Journal of South Valley University for Environmental Researches (JSVUER)
ISSN: 2090-7206
https://portal.svu.edu.eg/svu-src/index.php/jsvues/index

Effect of dietary phytase levels supplementation in improving soybean meal-based diets efficiency for Nile tilapia (Oreochromis niloticus)

Talaat N. Amer; Medhat E.A. Seden and Nader E. El-Tawil*

Department of Fish Nutrition, Central Laboratory for Aquaculture Research (CLAR), Agriculture Research Center, Egypt.

*Corresponding author's e-mail: talaatnagy@yahoo.com

Abstract

The present study was conducted to determine the effect of dietary phytase (PHY) levels (500, 1000, 1500 FTU) supplementation to soybean meal (SBM) based diets on feed utilization and the potential replacement of fishmeal (FM) entirely by SBM in Nile tilapia diets. Five isonitrogenous and isocaloric diets containing 28% crude protein and 425 kcal/100 g diets were formulated. The treatments were: (T_1) positive control diet (FM-based), (T_2) negative control diet (SBM-based), (T_3) SBM-based+ 0.50 g PHY/kg, (T_4) SBM-based+ 1.0 g PHY/kg and (T_5) SBM-based+ 1.50 g PHY/kg. Each diet was offered twice daily to apparent satiation in triplicate fish groups (3.12 g/fish) for 11 weeks. The highest significant means ($p \leq 0.05$) of growth performance and survival rate values and the best values of feed conversion ratio, protein efficiency ratio, protein productive value %, and energy retention% were achieved by fish fed FM-based diet and SBM-based diet supplemented with 1.0 g PHY/kg compared to the other treatments. Insignificant differences ($p > 0.05$) were found in body composition among treatments groups. Results of this study showed that the addition of phytase as mix to soybean meal-based diets at a rate of 1000 FTU/kg can improve the nutrient utilization and growth performance of Nile tilapia. Also, it has an economic return and reduced the feed cost.

Keywords:

Nile tilapia, soybean meal, plant diets, phytase enzyme, growth performance, nutrient utilization.

INTRODUCTION

Aquaculture is recognized as the fastest growing agri-business sector and has thus become an important component of global food supply (FAO, 2014). Expanded aquaculture production will require more fish feed, which will in turn require higher quantities of alternate plant protein sources to reduce the dependency on fishmeal without reducing the performance and provide more economic and environmentally friendly aquaculture (Moustafa and Amer, 2017).

Soybean meal now is considered to be the most promising alternative source of plant protein in compound aquatic feed due to its high protein content, excellent amino acid profile, low cost, availability and steady supply as compared to the other plant protein sources (Castillo and Gatlin, 2015). However, one of the major problems associated with the use of plant proteins in fish feed is the presence of anti-nutritional factors (ANFs), such as phytate " phytic acid " (Amer, 2017).

Phytate (*myo*inositol-1,2,3,4,5,6 -hexaphosphate phosphohydrolase) is an indigestible form of phosphorus, the main storage form of phosphorous (P) in plants cannot be digested and has a low bioavailability for monogastric animals due to absence of an intestinal phytase thereby increasing discharge into aquatic environment (NRC, 2011; Rostami and Giri 2013). In addition, phytate have a negative effect on growth performance, nutrient, protein and energy utilization (El-Tawil, 2015). Phytate also can chelate with other minerals to decrease their bioavailability to fish.

The need for good quality of feed ingredients from plant protein sources with improved nutritional value, economic viability, and growing awareness of the environment has led to a rise in the use of exogenous enzymes in fish diets in recent years (Moustafa and Amer, 2017).

Phytase supplementation can hydrolyze phytate and increase the concentration of minerals like magnesium, calcium, manganese, and zinc in plasma, bone and the whole body (Hassan *et al.*, 2013). Phytase has been used by aqua feed industries to enhance the growth performance, nutrient utilization and bioavailability of macro and micro minerals in fish (Hassan *et al.*, 2013).

The reported results of recent research concerning impact of dietary phytase supplementation on protein and amino acid bioavailability, fish growth performance and plant nutrients utilization in aquaculture diets have been inconsistent (Castillo and Gatlin, 2015; Adeoye *et al.*, 2016). Therefore, the objective of the present study was to investigate the effects of phytase on growth performance, feed utilization of Nile tilapia fed at all-plant based diets.

MATERIALS AND METHODS

The present study was conducted to investigate the feed utilization and growth performance of Nile tilapia fry, *O. niloticus* fed on all-plant based diets supported with gradual levels of phytase enzyme.

Diets formulation and preparation:

Phyzyme® XP 10000 (TPT/L) used in the present work is a powdered micro-granulated phytase enzyme preparation contained 10000 FTU/g produced by Danisco Animal Nutrition, Danisco (UK) Limited, PO Box 777, Marlborough, Wiltshire, SN8 1XN, United Kingdom.

Fifteen glass aquaria were randomly assigned to 5 triplicate treatments. Each treatment attributed to one of the experimental diets. Five isonitrogenous and isocaloric diets were formulated with natural ingredients to provide 28% protein and 425 kcal/100 g diet according to the known nutritional requirements of tilapia (NRC, 2011) (Table 1).

