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Effects of dietary protein and Phytase on growth performance and body composition of Nile tilapia, *Oreochromis niloticus* L.

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ABSTRACT

The effects of dietary protein (35%, 40%, 50%) on different growth and Hematological parameters were investigated for Nile tilapia, *Oreochromis niloticus* L. Generally, there was a progressive increase in growth parameters like weight gain, Average daily gain/fish comparing with lower protein content diets without Phytase supplemented. Fish fed on diet containing 50 % of protein supplemented with highest Phytase dose (10.000 unit/kg diet) reflected highest of final weight (69.72 g/ fish). On the other hand, feeding on diets containing 35%, 40 and 45 % of protein supplemented with Phytase reached 46.91, 58.32, 65.81 g/ fish comparing with same diets without adding Phytase 45.88, 57.64, 63.27, 67.22 respectively. Distinguish significant decrease was founded for Feed conversion ratio FCR and Protein efficiency ratio and remarked high dietary protein levels (1.6 and 1.37) comparing with control diet (2.4, 1.62) respectively. Haemoglobin content and Total protein (TSP) reflected significant increasing relation with the increase of protein content in diets. By contrary, Nile tilapia Albumin and Globulin concentrations in serum were changed insignificantly. According to our obtained results, it could be concluded that artificial feed with 45% and 50 % dietary protein supplemented with 7.000 and 10.000 Phytase (unit/kg diet) improve and stimulate many growth performance parameters body composition and hematological features of Nile tilapia *Oreochromis niloticus* (L).

***Keywords*: *Oreochromis niloticus,* dietary protein, Phytase, growth performance**

Introduction

Fish consider high quality food with distinguishable protein and nutrients elements with unique important for human health and growth (Olaifa *et al*., 2010). Globally, aquaculture considers the fastest growing food producing sector and represents an important component in food national security (Ibrahem *et al*., 2010). Recently aquaculture playing an important role in the world fish production. According to Food Administration Organization (FAO), aquaculture considers the fastest growing food production activities in the world (FAO1997). Dogma of aquaculture industry is optimizing growth with efficient economical feed dietary to produce high quality fish (Bello *et al*., 2012). Thus, supplementary feeding plays key roles in aquaculture nutritional and economical success. Tilapia belongs to the Cichlidae remarked with very diversities and widely distribution throughout Africa, South America and other parts of the world. In Egypt, Tilapias represent about 52.8% of the total freshwater fish yield (Gafred, 1996). In the light of this fact, Tilapias considered the main target for Egyptian aquaculture industry. More than half of total production cost in modern intensive aquaculture count for Feed formulations (Ibrahem *et al*., 2010). The aquaculture feed industry depending on the fishmeal, which considers the most effective protein source for fish feed. However, Limited supply, high cost considers important challenges for fishmeal employment (New and Wijkstro¨m, 2002, Baruah *et al*., 2004). Thus, replacement fishmeal with extensively available plant or grain by-products is getting increasing concerns to develop of low-cost fish feed (Carter and Hauler, 2000; Gatlin *et al*., 2007). Recently, Soybean meal is considered the most nutritious and applied as a protein source in different fish diets (Lovell, 1988). Among a large number of feed additives available to improve fish growth performance, enzyme (Phytase) can promote growth through increasing nutrients bioavailability in plant protein. This investigation was conducted to assess the influence for increasing dietary protein level with 30, 35, 40 and 45 % crude protein of Soybean (*Glycine max*) supplemented with3,000 units, 5,000 units, 7,000 units and 10,000 units

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Phytase/kg diet making diet/treatments on *O. niloticus* growth performance. To achieve this goal, different growth (Weight gain (g), % weight gain, Average daily gain/fish, Feed conversion ratio FCR and Protein efficiency ratio) and Hematological parameters (Erythrocyte counts, Haemoglobin content, total protein (TSP), Albumin, Globulin and Albumin / Globulin ratio) will employed.

Materials and methods

Site of work:

The experiments were conducted at first fish Nutrition lab, Animal and Fish Production Department, Faculty of Agriculture Saba Basha, Alexandria University, city-Egypt. During four months from April to July 2018. Experimental aquaria: ten glass aquaria filled with 35-liter volume were filled with dechlorinated tap water for each. Through electric air compressor, water in each aquarium was provided with sufficient aeration. Furthermore, siphoning used daily for removing faeces and other remains before establishment feeding experiments and full cleaning was carried out for each aquarium every day.

