# Effect of Biogen as feed additives on growth and feed utilization of Nile tilapia, *Oreochromis niloticus*

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# Abstract

The present work aimed to investigate the effect of incorporation of increasing levels of Biogen in tilapia diets on growth performance and feed utilization of Nile tilapia (Oreochromis niloticus). The experimental diets were formulated and Biogen was incorporated in five different doses, 0, 0.1, 0.2, 0.3 and 0.4% for the diets D1, D2, D3, D4 and D5, respectively and tested in 15 glass aquaria (3 replicates for each treatment). The experimental period lasted after 90 days. At the end of this experiment (90 days) the highest average body weight (BW) was recorded for fish group fed the diet D2 followed in descending order by those in groups D4, D2, D5 and control group D1 with significant differences between the different diets and the same trend was also observed for body length (BL), weight gain (WG), specific growth rate (SGR), feed conversion ratio (FCR) and protein efficiency ratio (PER). Feed intake during the entire period found to be 28.15, 32.30, 26.33, 27.49 and 26.93 g for the diet enriched by, 0, 1, 2, 3, and 5 g Biogen/kg diet, respectively and the differences between the FI values were significant (P<0.05). Protein content in whole fish body was significantly (P<0.05) decreased with the increasing levels of Biogen and the highest protein content was obtained in fish fed control diet. The lowest EE content and ash content were obtained for fish fed the control diet.

## **1. Introduction**

Biogen product is an effective growth promoter feed additive used in diets of poultry and livestock (EI-Banna *et al*, 2001). The main ingredients of Biogen are ellicien (the product of garlic) + Ginsieng + *Bacillus subtitles* + high unit hydrolytic enzymes (amylolytic, lipolyric, proteolytic and cell separating enzymes).

The high unit hydrolytic enzyme group of Biogen my make the starch, fat and protein of feeds to be entirely dissociated and absorbed in gastrointestinal tracts of the poultry and domestic animals (Yang and Yu, 1990). Nikitine *et al.*, (1995) found that dietary Biogen supplementation improved growth and feed efficiency and it can help in elimination of environmental pollution such as stinking odour. Safinaz (2000) reported that addition of Biogen to fish diet at level of 0.3% improved the normal physiological function of *O. niloticus* exposed to 30 ppm phenol. Moreover, the Biogen minimized the direct and in drastic negative effects of phenol on immune response of exposed *O. niloticus* also it prevents the accumulation of phenol in the muscles.

Biogen can enhance the metabolism of fish body cells, raise the efficiency of feed utilization and balance the secretion of various secretary glands. Moreover, it increases the vitality of cells by supplying oxygen to whole body, improves the immune responses, helps to excrete heavy metals, inhibits aflatoxin and motions the normal endocrine system. Biogen has bactericidal effects and increases the palatability of feed, promotes the secretion of digestive fluids and stimulates the appetite (Abdel-Hamid *et al.*, 2002). Also, Biogen was found to improve the immunity of Nile tilapia fingerlings (Diab *el al.*, 2002). Biogen at level of 2 kg/ton feed can be recommended in fish farming to improve production since it enhances the growth rate of tilapia (Bayoumi, 2004).

Elam (2004) indicated that, supplementation of *O. niloticus* and *Mugil cephalus* by 0.1 or 0.2% Biogen significantly increased serum albumin and globulin indicating that Biogen can

improve the general health and immune system of fish and decreased serum cholesterol and glucose in all treated groups compared to control group. Also allicin enhance the blood circulation in fish body specially in gills resulting in increase the ability of fish to use any little amount of dissolved oxygen. Biogen also contain ginseng extract that needed by the fish body to maintain its physiological functions, and have the ability to enhance the natural body resistance through activation of immune cells. Biogen also have a response as a palatability enhancer with better taste that increase the feed consumption by fish and consequently increase the growth rate and this my be due its content of different kind of enzymes that increase the digestibility and absorbability of feed and activation of the intestinal villi.