The experimental diets were formulated to be: T_1 contained both herring fishmeal (FM) and soybean meal (SBM) as the main protein source (FM-based) to serve as a positive control. T_2 contained SBM as the main protein source in diet (SBM-based diet) served as a negative control. The last three SBM diets were supplied with 3 gradual levels (500, 1000, 1500 FTU) of the exogenous dietary phytase (PHY) as follow: T_3 contained 0.50 g PHY/kg diet (SBM-based+ PHY_1), T_4 contained 1.00 g PHY/kg diet (SBM-based+ PHY_2) and T_5 contained 1.50 g PHY/kg diet (SBM-based+ PHY_3). The dietary ingredients were homogeneously grounded to 500 µm and thoroughly mixed. Then sufficient amount of water (about 400 ml/kg diet) was added and mixed to obtain stiff dough which was passed through a 1.5 mm die mincer. The pelleted diets were air dried by electric fan at room temperature for 24 hrs.

All diets were packed in sealed plastic bags and kept stored at 4 °C until use. The chemical composition of the experimental diets is presented in Table (2).

Fish and husbandry conditions:

The experiment was performed at the department of fish nutrition, Central Laboratory for Aquaculture Research (CLAR) Abbassa, Abu-Hammad, Sharkiya governorate, Egypt. Nile tilapia (*Oreochromis niloticus*) fry were obtained from CLAR hatchery ponds. Fish were held in an indoor tank and fed the basal diet (T_1) for two weeks as an acclimation period to the laboratory conditions prior to the trial. Fifteen fish with an average initial body weight of (3.12±0.14 g) were weighed and stocked into each of 100 L glass aquaria (3 replicates of 5 treatments). One half of water in each aquarium was changed daily to avoid accumulation of the metabolites. Each aquarium was supplied with an air stone for continuous aeration using an electrical air pump to maintain oxygen level.

All fish were fed to apparent satiation, twice a day, 6 days/week for 11 weeks. During the course of the experiment, all fish were collected from each aquarium every two weeks and collectively weighed.

Sampling, Analytical Procedure and Measurements:

Samples of the experimental diets were taken in a powder form, and preserved in well-sealed plastic bags in a refrigerator for the chemical analysis. Fish were sampled at the beginning and at the end of the trial from each tank, dried and immediately stored at −20 °C pending analyses. Diets and carcass samples were submitted to proximate composition analysis according to the standard methods of AOAC (1990) for moisture, crude protein, total lipids, and ash. Moisture content was estimated by drying the samples at 85°C in a drying oven (GCA, model 18EM, Precision Scientific group, Chicago, Illinois, USA) until a fixed weight was achieved. Crude protein was estimated by multiplying the nitrogen content which was determined using a micro-Kjeldahl apparatus (Labconco Corporation, Kansas, Missouri,USA) by 6.25. Lipid content was determined by petroleum ether extraction in a Soxhlet apparatus (Lab-Line Instruments, Inc., Melrose Park, Illinois,USA) at 40-60 °C for 16 h. Ash was determined by combusting dry samples in a muffle furnace (Thermolyne Corporation, Dubuque, Iowa, USA) at 550 °C for 6 h. Crude fiber was estimated according to Goering and Van Soest (1970) and the nitrogen free extract (NFE) was calculated as:

NFE (%) =100 − (% crude protein + % crude lipid + % crude fiber + % ash). Gross energy was calculated according to NRC (1993).

Water quality analysis:

Routinely, water samples were collected biweekly throughout the experimental period from each aquarium for water quality measurements. Water temperature and dissolved oxygen were measured with an YSI model 58 oxygen meter (Yellow Spring Instrument Co., Yellow Spring, Ohio, USA). While the pH was measured using a pH–meter (Digital Mini-pH Meter, model 55, Fisher Scientific, USA). Unionized ammonia, total alkalinity and total hardness were determined according to Boyd and Tucker (1992).

Growth performance parameters:

The following equations were used to calculate growth performance and survival rate parameters:

Weight gain (WG) = W_1-W_0.

Daily weight gain (DWG) = (W_1-W_0)/T.

Specific growth rate (SGR%/day) = [(Ln W_1− Ln W_0)/T] × 100.

Where, Ln = natural log, W_0 = Initial body weight (g), W_1= Final body weight (g) and T= Time (day).

Survival rate (%) = 100 × (fish No. at the end/ fish No. stocked at the beginning).

Feed utilization parameters:

The following equations were used to calculate the feed utilization parameters:

Feed intake (FI) = total feed consumed over 11 weeks (g)/ fish number.

Feed conversion ratio (FCR) = feed intake (g)/body weight gain (g).

Protein efficiency ratio (PER) = total weight gain (g)/protein intake (g).

Protein productive value (PPV %) = 100 (protein gain/protein intake).

Energy retention (ER %) = 100 (gross energy gain/gross energy intake).

Statistical Analyses:

Data were submitted to one-way ANOVA and were expressed as the mean ± SD of the replicates. All statistical analyses were performed as described by Dytham (1999) using SPSS, version 10 (SPSS Inc. 1999). Significant differences ($p\leq 0.05$) among means were tested by the method of Duncan (1955).

Economical evaluation:

The simple economic analysis was based on local retail sale market price of all the dietary ingredients at the time of the study. These prices were as follows: herring fish meal, 30.00; soybean meal, 8.00; yellow corn meal, 4.00; wheat bran, 3.50; corn oil, 20.00; starch 10.00, vitamins mixture,15; minerals mixture, 7.00 and phytase enzyme, 90 LE/kg.