Experimental diets

Nine diets were formulated at 30, 35, 40 and 45 %crude protein supplemented with3,000 units, 5,000 units, 7,000 units and 10,000 units Phytase/kg diet making diet/treatments. Each dietary represented with three replicates, with 20 fish each with 5.98 ± 0.1g as standard length. After fish acclimatization for 14 days before the experiment which lasted for 16 weeks, fish were fed at 5% body weight twice daily. Weight variation were noticed and recorded weekly.

Treatment of soybeans and diet formulation

Soybean (*Glycine max*) bought from a local market and processed by heat treatment method. Soybean was dried for 10 hours at 70℃ then ground into fine powder to compose a meal. Then, 5.0 kg soybeans meal was mixed with 5.0 liters of water and dried at 70℃ for 8 hours and reblended and stored at ambient temperature till use. Finally, Phytase (enzymes) were supplemented into each mixture at with 3,000 units, 5,000 units, 7,000 units and 10,000 units Phytase/kg diet. Proximate composition and gross energy of the experimental diets are shown in Table (1).

Table 1: Composition of experimental diets /100 g with three lipid levels.

Ingredients	Diets (% protein)								
	Control	35 with Phytase	35 without Phytase	40 with Phytase	40 without Phytase	45 with Phytase	45 without Phytase	50 with Phytase	50without Phytase
Fish meal	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0
Soybean	25.0	35.0	35.0	40.0	40.0	45.0	45.0	50.0	50.0
Maize yellow corn	59.0	49.0	49.0	44.0	44.0	39.0	39.0	34.0	34.0
Vitamins &minerals mix	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Vegetable oil	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Phytase (unit/kg diet)	0.0	3,000	0	5,000	0	7,000	0	10,000	0
Total	100	100	100	100	100	100	100	100	100

Biological evaluation

For a period of 16week, fish length and weight were measured to nearest 0.1cm and weighed to the nearest 0.1g every 15 days for each aquarium. Ten fishes from each aquarium were randomly collected for whole fish body evaluation. For moisture content estimation, sample was dried at 105° C to constant weight in an oven. Kjeldahl methods was applied to estimate Protein content as N×6.25 and ash by heating at 550° C for 24h in ashing furnace.

The following equation was used to evaluate fish growth performance:

Mean weight gain = [final mean weight (g) – initial mean weight (g)]
Specific growth rate (SGR) in terms of dry weight was calculated according to Xie *et al*., (1997) as follow: SGR=100× (Ln FBW-Ln IBW)/t
Survival percentage of each group was recorded according to Richardson *et al*. (1985).
Feed conversion ratio (FCR) and (PER) were calculated according to Dato-Cajegas and Yakupitiyage (1996) as follow:
FCR = dry feed (g)/ weight gain (g)
PER=wet weight gain (g)/amount of protein fed (g)
Protein productive value (PPV) = (Final fish body protein - initial body protein)/ Crude protein intake × 100

To investigate influence of different protein and Phytase levels, hematological parameters of *Oreochromis nilotocus* was recorded. into heparinized tubes, blood was directly collected from the caudal artery and Sahli haemometer system was applied to estimate Haemoglobin content. In final, Serum protein and cholesterol were measured using standard Kits (Biolabs Laboratory Kits).

Statistical analysis

Resultant data were subjected to one-way analysis of variance (ANOVA) using SPSS (Statistical Package for Social Sciences 2006 version 15.0). Duncan multiple range test was used to evaluate differences among individual means.

Results

Experimental diet Proximate composition before and after the experiment

Nine applied diets reflected significant variation for crude protein content was recorded for and ranged between 25.61 and 49.82 %. Highest crud protein content was remarked diet with 50 % protein comparing with control diet (only showed25.61 % of protein). Ash content, and crude fiber were similar and ranged between 7.81 and 7.53, 5.71 and 5.24 for control and 50% protein content diets respectively. By contrary, varied dry matter content was reflected for different diets comparing with control. The highest value of dry matter was recorded for control diet (91.65). then, arranged dissentingly corresponded to diet with 50% protein content which reflected the lowest dry matter value (79.23) (Table 2).

