), preceded by diet 5 (40.01%), while diet 2 has the lowest value (39.84%). The crude fat and NFE were highest and assumed the same trend pattern in diet 2 (10.12%), diet 4 (10.11%), diet 3 (10.10%), diet 5 (9.98%), while diet 1 has the lowest value (9.34%) and diet 2 (34.02%), followed by diet 4 (33.39%), diet 3 (33.72%), diet 5 (33.28%), while diet 1 has the lowest value (33.17%) respectively. Diet 5 has the highest value (5.87%) of crude fibre, followed by diet 4 (5.61%), diet 3 (5.42%), diet 2 (5.13%) while diet 1 has the lowest value (4.50%) of crude fibre. The moisture content of the water lettuce leaf meal based diets ranged from 2.00% (diets 2, 3, 4 and 5) and 3.00% (diet 1). The gross energy value ranged from diet 5 (456.82 kcal/g) to diet 2 (460.21 kcal/g) and digestible energy value also ranged from diet 5 (304.04 kcal/g) to diet 1 (306.46 kcal/g).

Water quality parameters (WQP)

The mean water quality of the rectangular plastic trough system

at weekly intervals during the experimental period is presented in Table 4. The water quality was monitored every week throughout the feeding trials. The water temperature ranged between 26.14 to 27.80°C with mean water temperature of 27.11°C. The dissolved oxygen also ranged between 4.56 to 7.24 mg/l with mean dissolved oxygen value of 6.49 mg/l, pH 6.88 to 7.40 with mean pH of 7.02 and conductivity ranged from 270.00 to 532.00 $\mu\text{moh/cm}^3$ with mean value of 347.00 $\mu\text{moh/cm}^3$. These water quality ranges fall within acceptable range of APHA AWWWA WPCF (1999). The survival rates ranged from 56.67% to 86.67% with mean survival rate 71.67%. Diet 3 had the highest survival rate.

Growth and survival rates of African catfish (*Clarias gariepinus*) fed water lettuce leaf meal based diets

The growth and survival rates of African catfish (*Clarias gariepinus*) fed water lettuce leaf meal based diets is presented in Table 5. The mean body weight gain of 70.60 g was highest in fish fed 0% water lettuce leaf meal along with 100% maize meal (control diet), followed by diet 3 containing 50% water lettuce leaf meal along with 50% maize meal (69.70 g), and the lowest mean body weight gain of 38.32 g was observed with diet 5 containing 100% water lettuce leaf meal along with 0% maize meal. There was no significant difference ($p>0.05$) between the mean body weight gain for diet 1 (70.60 g), diet 2 (60.09 g) and diet 3 (69.70 g) while diet 4 (44.55 g) and diet 5 (38.32 g) exhibited significant difference respectively. The mean specific growth rate of 3.04%/day was highest in fish fed control diet containing 0% water lettuce leaf meal along with 100% maize meal, followed by diet 3 containing 50% water lettuce leaf meal along with 50%

Table 3: Proximate composition (% dry weight) of water lettuce leaf meal based diets.

Parameters	DT 1	DT 2	DT 3	DT 4	DT 5
	(0% WLLM)	(25% WLLM)	(50% WLLM)	(75% WLLM)	(100% WLLM)
Moisture content	3	2	2	2	2
Crude protein	41.56	39.84	39.86	39.88	40.01
Crude fat	9.34	10.12	10.1	10.11	9.98
Crude fibre	4.5	5.13	5.42	5.61	5.87
Ash	11.43	10.89	10.9	11.01	10.86
NFE*	33.17	34.02	33.72	33.39	33.28
Gross energy	459.07	460.21	458.91	457.76	456.82
Digestible energy	304.04	306.46	305.62	306.45	304.07

Table 4: Mean water quality of the rectangular plastic trough system at weekly intervals during the experimental period.

Weeks	Temperature (°C)	Dissolved oxygen (mg/l)	pH	Conductivity
0	26.5	7.1	6.88	270
1	27.8	7.24	6.88	276
2	27.8	7.24	6.88	276
3	26.14	4.56	6.88	532
4	27	5.82	6.9	408
5	27	6.44	6.9	378
6	27.5	6.8	7.4	314
7	27.14	6.7	7.4	322
Mean	27.11	6.49	7.02	347

maize meal (3.10%/day), and the lowest mean specific growth rate of 2.25%/day was observed with diet 5 containing 100% water lettuce leaf meal along with 0% maize meal. Diet 1 (3.04%/day), diet 2 (2.90%/day) and diet 3 (3.10%/day) showed no significant difference while diet 4 (2.48%/day) and diet 5 (2.25%/day) were significantly different in mean specific growth.