RESULTS

During the study span, water temperature ranged from 28.3 to 28.9°C, while pH ranged from 7.4 to 7.8. Dissolved oxygen level (DO) was higher than 5.75 mg DO/l, whereas, unionized ammonia concentration was lower than 0.2 mg NH_3/l throughout the study period. Total alkalinity and total hardness values ranged from 123 to 160 mg/l and 160-185 mg/l as $CaCO_3$, respectively. No significant differences ($p>0.05$) were detected among the treatments in water quality parameters.

Growth performance parameters [final body weight (FBW), daily weight gain (DWG) and specific growth rate (SGR %)] of Nile tilapia, *O. niloticus*, fed at different experimental diets over 11 weeks period are presented in Table 3 and Figure 1. FBW, WG and SGR were significantly ($p\leq0.05$) affected by different treatments. The significant highest growth performance values ($p\leq0.05$) were found with fish fed FM-based diet followed with fish fed SBM-based diet supplemented with 1.00 and 1.50 g PHY/kg diet. The differences among fish groups fed at FM-based diet and SBM-based diets supplemented with 1.00 and 1.50 g PHY/kg diet were insignificant ($p>0.05$). While fish fed SBM-diet (negative control) recorded the significant lowest growth performance parameters values ($p\leq0.05$) (Table 4).

High survival rate % was recorded in all the treatments (Table 3). At the end of the experiment, survival rate showed significant differences ($p\leq0.05$) among treatments. The best ($p\leq0.05$) survival rate (88.89%) was observed when fish was maintained at each FM-based diet or SBM-based+1.00 g PHY/kg diet, while the lowest value of survival rate (75.55%) was found with fish maintained at the SBM-based diet (negative control).

The results of feed intake (FI), feed conversion ratio (FCR), protein efficiency ratio (PER), protein productive value (PPV %) and energy retention (ER %) of fish fed diets supplemented with graded levels of the commercial phytase enzyme are shown in Table 4. With regard to FI, results revealed that, the highest FI value (33.84 g) was recorded for fish fed FM-based diet. This which differed significantly from those for the other diets ($p\leq0.05$), followed by fish fed SBM-based+ PHY_2 diet (32.93 g). The significant lowest FI value ($p\leq0.05$) was recorded for fish fed SBM-based diet (T_2) and SBM-based+ PHY_1 without any significant differences ($p>0.05$) between them.

The overall mean FCR values were significantly better (lower) (1.68 and 1.47) with both FM-based diet and SBM-based diet+ PHY_3, respectively than that in the other treatments. Insignificant differences ($p>0.05$) were found between T_1 and T_5 groups. The poorest FCR value ($p\leq0.05$) was determined as 1.78 with fish fed at SBM-based diet (T_2). Concerning the PER, the results showed that the highest PER value (2.11) was obtained in fish fed FM-based diet followed with fish fed SBM-based diet+ PHY_3 and SBM-based diet+ PHY_3 with insignificant differences ($p>0.05$) between them, while the lowest ($p\leq0.05$) PER value (1.99) was recorded in fish maintained at the SBM-based diet (negative control). While the significant highest PPV% value (33.71) was observed in fish fed FM-based diet. This which differed significantly from those for the other SBM-based diets ($p\leq0.05$). No significant differences ($p>0.05$) were found among fish groups fed at SBM-based diets with or without different dietary phytase supplementation levels in PPV%.

With regard to ER% as shown in Table 4, no significant differences ($p>0.05$) were observed in ER values among fish fed FM-based diet and SBM-based diet gropes supplemented with all PHY levels. The highest ER value (22.04%) was recorded with FM-based diet. The lowest ER value (20.84%) was recorded with SBM-based diet (T_2) without phytase supplementation.

The results of the whole body composition of tilapia fed the experimental diets are displayed in Table 5. There were no significant differences ($p>0.05$) in the body moisture (ranged between 74.32% and 74.57%), dry matter (ranged between 25.43% and 25.68%) and protein (ranged between 15.39% and 15.89%) of Nile tilapia fed the dietary treatments. Body lipid content values (ranged between 5.22% and 5.61%). The highest significant values ($p \leq 0.05$) of body lipid were found with fish fed SBM-based diets supplemented with 1.00 and 1.50 g phytase/kg diet. Dry matter content ranged between 15.17% and 17.42%. The highest significant values ($p \leq 0.05$) of body ash were found with fish fed SBM-based diets with or without phytase supplementation, while the lowest value was found with fish maintained at FM-based diet.

Economical efficiency calculations of the SBM-based diets based on cost one kg gain in weight of Nile tilapia in comparison with the FM-based diet are shown in Table (6). It's clear that, PHY supplementation at 1.00 g /kg level in SBM-based diets reduced feed price from 8.91 LE /kg to 7.28 LE/ kg which in turn reduced the feed cost to produce one kg fish gain by 11.57% compared to fish fed control diet (FM-based diet).

DISCUSSION

All values of the water quality parameters measured were within the acceptable range for the normal growth of tilapia as mentioned by Boyd (1984). Moustafa and Amer (2017) showed that supplementation SBM diets with multi-enzyme, Natuzyme® significantly enhanced the apparent retention rate % of both phosphorus and nitrogen by 63.3% and 17.7%, respectively. They concluded that phytase had a positive effect to break down phytate-P in soybean meal which contains 50-80% of phosphate in phytate-P form. So, phytase may be considered as an "environmentally-friendly" additive in plant based diets which in turn subsequently limit or at least postpone the eutrophication problem.