## 2. Material and methods

The present study was carried out at the Laboratory of Fish Nutrition, Faculty of Agriculture, Banha University. The aim of this experiment is to investigat the effect of incorporation of increasing levels of Biogen in tilapia diets on growth performance and feed utilization parameters of Nile tilapia (*Oreochromis niloticus*).

Fifteen rectangular aquaria  $100 \times 40 \times 50$ cm (200 liter) were filled by 160 liter freshwater for each aquarium and used for the experiment to represent different 5 experimental treatments (3 replicates) and each aquarium was stocked with 20 fish with an initial weight ranged from 2.18 to 2.25 g. The experimental fish were obtained from Abbassa hatchery, Abbassa village, Abu-Hammad district, Sherkia Governorate, Egypt. The experimental fish were transported in a 50 liter plastic bags filled with freshwater and oxygen to the laboratory, and after arrival to the laboratory fish were stocked in fiberglass tanks for two weeks before start the experiment for acclimatization where all fish were fed the control diet at a rate of approximately 3% of their average body weight to be adapted to pelleted feeds. After the acclimatization the experimental fish were distributed randomly into the experimental aquaria representing the five treatments studied. At stocking, body weight and body length of fingerlings per aquarium were individually recorded.

The aquaria were cleaned and water was replaced every four days, dissolved oxygen was maintained at 3-6 mg/L by continuous aeration (estimated by using dissolved oxygen meter) and water temperature at 23 to 27°C. The basal diets was formulated and Biogen added at five different doses, 0, 0.1, 0.2, 0.3 and 0.4% for the diets D1, D2, D3, D4 and D5, respectively (Table 1). Biogen was supplied from China Way-Taiwan company, a new trade name for probiotic (Khalil *et al.*, 2001), which composed of Allicin (the active principle of garlic), high-unit hydrolytic enzymes (proteolytic lipolytic, amylolytic and cell separating enzymes) *Bacillus subtilis*, and Ginseng extract. Fish fed diets at a daily rate of 3% of total biomass 6 day/week (twice daily at 9.00 am and 3.00 pm). Every two weeks, total fish was taken from each aquarium then weighed and the amount of feed was adjusted according to the changes in body weight throughout the experimental period.

Growth performance parameters were measured by using the following equations: Condition factor (K) =  $(W/L^3) \times 100$  Where, W = weight of fish in "grams" L = total length of fish in "cm"

Specific growth rate (SGR) =  $\frac{\text{LnW2} - \text{LnW1}}{\text{t}} \times 100$ 

Where, Ln = the natural log,  $W_1 =$  first fish weight,  $W_2 =$  the following fish weight in "grams" and t = period in days.

Weight gain = final weight (g) – initial weight (g)

Feed conversion ratio (FCR = Feed ingested (g)/Weight gain (g)

Protein efficiency ratio (PER) = Weight gain (g)/Protein ingested (g)

| Feed ingredients                 |                                  | Experimental diets |        |        |        |  |  |
|----------------------------------|----------------------------------|--------------------|--------|--------|--------|--|--|
|                                  | Diet1                            | Diet2              | Diet3  | Diet4  | Diet5  |  |  |
| Fish meal (65%)                  | 16                               | 16                 | 16     | 16     | 16     |  |  |
| Yellow corn                      | 28                               | 28                 | 28     | 28     | 28     |  |  |
| Soybean meal (40%)               | 40                               | 40                 | 40     | 40     | 40     |  |  |
| Wheat bran                       | 10.5                             | 10.4               | 10.3   | 10.2   | 10.1   |  |  |
| Vegetable oil                    | 2.5                              | 2.5                | 2.5    | 2.5    | 2.5    |  |  |
| Vit. & Min. mixture <sup>1</sup> | 3.0                              | 3.0                | 3.0    | 3.0    | 3.0    |  |  |
| Biogen                           | 0                                | 0.1                | 0.2    | 0.3    | 0.4    |  |  |
| Sum                              | 100                              | 100                | 100    | 100    | 100    |  |  |
| Chemical analysis                | (determined on dry matter basis) |                    |        |        |        |  |  |
| Dry matter (DM)                  | 7.44                             | 6.55               | 6.12   | 7.15   | 5.89   |  |  |
| Crude protein (CP)               | 30.18                            | 30.66              | 30.71  | 30.80  | 30.91  |  |  |
| Ether extract (EE)               | 4.44                             | 4.23               | 4.87   | 4.20   | 4.36   |  |  |
| Crude fiber (CF)                 | 9.33                             | 10.22              | 10.10  | 10.24  | 10.66  |  |  |
| Ash                              | 10.12                            | 10.14              | 10.33  | 10.45  | 10.15  |  |  |
| NFE <sup>2</sup>                 | 45.93                            | 44.75              | 43.99  | 44.31  | 43.92  |  |  |
| ME (Kcal/kg diet) <sup>3</sup>   | 2610                             | 2609               | 2607   | 2600   | 2595   |  |  |
| P/E ratio <sup>4</sup>           | 115.63                           | 117.52             | 117.80 | 118.46 | 119.11 |  |  |