Table 2: Proximate chemical composition (%) of the experimental diets.

Ingredients	Control	Diets (% protein)			
		35	40	45	50
Dry matter	91.65	88.34	83.84	81.55	79.23
Crude protein	25.61	35.01	40.11	44.97	49.82
Crude fiber	5.71	5.66	5.42	5.27	5.24
Ash	7.81	7.76	7.66	7.61	7.53
DE (kJ g±1) ²	0.064	0.063	0.062	0.61	0.59

-Nitrogen free extract by difference values
- Standard physiological values of 18.8 k j / g protein, 13.8 kj/g carbohydrate and 33.5 kj /g fat was used to estimate Metabolizable energy according to Brett and Groves 1979).
- Gross energy contents (Kcal /kg) according to NRC (1993) using following calorific values 5.64, 9.44 and 4.11 kcal/Kg whole bodies of protein, fat and carbohydrate respectively.

Growth performance parameters for Nile Tilapia fed protein and Phytase diets

As shown in Table (3), different growth performances parameters of *Oreochromis niloticus* were recorded. Generally, diets with increasing protein content supplemented with Phytase improved growth performance parameters comparing with lower protein content diets without Phytase supplemented. Fish fed on diet containing 50 % of protein supplemented with highest Phytase dose (10.000 unit/kg diet) reflected highest of 69.72 g/ fish final weight. On the other hand, feeding on diets containing 35%, 40 and 45 % of protein supplemented with Phytase reached 46.91, 58.32, 65.81 g/ fish comparing with same diets without adding Phytase 45.88, 57.64, 63.27, 67.22 respectively. Fur2thermore, diets statistical evaluation showed that fish fed on diet containing 50 % protein with

10.000 unit/kg diet of Phytase reflected significant (P< 0.05) higher weight followed in a decreasing order (P< 0.05) by the 35, 40 and 40 % protein and the control groups respectively. Similar growth parameters was recorded for % weight gain when fish fed on diet containing 50 % of protein supplemented with highest Phytase dose (10.000 unit/kg diet) recorded the highest value (208.1 %) followed in a significant (P< 0.05) dissentingly order for by the 35, 40 and 45 % protein and the control groups respectively. Significant increasing for Feed conversion ratio (FCR) with decreasing of the protein content in diets with Phytase reached to lowest value of 1.6 in fish fed at 50 % of protein supplemented with highest Phytase dose (10.000 unit/kg diet) comparing with control sample (2.4). Furthermore, protein efficiency ratio (PER) varied between 1.62 and 1.37 for control and 50 % of protein supplemented with highest Phytase dose (10.000 unit/kg diet) diets respectively. Group fed the 50 % of protein supplemented with highest Phytase dose (10.000 unit/kg diet) show higher PER value (P< 0.05) compared to the control and 35, 40 and 45 % protein content diets. However, variation in this trail among the 35 and 40 % of protein with or without Phytase supplement groups, were insignificant. Our investigated results showed that fishes fed diets with Phytase reflected superior mean weight gain, Feed conversion ratio FCR, Protein efficiency ratio than dietary of fish fed group with high protein content without Phytase which indicated positive relation between increasing growth performances parameters and increasing Phytase addition levels in diets.

Table 3: Growth performance parameters of *Oreochromis niloticus* influenced with different protein and Phytase levels.

		Diets (% protein)							
Growth parameters	**Control**	**35 without Phytase**	**35 with Phytase**	**40 without Phytase**	**40 with Phytase**	**45 without Phytase**	**45 with Phytase**	**50 without Phytase**	**50 with Phytase**
No. of fish / aquarium	20	20	20	20	20	20	20	20	20
Initial average weight (g)	22.63	22.63	22.63	22.63	22.63	22.63	22.63	22.63	22.63
Final average weight (g)	41.76	45.88	46.91	57.64	58.32	63.27	65.81	67.22	69.72
Weight gain (g)	19.13	23.25	24.28	32.36	35.69	40.64	43.18	44.59	47.09
% weight gain	84.5	102.7	107.3	143.0	157.7	179.6	190.8	197.0	208.1
Average daily gain/fish	0.17	0.21	0.22	0.29	0.32	0.36	0.38	0.39	0.42
Survival %	100	100	100	100	100	100	100	96	96
Feed conversion ratio FCR	2.4	2.2	2.2	2.0	2.0	1.8	1.8	1.6	1.6
Protein efficiency ratio	1.62	1.61	1.61	1.58	1.58	1.51	1.49	1.37	1.37