The interest of phytase supplementation in aquaculture research is to escape from anti-nutrient behavior of phytic acid and improve the nutritive value of plant-based diet and also to minimize the excretion of phosphorus to the water environment so as to minimize pollution and eutrophication (Moustafa and Amer, 2017).

In the present study, the response of Nile tilapia fry to plant diets supplemented with exogenous dietary phytase (PHY) was investigated. Nile tilapia as an omnivorous fish able to digest plant material better than carnivorous fish (Amer, 2017). Therefore, soybean meal was expected to be suitable to replace fishmeal in fish diets to reduce the cost of fish feed by the addition of enzyme. However, this study clearly demonstrated that 100% of the FM protein could be replaced by SBM protein in Nile tilapia diets by inclusion of dietary phytase.

Supplementation of SBM-based diets with PHY maintained a similar growth rate as the rate provided by the FM-based diet. This may be attributed to the improvement in protein bioavailability, amino acids absorption and increasing the availability of phytate phosphorous which is an essential nutrient for growth, skeletal development and reproduction in fish in SBM-based diets (Cao *et al.*, 2008). Nile tilapia fed the PHY supplemented diets performed significantly better ($p<0.05$) than those fed control SBM-based diet. These results showed that PHY supplementation in SBM-based diet eliminated the negative effect of phytate in SBM-based diet.

Efficacy of microbial phytase is governed by numerous interactive factors. These may include dietary substrate levels, fish species, the inclusion rate and source of phytase. Cao *et al.* (2007) reviewed that phytase dose at a level of 250 – 2000 FTU/kg feed is usually considered optimum for many fish species.

The results of the present work suggested that the supplementation of PHY (1.00 g/Kg) in SBM-based diets can significantly improve growth and feed utilization parameters of Nile tilapia *O. niloticus*. Similarly, Ng and Chong (2002) showed that phytase enzyme supplementing to tilapia diet containing 40% palm kern meal significantly improved growth and feed efficiency. The higher growth performance parameters (FBW, WG, DWG and SGR %) were observed with fish groups maintained at FM-based diet and SBM-based diet supplemented with PHY_2. Our results were also agreed with Liebert and Portz (2005). They compared two different sources of microbial phytase, SP1002 and Ronozyme@_P in Nile tilapia diet. They found that supplementation of phytase SP1002 at a level of 750 FTU/kg diet was sufficient to improve growth, feed conversion, protein deposition, while supplementation of at least 1000 FTU/kg of Ronozyme@_P resulted in intermediate growth. In contrast, Yigit *et al.*, (2016) informed that the addition of phytase as mix to soybean meal-based diet could not increase growth and nutrient digestibility in trout. The authors attributed the non – response of trout to exogenous enzymes supplementation to a variety of dietary factors such as the concentration and sources of protein in the diet and enzymes which were affected by different temperatures and pH levels.

The positive effect of the dietary phytase on fish growth performance in the present study may be due to its ability to break the bonds between phytate-minerals and phytate-protein in SBM-based diets and increase the availability of amino acids (Amer, 2017). The experimental diets in the present study were not exposed to any heat treatments that may have led to enzymes being highly active. In addition, Hassaan *et al.* (2013) reported that the balance of Ca:P ratio in the experimental diets may improve the bioavailability and utilization of plant phosphorus by fish, bone mineralization and protein digestibility.

The effect of phytase in the bioavailability of nutrients and minerals, protein digestibility, amino acids utilization, growth performance and reduction of anti-nutritional factors of plant-based fish diets have been reported by a certain number of studies (Castillo and Gatlin, 2015; Adeoye *et al.*, 2016; Amer, 2017). It was hypothesized that phytase supplementation can improve protein digestibility and growth performance by improving the activities of endogenous enzymes and the efficacy of the digestion process (Moustafa and Amer, 2017).

The results on fish survival showed better survival rate among those fish fed FM-based diet and SBM-based + PHY_2 with insignificant differences ($p>0.05$) between them. While fish fed SBM-based diet (negative control) achieved the significant lowest ($p>0.05$) survival rate values. The dietary phytase supplementation did not exert any negative effect on the survival rate of tilapia among treatments in the present study. Similarly, Cao *et al.* (2008) reported that inclusion of commercially prepared phytase preparations didn't affect survival rates of Nile tilapia fed SBM-based diets. Also Yigit *et al.* (2018) showed that, there were no differences in the body composition among rainbow trout (*Oncorhynchus mykiss*, Walbaum) groups fed at SBM-based diets supplemented with dietary phytase.

The results showed that feed intake was higher among fish fed FM-based diet. This may be due to the high palatability of FM-based diets more than all-plant diets. While when SBM-based diets were supplemented with 1.00 and 1.50 g PHY/kg, feed intake showed higher levels compared with the control SBM-based diet due to enhanced release of nutrients of plant based diets by breaking down the bonds of phytate-protein and phytate-minerals complexes (Amer, 2017).