The best food conversion ratio of 2.45 was recorded in control diet, followed by diet 3 (2.53) with no significant difference ($p > 0.05$) between the food conversion ratio of diets 1, 2 and 3 as shown in **Table 5**. The poorest food conversion ratio of 4.38 was recorded in fish fed 100% water lettuce leaf meal without maize meal (diet 5). It should be noted that the lower the value of food conversion ratio, the better for the fish as less diets will be converted into flesh. This implies that the *Clarias gariepinus* Juveniles utilized diets 1, 2 and 3 better than diets 4 and 5. The best feed efficiency of 40.94% was recorded in control diet, followed by diet 3 with approximately 40%. There was no significant difference ($p > 0.05$) between the feed efficiency of empirical diets 1, 2 and 3 as shown in **Table 5**. The poorest feed efficiency of 24.43% was recorded in fish fed 100% water lettuce leaf meal without maize meal (diet 5). The feed efficiency is simply the reciprocal of food conversion ratios (1/FCR), the greater the feed efficiency, the better for the fish as less diets will be converted into flesh. FEs greater than 50% are considered good i.e. $\frac{3}{4}$ growth. The mean protein efficiency ratio of 1.01 was highest in fish fed control diet containing 0% water lettuce leaf meal along with 100% maize meal, followed by diet 3 containing 50% water lettuce leaf meal along with 50% maize meal (0.93), and the lowest mean protein efficiency ratio of 0.61 was observed with diet 5 containing 100% water lettuce leaf meal along with 0% maize meal. The significant difference ($p > 0.05$) for PER assumed the same trend pattern for MGR, SGR and FCR in diet 1 (1.01), diet 2 (0.93) and diet 3 (1.00) and in diet 4 (0.71) and diet 5 (0.61) respectively as shown in Figure 1. The mean total fish production of 7.49 kg/m³ was highest in fish fed diet 3 containing 50% water lettuce leaf meal along with 50% maize meal, followed by diet 1 containing 0% water lettuce leaf

meal along with 100% maize meal (5.76 kg/m³), and the lowest mean total fish production of 3.00 kg/m³ was observed with diet 5 containing 100% water lettuce leaf meal without maize meal. There was no significant difference ($p > 0.05$) between the mean total fish production of diet 1 (5.76 kg/m³) and diet 3 (7.49 kg/m³) and no significant difference between diet 2 (4.62 kg/m³), diet 4 (3.43 kg/m³) and diet 5 (3.00 kg/m³) respectively.

Acceptability of the diet is the common problem associated with alternative sources of feed stuffs (Rodriguez et al., 1996). In this present investigation, all the experimental diets were accepted by *Clarias gariepinus* Juveniles and utilized differently as evidenced in growth response parameters. This implies that the levels of incorporation of water lettuce leaf meal did not affect the palatability of the diets but utilization. This might be due to the processing method employed in this study. This sun drying and the grinding techniques might reduce the amounts of anti-nutritional factors in the water lettuce leaf meal thereby increasing its palatability in *Clarias gariepinus*. The observation agrees with the report of (Fagbenro, 1999; Francis et al., 2001 and Siddhuraju and Becker, 2003). These workers reported that reduction in anti-nutritional factors by different processing techniques resulted in better palatability and growth in fish. This feeding trial reveals that the maize-basal diet (control diet) performed best in term of growth, food conversion ratio and protein efficiency ratio than diets with varying inclusion levels of sundried water lettuce leaf meal and its inclusion affected the growth patterns of experimental fish. Diets with high inclusion levels (diet 4 and diet 5) resulted in reduced growth of *Clarias gariepinus* juveniles. The other treatments which include water lettuce leaf meal, however, showed incredible results. The fish fed 50% water lettuce leaf meal along with 50% yellow maize meal was observed to be best among the treatments, with final weight of 84.37 g, 1.24 g daily growth rate, 3.10%/day specific growth rate, 1.00 protein efficiency ratio, 7.49 kg/m³ total fish yield and 86.67% survival rate. This result agrees with other works of Fasakin et al., 2001 and Obasa et al., 2013. These workers reported that at above 50%

Table 5: The growth and survival rates of African catfish (*Clarias gariepinus*) fed water lettuce leaf meal based diets.