Chemical analyses of fish whole body are shown in Table (4). Moisture content differences was not significantly (p>0.05) among the diets with 35 % of protein with or without Phytase supplement groups and control diet (70.33 to 70.55). Superior value of protein content was obtained with fish fed diet with 50 % of protein supplemented with highest Phytase dose (10.000 unit/kg diet) (71.98±0.2) compared to the control diet group. Ash content was not significantly different (p>0.05) among the 35 and 40 % of protein with or without Phytase supplement groups and control diet. However, highest ash content was recorded after feeding on 50 % of protein supplemented with highest Phytase dose (10.000 unit/kg diet) (28.90) compared to control and other diet groups. There were no significant differences (p>0.05) in Crude lipid among the diets with 35 and 40 % of protein with or without Phytase supplement groups and control diet (22.79 to 22.24). While fish fed with 50 % of protein supplemented with highest Phytase dose (10.000 unit/kg diet) showed significant decrease in body protein content (20.11).

Table 4: *Oreochromis niloticus* whole fish body Chemical composition after 4 months rearing period.

Item	Control	Diets (% protein)							
		35 with Phytase	35 without Phytase	40 with Phytase	40 without Phytase	45 with Phytase	45 without Phytase	50 with Phytase	50without Phytase
Moisture	70.33±0.4	70.47±0.1	70.55±0.1	68.72±0.2	68.23±0.2	64.84±0.1	64.66±0.2	60.63±0.2	61.32±0.1
Crude protein	53.12±0.2	55.89±0.3	55.83±0.2	59.24±0.1	59.98±0.1	66.27±0.2	66.45±0.1	71.98±0.2	71.98±0.2
Crude ash	21.52±0.2	21.82±0.1	21.88±0.3	21.72±0.2	25172±0.3	27.64±0.3	27.66±0.2	28.91±0.3	28.90±0.3
Crude lipid	22.79±0.1	22.92±0.2	22.90±0.2	22.10±	22.24±0.1	21.62±0.1	21.76±0.3	20.02±	20.11±0.2

Hematological parameters for Nile Tilapia fed protein and Phytase diets

Influence of different levels of protein and Phytase levels on some hematological parameters of *Oreochromis niloticus* are shown in Table (5) and Figs. (1). Haemoglobin content was increasing significantly (p< 0.05) with the increase of protein content in diets. Lowest Haemoglobin content was recorded for control diet (9.62±1.2 gm/100ml blood) comparing with 50 % of protein supplemented with highest Phytase dose (10.000 unit/kg diet) which reflected highest Haemoglobin content (13.44±0.31100ml blood). Total protein (TSP) values increased significantly (p< 0.01) with increasing protein content in diets (4.92±0.22, 5.22±0.25, 5.20±0.23, 5.67±0.62, 5.62±0.16, 6.35±0.27, 6.11±0.27, 7.00±0.15 and 7.12±0.23 mg /100 ml), for control, 35, 40, 45 and 50 % protein content diets with and without Phytase respectively. On the other hand, Nile tilapia Albumin and Globulin concentrations in serum were changed insignificant trend.

Table 5: Hematological parameters variations of *Oreochromis niloticus* at experiment ending.

Growth parameters	Control	Diets (% protein)							
		35 with Phytase	35 without Phytase	40 with Phytase	40 without Phytase	45 with Phytase	45 without Phytase	50 with Phytase	50 without Phytase
Erythrocyte counts (106 /mm3)	2.380±0.11	2.54±	2.52±0.18	2.67±0.26	2.63±0.71	2.87±0.31	2.84±0.35	3.12±0.15	3.11±0.11
Haemoglobin content (gm/100ml blood)	9.62±1.2	10.33±0.01	10.24±0.12	11.65±12	11.42±0.18	12.45±0.19	12.31±0.22	13.44±0.31	13.23±0.15
Total protein (TSP) (gm/100ml serum)	4.92±0.22	5.22±0.25	5.20±0.23	5.67±0.62	5.62±0.16	6.35±0.27	6.11±0.27	7.00±0.15	7.12±0.23
Albumin (TSA)	0.44±0.18	0.47±0.31	0.46±0.61	0.51±0.26	0.50±0.22	0.62±0.55	0.62±0.41	0.69±0.32	0.67±0.16
Globulin (TSG)	4.39±12	4.89±0.33	4.81±12	5.72±0.31	5.25±0.21	6.21±0.13	6.22±0.23	6.58±0.63	6.53±0.33
Albumin / Globulin ratio	0.10±0.81	0.09±0.32	0.09±0.22	0.09±0.23	0.09±0.26	0.1±0.14	0.1±0.12	0.1±0.18	0.1±0.11