It is clear that supplementation of SBM-based diets with PHY positively affected feed efficiency parameters such as FCR, PER, PPV% and ER%. In the present study, PHY supplementation significantly improved feed intake and FCR of *O. niloticus* fry fed on plant based diets. These results agree with those found by several authors who reported that dietary phytase can improve nutrient digestibility of plant-based fish diets and result in better growth performance and feed efficiency (Amer, 2017; El-Tawil, 2015). The enhancement of FCR in fish fed diets containing enzymes in the present work coincided with the increased protein utilization. This is in consistence with the findings of the other studies which evaluated the inclusion of phytase in diet containing high level of plant sources (Amer, 2017; Adeoye *et al.*, 2016).

Phytate can nonselectively bind to proteins and it has been shown to inhibit activities of enzymes including pepsin, trypsin and alpha-amylase (Rostami and Giri 2013) as well as to decrease protein digestibility. The better FCR, PER and PPV% in the fishes fed phytase treated SBM-based diets can be attributed to the enhancement of the protein availability to the fishes and the presence of anti nutritional factors in permissible limit in the test diets.

The body composition of fish is primarily influenced by diet composition, feeding practices, fish size, and can be controlled through nutrition. However, the results of body composition in the present study didn't reveal any significant differences (p>0.05) in moisture, dry matter and protein content when FM was completely replaced by SBM with or without supplementation with phytase. This is in accordance with Amer *et al.* (2015) who concluded that FM can be completely replaced by SBM in Nile tilapia diets by the inclusion of L-carnitine at 300 g/kg without any significant differences in body composition. Also, Amer (2017) didn't find any significant differences in whole body composition of Nile tilapia fry fed diets supplemented with exogenous multi-enzyme preparations. Similar trend was detected by Yigit *et al.*, (2018) who indicated that dietary phytase supplementation in the diets did not have any effect on the whole body composition of rainbow trout (*Oncorhynchus mykiss, Walbaum*) fed soybean meal-based diets. This inconsistency may be attributed to differences in feed ingredients, nutritional quality of plant ingredients, water quality, fish species, size and culture and experimental conditions.

Fishmeal replacement with extensively available plant or grain by-products is getting increased attention for the development of low-cost fish feed. The interest of the feed industry towards dietary enzymes would increase for better protein economy of plant-based aqua feeds (Amer, 2017). The low FCR value obtained with phytase supplemented groups lowered final cost of production than FM-based or SBM-based diet. SBM-based diet supplemented with 1.00 g phytase provides the fish culturist to save 70 g feed per kg body weight gain, which is 5.93% less than the feed required for the SBM-based diet group (negative control). Also, the results clearly showed that inclusion of 1.00 g phytase in SBM-based diet reduced feed cost by 11.57% compared to the FM-based diet. That would save 5.79% of fish culture costs because feeding cost accounts for over 50% of production costs of aquaculture (Amer *et al.*, 2015; Amer, 2017).

Amer (2017) concluded that multi-enzyme preparation supplementation to soybean-based diet for Nile tilapia reduced feed cost by 17.55% compared to the FM-based diet which save 8.78% of fish culture costs. These findings showed that, using multi exogenous enzymes ('cocktails') have more economic return than using purified enzymes as phytase through its success in providing the formulation of cost effective diets.

The present findings suggest that supplementation of commercially available dietary phytase at a rate 1.00 g phytase / kg gives the possibility of total replacement of fishmeal by soybean meal in Nile tilapia diets by improving the utilization of soy bean meal-based diets. The inclusions of phytase in Nile tilapia feed enhance the nutrient utilization and improve fish performance which will have an economic return by reducing the feed cost.

REFERENCES

Adeoye, A.A.; A. Jaramillo-Torres; S.W. Fox; D.L. Merrifield and S.J. Davies, 2016. Supplementation of formulated diets for tilapia (Oreochromis niloticus) with selected exogenous enzymes: Overall performance and effects on intestinal histology and microbiota. Animal Feed Science and Technology, 215: 133–143.

Amer, T.N.; M.E.A. Seden; N.E. El-Tawil and M.H. Ahmad, 2015. Effect of total replacement of fishmeal by soybean meal with different levels of L-carnitine on growth performance and feed efficiency of Nile tilapia (Oreochromis niloticus) diets. 5th Conference of Central Laboratory for Aquaculture Research, Hurghada, 31 March - 3 April 2015 in partnership with Abbassa International Journal for Aquaculture and Egyptian Journal For Aquaculture, Spiceal Issue. 26-43.

Amer T. N. A., 2017: Optimizing all plant-based diets utilization for Nile tilapia (Oreochromis niloticus) using exogenous multi-enzymes mixture (Natuzyme®). Abbassa Int. J. Aqua., 10 (1):40-61.

AOAC., 1990. Official Methods of Analysis of the Association of Official, Analytical Chemists, 15th ed. Association of Official Analytical Chemists, Arlington, VA.

Boyd, C.E., 1984. Water quality in warm water fish ponds. Auburn University, Agriculture Experimental Station, Auburn, AL, USA.

Boyd, C.E. and C.S. Tucker, 1992. Water quality and pond soil analyses for aquaculture. Alabama agricultural experiment station, Auburn university, Alabama, 183 pp.

Cao L., Wang W.,Yang C., Yang Y., Diana J., Yakupitiyage A., Luo Z., Li D., 2007: Application of microbial phytase in fish feed. Enzyme and Microbial Technology 40: 497–507.

Cao L., Yang Y., Wang W. M., Yakupitiyage A., Yuan D. R. Diana J. S., 2008: Effect of pretreatment with microbial phytase on phosphorus utilization and growth performance of Nile tilapia (Oreochromis niloticus). Aquaculture Nutrition 14, 99–109.