Treatments	0%	25%	50%	75% WLLM	100% WLLM
	WLLM	WLLM	WLLM		
Mean initial weight (g)	15.05 ^a	14.70 ^a	14.67 ^a	14.51 ^a	14.84 ^a
Mean final weight (g)	86.32 ^a	74.79 ^{ab}	84.37 ^a	58.94 ^{bc}	53.17 ^c
Mean weight gain (g)	70.60 ^a	60.09 ^{ab}	69.70 ^a	44.33 ^{bc}	38.32 ^c
Relative weight gain (%)	449.45 ^a	408.63 ^{ab}	474.98 ^a	303.68 ^{bc}	258.25 ^c
Daily growth rate (g/day)	1.26 ^a	1.07 ^{ab}	1.24 ^a	0.79 ^{bc}	0.68 ^c
Specific growth rate (%/day)	3.02 ^a	2.90 ^{ab}	3.10 ^a	2.48 ^{bc}	2.25 ^c
Feed intake (g)	173.07 ^a	160.71 ^a	172.53 ^a	156.38 ^a	158.60 ^a
Protein fed (%)	69.23 ^a	64.28 ^a	69.02 ^a	62.55 ^a	63.44 ^a
Food conversion ratio (FCR)	2.45 ^b	2.69 ^b	2.53 ^b	3.58 ^{ab}	4.38 ^a
Feed efficiency (FE)	40.81 ^a	37.17 ^{ab}	39.52 ^a	27.93 ^{bc}	22.83 ^c
Protein efficiency ratio (PER)	1.01 ^a	0.93 ^{ab}	1.00 ^a	0.71 ^{bc}	0.61 ^c
Survival rate (%)	66.67 ^b	61.67 ^b	86.67 ^a	58.33 ^b	56.67 ^b
Total fish production (kg/m ³)	5.76 ^{ab}	4.62 ^{bc}	7.49 ^a	3.43 ^{bc}	3.00 ^c
Culture period (days)	56	56	56	56	56
Stocking density	20	20	20	20	20

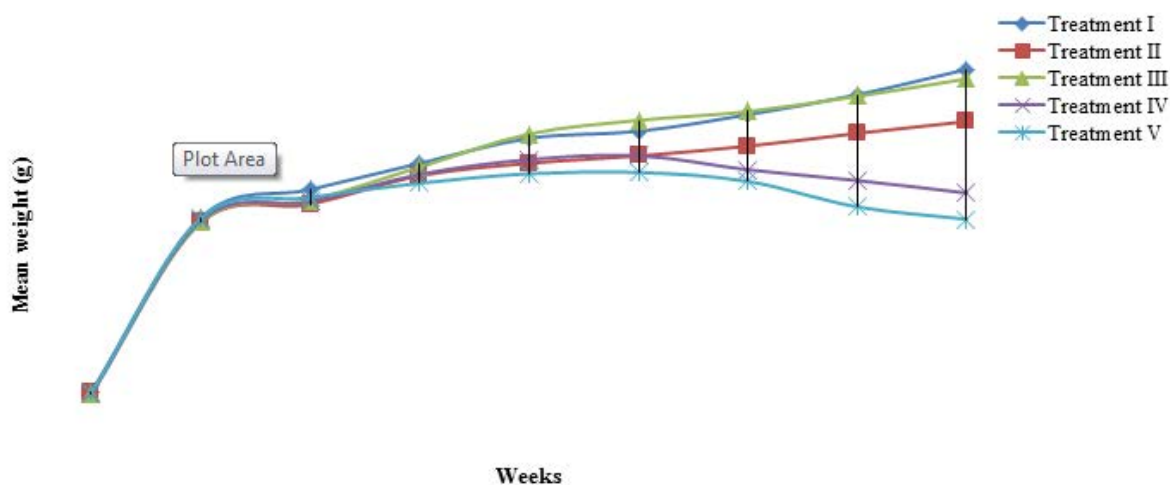


Figure 1: Growth curves of *Clarias gariepinus* fingerlings fed water lettuce based diets for 8 weeks.

inclusion levels, growth, feed utilization and digestibility reduced due to the presence of anti-nutritional factors in *Azolla Africana* leaf meal and fermented mango seed meal respectively. In this present study, inclusion of water lettuce leaf meal at 75% reduced growth rate and feed utilization of *Clarias gariepinus* juveniles. Omojowo et al. (2010) reported that 25% inclusion of mango peel meal in the diet of *O. niloticus* fingerlings supported growth. El-Sayed et al. (2010) reported that dried citrus pulp meal could also replace 10% maize meal for tilapia diets. However, Imgbian and Zarmai, (2011) also reported that palm oil sludge meal could replace maize meal up to 50% inclusion level for *H. longifilis* fingerlings. Throughout the feeding trial, the water quality in all treatments was within the optimum range required by catfish Boyd (1979) and Ajani and Akinwale (2001).

Conclusion

The cost effectiveness in terms of protein-sparing action of diet 3 accounts for its quality for better performance. The practical diet with water lettuce leaf meal could be partially included in maize diets fed to *Clarias gariepinus* juveniles up to 50% to facilitate fish acceptability, ingestion and utilization for better yields in aquaculture.

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