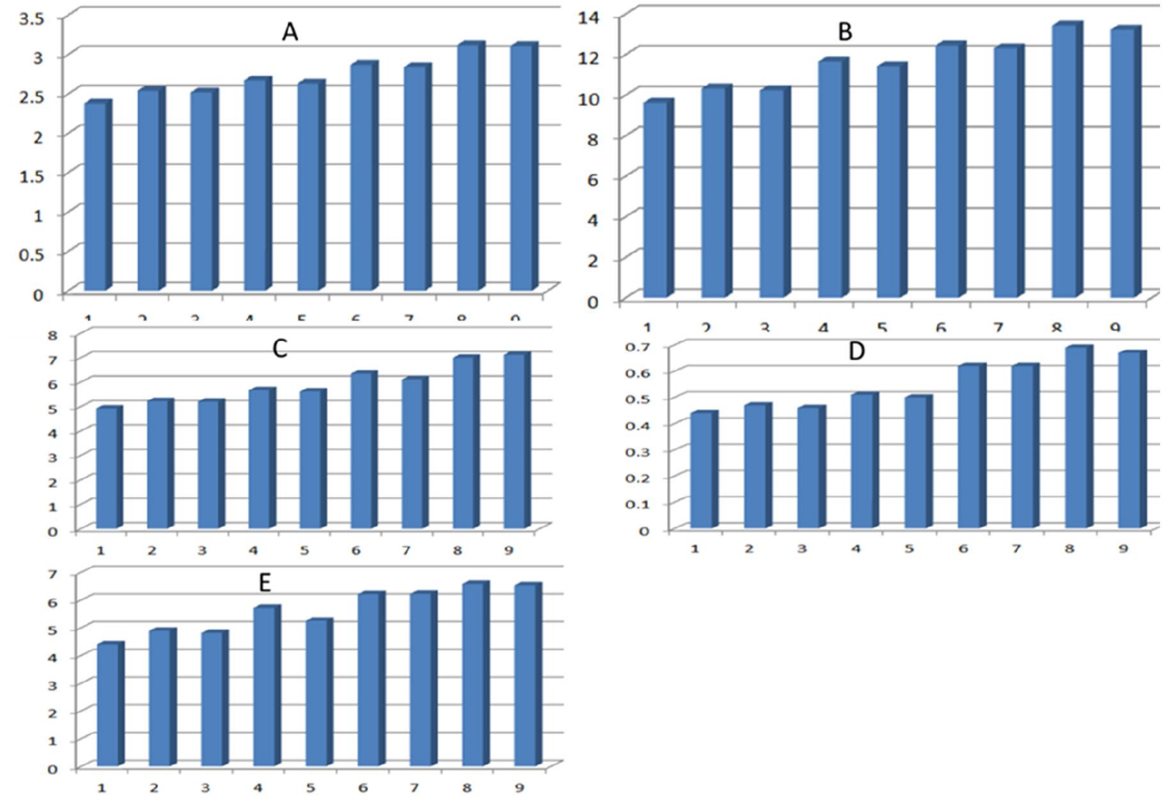

Fig. 1: Changes in (A) Erythrocyte counts, (B) Haemoglobin content, (C) Total protein, (D) Albumin, (E) Globulin of nine diets for *Oreochromis niloticus* at the end of the experiment.

Where:

1- Control.