Castillo, S. and D.M. Gatlin, 2015. Dietary supplementation of exogenous carbohydrase enzymes in fish nutrition: A review. Aquaculture, 435: 286-292.

Duncan, D.B., 1955. Multiple range and multiple F tests. Biometrics, 11: 1-42.

Dytham, C., 1999. Choosing and using statistics: a biologist's guide. Blackwell Science Ltd., London, UK.

El-Tawil, N.E., 2015. Improving feed efficiency of plant diets by exogenous enzymes additive and heat treatment of soybean for Nile tilapia, Oreochromis niloticus. Egyptian Journal of Nutrition and Feeds, 18 (3): 473- 481.

FAO (Food and Agriculture Organization of the United Nations), 2014. The state of fisheries and aquaculture. Opportunities and challenges. Rome. 223 pp. (Also available at http://www.fao.org /3/a-i3720e.pdf).

Goering, H.K. and P.J. Van Soest, 1970. Forage analyses. Agriculture Handbook 379. Agric. Res. Serv. U.S. Dept. of Agriculture.

Hassaan, M.S.; M.A. Soltan; H.M. Agouz and A.M. Badr, 2013. Influences of calcium/phosphorus ratio on supplemental microbial phytase efficiency for Nile tilapia (Oreochromis niloticus). Egyptian Journal of Aquatic Research, 39: 205–213.

Liebert F, Portz L., 2005. Nutrient utilization of Nile tilapia Oreochromis niloticus fed plant based low phosphorus diets supplemented with graded levels of different sources of microbial phytase. Aquaculture;248:111–9.

Ng W.K., Chong K.K. 2002. The nutritive value of palm kernel and the effect of enzyme supplementation in practical diets for red hybrid tilapia (Oreochromis sp.). Asian Fish Sci. 15:167–176.

NRC (National Research Council), 1993. Nutrient requirements of warm water fishes and shellfishes. The National Academy Press, Washington, D.C.:102p.

NRC (National Research Council), 2011. Nutrient requirements of fish and shrimp. The National Academy Press, Washington, D.C.: 360p.

Rostami H. and Giri A., 2013. An overview on microbial phytase and its biotechnological applications.International Journal of Advanced Biotechnology and Research 4(1):62-71.

SPSS Inc. SPSS Base 10.0 for Windows User's Guide. SPSS Inc; Chicago, IL, USA: 1999.

Y.T.A. Moustafa; T.N. Amer, 2017. Toward more environmentally friendly aquaculture: Effect of dietary exogenous enzymes (Natuzyme®) on nitrogen and phosphorus retention and excretion in Nile tilapia (Oreochromis niloticus). 1st International Conference and Exhibition on Sustainable development of Aquaculture of Central Laboratory for Aquaculture Research In Cooperation with Worldfish, 20- 22 November, 2017. Vol. 2: 478- 501.

Yigit, N.O.; S.B. Koca; B.I. Didinen and I. Diler, 2016. Effect of protease and phytase supplementation on growth performance and nutrient digestibility of rainbow trout (Oncorhynchus mykiss, Walbaum) fed soybean meal-based diets. Journal of Applied Animal Research, 1-4.

Yigit,N.O; S.B. Koca, B. I. Didinen and I. Diler, 2018. Effect of protease and phytase supplementation on growth performance and nutrient digestibility of rainbow trout (Oncorhynchus mykiss, Walbaum) fed soybean meal-based diets. Journal of applied animal research, 1: 29-32.

Table 1. *Composition of different experimental diets used in this experiment.*

Ingredients	FM-based diet T_1	SBM-based diet T_2	SBM-based diet + Phytase (g/kg)		
			T_3 PHY_1 (0.50)	T_4 PHY_2 (1.00)	T_5 PHY_3 (1.50)
Herring Fish meal	10.50	0.00	0.00	0.00	0.00
Soybean meal	35.00	55.00	55.00	55.00	55.00
Wheat bran	18.70	13.00	13.00	13.00	13.00
Yellow corn	26.50	21.00	21.00	21.00	20.00
Corn oil	3.30	5.00	5.00	5.00	5.00
Starch	3.00	3.00	2.95	2.90	2.85
Vitamins premix [1]	1.00	1.00	1.00	1.00	1.00
Minerals premix [2]	2.00	2.00	2.00	2.00	2.00
Phytase (PHY) [3]	0.00	0.00	0.05	0.10	0.15
Total	100.00	100.00	100.00	100.00	100.00

[1]-Vitamins premix (per kg of premix): thiamine, 2.5 g; riboflavin, 2.5 g; pyridoxine, 2.0 g; inositol, 100.0 g; biotin, 0.3 g; pantothenic acid, 100.0 g; folic acid, 0.75 g; para-aminobenzoic acid, 2.5 g; choline, 200.0 g; nicotinic acid, 10.0 g; cyanocobalamine, 0.005 g; α-tocopherol acetate, 20.1 g; menadione, 2.0 g; retinol palmitate, 100,000 IU; cholecalciferol, 500,000 IU.
[2]-Minerals premix (g/kg of premix): $CaHPO_4.2H2O$, 727.2; $MgCO_4.7H2O$, 127.5; KCl 50.0; NaCl, 60.0; $FeC_6H_5O_7.3H_2O$, 25.0; ZnCO3, 5.5; $MnCl_2.4H_2O$, 2.5; $Cu(OAc)_2.H_2O$, 0.785; $CoCl_3..6H2O$, 0.477; $CaIO_3.6H_2O$, 0.295; $CrCl_3.6H_2O$, 0.128; $AlCl_3.6H_2O$, 0.54; Na_2SeO_3, 0.03.
[3]-Phyzyme® XP 10000 (TPT/L): Powdered micro-granulated phytase enzyme preparation containing 10000 FTU/g.