Table (1): Composition and chemical analysis of the experimental diets.

<sup>1</sup> Vitamin & mineral mixture/kg premix : Vitamin D<sub>3</sub>, 0.8 million IU; A, 4.8 million IU; E, 4 g; K, 0.8 g; B1, 0.4 g; Riboflavin, 1.6 g; B6, 0.6 g, B12, 4 mg; Pantothenic acid, 4 g; Nicotinic acid, 8 g; Folic acid, 0.4 g Biotin,20 mg, Mn, 22 g; Zn, 22 g; Fe, 12 g; Cu, 4 g; I, 0.4 g, Selenium, 0.4 g and Co, 4.8 mg.

<sup>2</sup>Nitrogen free extract (NFE) =100-(CP+EE+CF+Ash)

<sup>3</sup>Metabolizable energy was calculated from ingredients based on NRC (1993) values for tilapia.

<sup>4</sup> Protein to energy ratio in mg protein/Kcal ME.

At the end of the experiment, three fish were randomly sampled from each aquarium and subjected to the chemical analysis of whole fish body. Moisture, dry matter (DM), ether extract (EE), crude protein (CP), crude fiber (CF) and ash content of diets and fish were determined according to the methods described in AOAC (1990): dry matter after drying in an oven at 105°C until constant weight; ash content by incineration in a muffle furnace at 600°C for 12 hrs; crude protein (N × 6.25) by the kjeldhal method after acid digestion; and ether extract by petroleum ether (60-80°C) extraction.

Statistical analysis of the obtained data was analyzed according to SAS (1996). Differences between means were tested for significance according to Duncan's multiple rang test as described by Duncan (1955). The following model was used to analyze the obtained data:  $Y_{ii} = \mu + \alpha i + e_{ii}$ 

Where:  $Y_{ij}$  = the observation on the ij<sup>th</sup> fish eaten the i<sup>th</sup> diet;  $\mu$  = overall mean,  $\alpha_i$  = the effect of i<sup>th</sup> diet and  $E_{ij}$  = random error assumed to be independently and randomly distributed (0,  $\delta^2 e$ ).

#### 3. Results and discussion

#### 3.1.Body measurements

At the end of experiment (after 90 days from the experimental start) results of Table (2) indicated that, the highest significant average body weight (BW) was recorded for fish group 3 followed in descending order by those in groups 4, 2, 5 and 1 (control group). Also, the longest BL was recorded in group 4 followed by those in group 3. The succession of decreasing length was observed in group 2 and group 5 and the bottom length was related to group 1 which was fed on basal diet (control). The highest K values (1.87) were recorded by

the fish in both group 1 and 3 which was fed on basal diet and 2 g of Biogen/kg feed followed by those in groups 2, 5 and 4, respectively.

Results of the present study showed that there was a significant increase in BW of Nile tilapia as Biogen level increased till 0.2% then decreased at level of 0.4%. At the same time, all diets containing Biogen levels (0.1 to 0.4%) gave heavy BW than those fed on the control diet.