2- Diet with 35 % of protein with Phytase (3,000 (unit/kg diet))

3- Diet with 35 % of protein without Phytase

4- Diet with 40 % of protein with Phytase (5,000 (unit/kg diet))

5- Diet with 40 % of protein without Phytase

6- Diet with 45 % of protein with Phytase (7,000 (unit/kg diet))

7- Diet with 45 % of protein without Phytase

8- Diet with 50 % of protein with Phytase (10,000 (unit/kg diet))

9- Diet with 50 % of protein without Phytase

Discussion

For all investigated tilapia, progressive increasing was founded for growth performance corresponding with increasing dietary protein level, which clear in light of findings that growth performance may increase with increasing dietary protein up to 45 and 50%, as supported by regression analysis for tilapia fish (Zeitoun, Ullrey and Magee 1976). Our previous findings was in disagreements with most economical dietary protein level for tilapia growth which cleared that growth was increasing for tilapia fry and juvenile fish at 40 % of dietary protein level and 30% for adult fish (96 and 264 g). Other tilapia species, the growth of O. mossambicus (Peters) fry increasing at a dietary protein concentration more than 40% (Jauncey 1982) and above 35% for Tilapia zillii (Gervais) (Mazid, Tanaka, Katayama, Asadur Rahman, Simpson & Cichister 1979).

In general, protein requirement for many fish species including tilapia decreases with increasing size and age (Wilson 1989). Supported with previous data, Balarin and Haller (1982) concluded that tilapia weighing less than 1 g should be fed 35±50% protein, 1-5 g fish 30-40%, and 5-25 g fish 25-30% protein. Our findings for lacking influence of control diet could be cleared in the light of previous findings. More support was added to our obtaining results by El-Sayed and Teshima (1991). They explained that, dietary protein requirement of tilapia fry (< 1 g) is generally high and ranges from 35% to 56%. Furthermore, varied growth parameters for our diets under study were in accordance with Winfree and Stickney (1981) findings. They fund that, different optimized dietary protein levels resulting in maximum growth for tilapia fingerlings (2±7.5 g) have been was 34% for O. aureus and 40% protein diet for O. niloticus fingerlings Gunasekera *et al*., (1995). Effective dietary protein level for our investigation was in agreement with economical dietary protein level for juvenile Nile tilapia growth which found to be higher (40%) than that (30%) (Siddiqui, Howlader and Adam (1988). More support was added to our results by Wee and Tuan (1988). They reported that effective increasing in growth of 25 and 50 g O. niloticus was recorded with increasing dietary protein levels from 20% to 50%. Our findings indicated that Protein efficiency ratio (PER) was decreased with increasing fish weight and dietary protein level which represented as negative relationship with dietary protein level. By contrary of our results, Dabrowski (1979) showed that relation between PER and dietary protein level was varied from species to species. Furthermore, many studies support our findings about Protein efficiency ratio (PER) through clearing that FCR and PER decreasing with increasing dietary protein content (Jauncey (1982) and De Silva, Gunasekera & Atapattu (1989).

Positive corresponding relation between growth performance parameters and increasing Phytase levels in diets could be attributed to two main causes. Firstly, increasing phytate phosphorus (P) liberation from diets via Phytase enzymes, which utilized to gain better growth performance through the fishes. Secondly, increasing bio available and digestible dietary nutrients via the Phytase enzymes. Our obtained results was in accordance with findings of Jackson *et al.,* (1996) who indicated weight gain increasing in channel catfish fed with poor plant protein sources supplemented with Phytase. Our obtained result for applied Phytase as supplemented for fish feed dietary was in agreements with obtained findings by various authors (Cheng and Hardy, 2002 and Zongjia *et al*., 2003). On the other hand, some studies showed no effect of dietary Phytase on weight gain of various fish species specially fed plant-based diets. (Vielma *et al*., 2000, Masumoto *et al*., 2001, Yan and Reigh 2002, Sajjadi and Carter 2004, Yoo *et al*., 2005).

Conclusion

This investigation was carried out to evaluate influence of different dietary protein levels and Phytase on growth performance parameters for Nile tilapia *Oreochromis niloticus* (L). Nine diets were formulated at 30, 35, 40 and 45 % crude protein of Soybean (Glycine max) supplemented with3,000 units, 5,000 units, 7,000 units and 10,000 units Phytase/kg diet making diet/treatments. Diets chemical compositions showed significant variation for crude protein for nine diets and ranged between 25.61 for control diet and 49.82 % for diet with 50 % protein. Ash content, and crude fiber were similar and ranged between 7.81 and 7.53, 5.71 and 5.24 for control and 50% protein content diets respectively. In general, diets with increasing protein content supplemented with Phytase improved growth performance parameters like, weight gain, Average daily gain/fish comparing with lower protein content diets without Phytase supplemented. Furthermore, significant decrease was recorded for Feed conversion ratio FCR and Protein efficiency ratio and remarked high dietary protein levels (1.6 and 1.37) comparing with control diet (2.4, 1.62) respectively.