Table 2. *Proximate analysis of experimental diets used in this experiment.*

Proximate analyses%	FM-based diet T_1	SBM-based diet T_2	SBM-based diet + Phytase (g/kg)		
			T_3 PHY_1 (0.50)	T_4 PHY_2 (1.00)	T_5 PHY_3 (1.50)
Dry matter	89.73	89.11	89.25	89.25	89.12
Crude protein	28.19	28.19	28.20	28.19	28.21
Crude fat	7.72	7.35	7.38	7.36	7.32
Crude fiber	6.80	7.43	7.51	7.50	7.54
Ash	9.91	10.00	10.21	10.10	10.12
NFE *	47.38	47.03	46.70	46.85	46.81
GE (kcal/100g)**	425.28	424.73	424.82	424.68	424.85

*NFE is nitrogen free extract and ** GE is gross energy.

Table 3. Growth performance parameters (means ± SE) of Nile tilapia (Oreochromis niloticus) fry fed at different experimental diets.

Items	FM-based diet T_1	SBM-based diet T_2	SBM-based diet + Phytase (g/kg)		
			T_3 PHY_1 (0.50)	T_4 PHY_2 (1.00)	T_5 PHY_3 (1.50)
FBW (g)	23.11±0.03^{a}	20.47±0.27^{d}	21.14±0.0^{c}	22.44±0.28^{ab}	22.16±0.0^{ab}
WG (g)	20.09±0.03^{a}	17.39±0.26^{d}	18.06±0.05^{c}	19.25±0.26^{ab}	19.07±0.06^{ab}
DWG (g)	0.29±0.00^{a}	0.25±0.05^{d}	0.26±0.00^{c}	0.27±0.00^{b}	0.27±0.00^{b}
SGR% /day	2.64±0.00^{a}	2.46±0.02^{d}	2.50±0.02^{cd}	2.53±0.0^{bc}	2.56±0.02^{ab}
Survival rate%	88.89±2.22^{a}	75.55±2.22^{b}	84.45±2.22^{a}	88.89±2.22^{a}	86.67±3.85^{a}

Mean values with the different superscript along the same row are significantly different ($p<0.05$).

Table 4. *Feed and nutrient utilization parameters (means ± SE) of Nile tilapia (Oreochromis niloticus) fry fed at different experimental diets.*

Items	FM-based diet T_1	SBM-based diet T_2	SBM-based diet + Phytase (g/kg)		
			T_3 PHY_1 (0.50)	T_4 PHY_2 (1.00)	T_5 PHY_3 (1.50)
FI (g)	33.84±0.10^{a}	31.00±0.18^{c}	31.00±0.12^{c}	32.93±0.55^{b}	32.54±0.15^{b}
FCR	1.68±0.01^{c}	1.78±0.01^{a}	1.64±0.02^{b}	1.65±0.01^{b}	1.57±0.00^{c}
PER	2.11±0.00^{a}	1.99±0.02^{c}	2.07±0.00^{b}	2.08±0.01^{ab}	2.08±0.00^{ab}
PPV %	33.71±0.12^{a}	31.48±0.55^{b}	31.79±0.44^{b}	31.94±0.84^{b}	31.98±0.35^{b}
ER%	22.04±0.18	20.84±0.23	21.23±0.31	21.29±0.11	21.59±0.28

Mean values with the different superscript along the same row are significantly different ($p<0.05$).

Table 5. *Body composition (Means ± SE) of Nile tilapia (Oreochromis niloticus) fry fed at different experimental diets.*

Treatments	Moisture %	Dry matter%	Protein%	Fat%	Ash%
T_1	74.48±0.12	25.52±0.12	16.04±22	5.23±0.12^{b}	15.17±0.04^{b}
T_2	74.57±0.13	25.43±0.13	15.89±0.24	5.23±0.05^{b}	17.42±0.18^{a}
T_3 (0.50g PHY/kg)	74.46±0.27	25.54±0.27	15.51±0.17	5.22±0.09^{b}	17.28±0.04^{a}
T_4 (1.00g PHY/kg)	74.32±0.18	25.68±0.18	15.39±0.18	5.61±0.11^{a}	16.95±0.40^{a}
T_5 (1.50g PHY/kg)	74.55±0.25	25.45±0.25	15.51±0.12	5.39±0.14^{ab}	16.67±0.27^{a}

Mean values with the different superscript along the same column are significantly different ($p<0.05$).