The growth promoting effect of Biogen which recorded at level of 2 g/kg feed could be attributed to the role of some of the Biogen enzymes (hydrolytic, amylolytic, lipolytic, proteolytic and cell separating enzymes) components which reached at certain level to act as a growth promoter through the increase in digestibility and absorbability of different nutrients in the gastrointestinal tract of fish. *B. subtilis* have been shown to produce digestive enzymes as amylase, protease, lipase which may enrich the concentration of intestinal digestive enzymes or its effect in improving digestive activity by synthesis of vitamins and co-factors or enzymatic improvement (Gatesoup, 1999). Gullian *et al.*, (2004) demonstrated a significant growth increase in shrimp inoculatd with *Bacillus* spp compared with the control. These probiotic effects could be the cause of the increased weight, digestion improvement or nutrient absorption.

Moreover, the growth stimulating effect may be also attributed to the various components of Biogen, where allicin has anti- microbial properties (the product of garlic) and ginseng which could have a growth promoting ability via prevention and treatment of sub- clinical infections, the same findings were observed by Galal *et al.*, (1997). Probiotics are also sometimes expected to have a direct growth promoting effect on fish either by a direct involvement in nutrient uptake, or by providing nutrients or vitamins (Ringo and Gatesoupe, 1998). Also, Khattab *et al.*, (2004) demonstrated that, final body weight of Nile tilapia *O. niloticus* was significantly (P<0.05) increased with increasing Biogen level in the diet and the highest final BW was recorded at 0.1% Biogen level.

| Diets               | No.  | Body wei   | ight/g Body leng |         | igth/cm | Conditic | Condition factor |  |
|---------------------|------|------------|------------------|---------|---------|----------|------------------|--|
|                     | INO. | Initial    | Final            | Initial | Final   | Initial  | Final            |  |
| Diet1 (Control)     | 60   | 2.24       | 9.46 c           | 5.15    | 7.98 c  | 1.63     | 1.87             |  |
| Diet2 (0.1% Biogen) | 60   | 2.23       | 11.58 b          | 5.07    | 8.57 ab | 1.62     | 1.84             |  |
| Diet3 (0.2% Biogen) | 60   | 2.18       | 13.11 a          | 4.97    | 8.89 a  | 1.76     | 1.87             |  |
| Diet4 (0.3% Biogen) | 60   | 2.25       | 12.49 ab         | 5.01    | 8.90 a  | 1.82     | 1.77             |  |
| Diet5 (0.4% Biogen) | 60   | 2.24       | 10.39 c          | 4.95    | 8.31 bc | 1.86     | 1.81             |  |
| Standard error      |      | $\pm 0.07$ | $\pm 0.40$       | ±0.09   | ±0.12   | ±0.15    | $\pm 0.06$       |  |
| .1. 1 1             | 0.11 | 11 1.00    | . 1              | 1       | 1:00    |          |                  |  |

Table (2): The effect of increasing levels of Biogen in the diets on body weight (BW), body length (BL) and condition factor (K) of Nile tilapia.

Averages within each column followed by different letters are significantly different (P<0.05)

Elam (2004) found that live BW of Nile tilapia, *O. niolticus* and *Mugil cephalus* significantly improved when the experimental diets enriched by 2 kg Biogen/ton. Samra (2006) incorporated Azolla, blue green algae or Biogen in tilapia diets at a rate of 10, 10 and 2 kg/ton. He found that the greatest final body weights were achieved for fish group fed artificial feed enriched by Biogen.

In recent study Saleh (2007) incorporated Biogen in Shrimp, *Penaeus japonicus* diets at levels of 0, 0.1, 0.2, 0.3% and he found that, at the 1<sup>th</sup>, 2<sup>th</sup>, 3<sup>th</sup>, 4<sup>th</sup>, 5<sup>th</sup> and 6<sup>th</sup> weeks from the experimental start, BW and survival rate significantly increased as Biogen level increased from 0 to 0.1% then it decreased at level of 0.3%.

## 3.2. Body weight gain (WG) and Specific growth rate (SGR)

The highest average body weight gain (WG) was recorded in group 3 which fed on diet supplemented by 2 g of Biogen/kg feed followed in a descending order by those in groups 4,

2, 5 and group 1 which was fed on basal diet (control diet) and the same trend was also observed for specific growth rate (SGR).