References

Balarin, J.D. and R.D. Haller, 1982. The intensive culture of tilapia in tanks, raceways and cages. In: Recent Advances in Aquaculture (ed. by J.F. Muir & R.J. Roberts), pp. 265± 356. Croom Helm, London.

Baruah, K., N.P. Sahu, A.K. Pal and D. Debnath, 2004, Dietary Phytase: An ideal approach for a cost effective and low polluting aqua feed. NAGA World Fish Centre Quarterly, 27: 15–19

Bello, O.S., F.E. Olaifa, B.O. Emikpe and S.T. Ogunbanwo 2012, The effect of Walnut (*Tetracarpidium conophorum*) leaf and Onion (*Allium cepa*) bulb residues on the tissue bacteriological changes of Clarias gariepinus juveniles. Bulletin of Animal Heath and Production in African, 60 (2): 205 – 212

Carter C.G and R.C. Hauler, 2000, Fish meal replacement by plant meals in extruded feeds for Atlantic salmon, *Salmo salar* L., Aquaculture 185: 299–311

Cheng Z.J and R.W. Hardy, 2002, Effect of microbial Phytase on apparent nutrient digestibility of barley, canola meal, wheat and wheat middlings, measured in vivo using rainbow trout, Oncorhynchus mykiss. Aquaculture Nutrition 8:271–277

Dabrowski K., 1979. Feeding requirements of ®sh with particular attention to common carp. A review. Polish Archive for Hydrobiology 26, 135±158

Dato-Cajegas, C.R.S. and A. Yakupityage, 1996. The need for dietary mineral supplementation for Nile tilapia *Oreochromis niloticus* cultured in a semiintensive system. Aquacult., 144: 227-237.

De Silva, S.S., R.M.Gunasekera and D. Atapattu, 1989. The dietary protein requirements of young tilapia and an evaluation of the least cost of dietary protein levels. Aquaculture 80, 271±284.

El-Sayed A.M. and S. Teshima, 1991. Tilapia nutrition in aquaculture. Reviews in Aquatic Sciences 5, 247-265.

FAO, 1997. Food and Agriculture Organization ' Aquaculture Production Statistics, 1986-1995. FAO Fish .Cire No. 815 Rev .9Rome, Italy.

Gafred, (Gen. Auth. Fish .Res. Develop. Egypt), 1996 .Annual report for country fish production in 1996.

Gatlin, D.M., F.T. Barrows, P. Brown, K. Dabrowski, G.T. Gaylord, R.W. Hardy, E. Herman, G. Hu, A. Krogdahl, R. Nelson, K. Overturf, M. Rust, W. Sealey, D. Skonberg, E. J. Souza, D. Stone, R. Wilson and E. Wurtele, 2007, Expanding the utilization of sustainable plant products in aqua feeds: A review. Aquaculture Research 38: 551–579

Gunasekera, R.M., K.F. Shim and T.J. Lam, 1995. Effect of dietary protein level on puberty, oocyte growth and egg chemical composition in the tilapia, *Oreochromis niloticus* (L.). Aquaculture 134, 169-183.

Ibrahem, M.D, M. Fathi, S. Mesalhy and A.M. Abd El-Aty, 2010, Effect of dietary supplementation of insulin and vitamin C on the growth, haematology, innate immunity, and resistance of Nile tilapia (*Oreochromis niloticus*), Fish and Shellfish Immunology, 29: 241-24

Jackson, L.S., M.H. Li and E.H. Robinson, 1996, Use of microbial Phytase in channel catfish, *Ictalurus punctatus* diets to improve utilization of phytate phosphorus. Journal of the World Aquaculture Society, 27:309–313

Jauncey, A., 1982. The effect of varying dietary protein level on the growth, food conversion, protein utilization and body composition of juvenile tilapias (Sarotherodon mossambicus). Aquaculture 27, 43-54.