Table 6. *Economic efficiency for production of one kg gain of Nile tilapia (Oreochromis niloticus) fry fed at different experimental diets.*

Items	FM-based diet T_1	SBM-based diet T_2	SBM-based diet + Phytase (g/kg)		
			T_3 PHY_1 (0.50)	T_4 PHY_2 (1.00)	T_5 PHY_3 (1.50)
Price/ kg feed (L.E)	8.91	7.29	7.28	7.28	7.28
FCR (kg feed/kg gain)	1.58	1.78	1.72	1.71	1.71
Feed cost/kg gain (L.E)	14.08	12.97	12.53	12.46	12.45
Reduction in feed cost/ kg gain (L.E)*	0.00	1.12	1.56	1.63	1.63
Reduction in feed cost/ kg gain%**	0.00	7.93	11.04	11.57	11.57

* Reduction in feed cost per kg gain (L.E.) = feed cost per kg gain of FM-based treatment (L.E.) - feed cost per kg gain of SBM-based treatments (L.E.).

** Reduction in feed cost per kg gain (%) = 100 [Reduction in feed cost per kg gain (L.E.) / feed cost per kg gain of FM-based treatment (L.E.)].

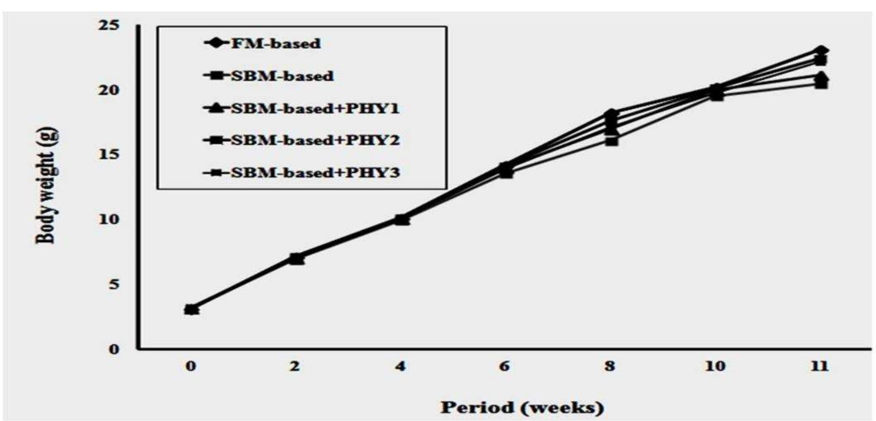

Figure 1. *Changes in mean body weight of Nile tilapia fed fishmeal-based diet and soybean meal-based diets supplemented with phytase (PHY) levels (0.5, 1, 1.5) for 11 weeks.*

Journal of South Valley University for Environmental Researches (JSVUER)
ISSN: 2090-7206
https://portal.svu.edu.eg/svu-src/index.php/jsvues/index

الملخــص العربــي

تأثير إضافة إنزيم الفيتيز على تحسين كفاءة علائق فول الصويا لأسماك البلطى النيلى

طلعـــت ناجـــى علـــى عامـــر ، مدحت السعيد سيدين ، نادر عزت عبد العظيم الطويل

قسم بحوث تغذية الأسماك - المعمل المركزى لبحوث الثروة السمكية - مركز البحوث الزراعية -وزارة الزراعة - مصر.

قسم بحوث تغذية الأسماك- المعمل المركزى لبحوث الثروة السمكية - مركز البحوث الزراعية.

تم إجراء هذه التجربة لدراسة تأثير إضافة مستويات مختلفة (٥٠٠، ١٠٠٠، ١٥٠٠ وحدة دولية) من إنزيم الفيتيز إلى العلائق النباتية المعتمدة على مسحوق فول الصويا كمصدر رئيسى للبروتين على الاستفادة من الغذاء، وكذلك إمكانية الاستبدال الكلى لمسحوق السمك بمسحوق فول الصويا فى علائق البلطى النيلى. تم إعداد خمس علائق متساوية فى محتوى البروتين (٢٨٪) والطاقة (٤٢٥ كيلو كالورى/ ١٠٠ جم غذاء). كانت المعاملات كالتالى: العليقة الأولى احتوت على مسحوق السمك (كنترول ١)، العليقة الثانية (نباتية) احتوت على مسحوق فول الصويا (كنترول ٢)، العليقة الثالثة والرابعة والخامسة (نباتية) مضافاً إليها الفيتيز بمستويات ٠,٥ ، ١,٠ ، ١,٥ جم فيتيز/كجم بالترتيب. غُذيت الأسماك مرتين يومياً إلى حد الإشباع فى ثلاث مكررات من مجموعات الأسماك (متوسط وزن السمكة ٣,١٢ جم) لمدة ١١ أسبوع. أوضحت النتائج أن أعلى قيم ذات دلالة إحصائية لأداء النمو ومعدل الإعاشة و كذلك أفضل قيم لمعدل تحويل للغذاء ونسبة كفاءة البروتين والقيمة الإنتاجية للبروتين وا حتجاز الطاقة تم تسجيلها مع الأسماك التى غُذيت على العليقة المحتوية على مسحوق السمك والعليقة النباتية المضاف إليها ١,٠ جم فيتيز/كجم بالمقارنة مع باقى العلائق. لوحظ أيضاً عدم وجود فروق ذات دلالة إحصائية فى تركيب الجسم بين المعاملات المختلفة. تشير النتائج إلى أن إضافة إنزيم الفيتيز إلى العلائق النباتية المحتوية على مسحوق فول الصويا بمعدل ١٠٠٠ وحدة دولية/كجم يمكن أن يحسن الاستفادة من الغذاء وكذلك أداء نمو أسماك البلطى النيلى. كما أن له عائد اقتصادى وأدى إلى خفض تكلفة الغذاء.