Lovell, R.T., 1988, Use of soybean products in diets for aquaculture species. Journal Aquaculture Product, 2: 27-52

Masumoto, T., B. Tamura and S. Shimeno, 2001, Effects of Phytase on bioavailability of phosphorus in soybean meal-based diets for Japanese flounder *Paralichthys olivaceus*. Fisheries Science, 67: 1075–1080

Mazid, M.A., Y. Tanaka, T. Katayama, M. Asadur Rahman, K.L. Simpson and C.O. Cichister, 1979. Growth response of Tilapia zillii ®ngerlings fed isocaloric diets with variable protein levels. Aquaculture 18, 115±122.

New, M.B and U.N. Wijkstrom, 2002, Use of fishmeal and fish oil in aqua feeds: Further thoughts on the fishmeal Trap, FAO Fish Circ. No. 975. FAO, Rome,

Olaifa, F.E., A.O. Oladapo and O.S. Bello, 2010, Assessment of Performance of *Clarias gariepinus* Juveniles on Diets Supplemented with Kola Pod Husk (*Cola nitida*). In Building a non- oil export based economy for Nigeria: The potential of value – added products from Agricultural residues (Edited by S. O. Jekayinfa). Cuvillier Verlag, Gottingen, Germany, Pp 255- 260.

Richardson, N.I., A.D. Higgs, M.R. Beames and R.J. McBride, 1985. Influence of dietary calcium, phosphorus, zinc and sodium phytate levels on cataract incidence, growth and histopathology in juvenile Chinook salmon, *Oncorhynchus tshayvytscha*. Journal of Nutrition 115:553-567.

Sajjadi, M. and Carter C.G., 2004, Dietary Phytase supplementation and the utilisation of phosphorus by Atlantic salmon, Salmo salar fed a canola-meal-based supplemented with Phytase. Journal of World Aquaculture Society 30:161–173

Siddiqui, A.Q., M.S. Howlader and A.A. Adam, 1988 Effects of dietary protein levels on growth, feed conversion and protein utilization in fry and young Nile tilapia, *Oreochromis niloticus*. Aquaculture 70, 63-73.

Vielma, J., T. Makinen, P. Ekholm and J. Koskela, 2000, Influence of dietary soy and Phytase levels on performance and body composition of large rainbow trout, *Oncorhynchus mykiss* and algal availability of phosphorus load. Aquaculture 183: 349–362

Wee, K.L. and N.A. Tuan, 1988. Effects of dietary protein level on growth and reproduction in Nile tilapia (*Oreochromis niloticus*). In: The Second International Symposium on Tilapia in Aquaculture (ed. by R.S.V. Pullin, T. Bhukasawan, K. Tonguthal & J.L. Maclean), pp. 401-410. ICLARM, Manila, Philippines.

Wilson, R.P., 1989. Protein and amino acid requirements of ®shes. In: Progress in Fish Nutrition (ed. by S. Shiau), pp. 51-76. National Taiwan Ocean University, Keelung, Taiwan.

Winfree, R.A. and A.A. Stickney, 1981. Effects of dietary protein and energy on growth, feed conversion efficiency and body composition of *Tilapia aurea*. Journal of Nutrition 111, 1001-1012.

Yan, W. and R.C. Reigh, 2002, Effects of fungal Phytase on utilization of dietary protein and minerals, and dephosphorylation of phytic acid in the alimentary tract of channel catfish, *Ictalurus punctatus* fed an all-plant-protein diet. Journal of World Aquaculture Society 33:10–22

Yoo, G.Y., Wang X.J., Choi S.M and Han K.M., 2005, Dietary microbial Phytase increased the phosphorus digestibility in juvenile Korean rockfish Sebastes schlegeli fed diets containing soybean meal. Aquaculture 243: 315–322

Zeilton, I.H., D.E. Ullrey and W.T. Magee, 1976. Quantifying nutrient requirements for ®sh. Journal of the Fisheries Research Board of Canada 33, 167-172.

Zongjia, J., Z.J. Cheng and R.W. Hardy, 2003, Effects of extrusion and expelling processing, and microbial Phytase supplementation on apparent digestibility coefficients of nutrients in full-fat soybeans for rainbow trout, *Oncorhynchus mykiss*. Aquaculture, 218: 501–514