Final BW and WG of Nile tilapia fed diets contained the different Biogen levels were significantly (P<0.001) higher than those fed the basal diet. Similar results were obtained for *P. monodon* (Rengpipat *et al.*, 1998 and 2000). Elam (2004) found that WG of Nile tilapia, *O. niolticus* and *Mugil cephalus* significantly improved (compared to control) when the experimental diets enriched by 2 kg Biogen/ton. In the same trend, Khattab *et al.*, (2004) demonstrated that, WG and SGR of Nile tilapia *O. niloticus* were significantly (P<0.05) increased with increasing Biogen level in the diet and the highest WG was recorded at 0.1% Biogen level. Also, Bayoumi 2004) reported that fish fed artificial diet supplemented with 2g of Biogen/kg diet exhibited highest SGR.

The improvement in WG and SGR of fish in group 3 due to the growth promoter effect of Biogen may be attributed to the fact that Biogen has a particular good flavor and appetizing function which can increase the palatability of feed, promote the secretion of digestive fluids and stimulate the appetite, also to the mode of action of probiotics (as one of Biogen components) which may operate by producing antibiotic substances and inhibiting harmful bacteria altering microbial metabolism and decrease intestinal pH (Bayoumi, 2004). Also the enhancement in body weight gain as a result of other various components of Biogen such as allicin, which is one of the garlic by-product which stimulated growth because of its thyroid like activity (El-Nawawy 1991).

| specific growth rate (SGR) of Nile tilapia. |                        |            |            |  |  |  |
|---|------------------------|------------|------------|--|--|--|
|   | Diets No. <sup>+</sup> | WG         | SGR        |  |  |  |
| Diet1 (Control)                             | 3                      | 7.23 e     | 1.60 e     |  |  |  |
| Diet2 (0.1% Biogen)                         | 3                      | 9.35 c     | 1.83 c     |  |  |  |
| Diet3 (0.2% Biogen)                         | 3                      | 10.90 a    | 1.98 a     |  |  |  |
| Diet4 (0.3% Biogen)                         | 3                      | 10.23 b    | 1.90 b     |  |  |  |
| Diet5 (0.4% Biogen)                         | 3                      | 8.15 d     | 1.71 d     |  |  |  |
| Standard error                              | 3                      | $\pm 0.08$ | $\pm 0.01$ |  |  |  |

Table (3): The effect of increasing levels of Biogen in the diets on weight gain (WG) and specific growth rate (SGR) of Nile tilapia.

Averages within each column followed by different letters are significantly different (P<0.05).

+ Average of 3 replicates (aquaria)

Lun et al. (1994) reported that allicin can activate and coordinate the function of various endocrine glands and thus enables them to secrete horomones especially the growth hormone, also allicin decrease the level of uric acid in fish feces resulting in the decreases in the level of ammonia in the water leading to a good water quality suitable for better growth rate. In addition, Biogen increase the resistance to the bad environmental condition and water pollution..

In the present study, probiotic bacteria of *B. subtilis* that present in Biogen increased as the Biogen dose increased. *B. subtilis* is currently being used for oral bacteriotherapy and bacterioprophylaxis of gastrointestinal disorders (mostly as a direct result of antibiotic treatment). Ingestion of significant quantities of *B. subtilis* is thought to restore the normal microbial flora following extensive antibiotic use of illness (Green *et al.*, 1999). In addition, Moriarty (1998) stated that use of *Bascillus* has been promoted and accepted within the industry due to it has not been associated with aquatic organism pathogies. The present study confirms the above findings. Gatesoupe (1999) reported that probiotic treatment decreased the proportion of pathogenic *Vibrino* spp in sediments and to a lesser extent in the water.

Probiotic actively inhibit the colonization of potential pathogens in the digestive tract by antibiosis or by competition for nutrients and space, and alteration of the microbial metabolism. It also improves the nutrition by detoxifying the potentially harmful compounds in feeds by denaturing the potentially indigestible components in the diet by hydrolytic enzymes including amylases and proteases, by producing vitamins such as biotin and Vitamin B12 by producing inhibitory compounds (Spanggaard *et al.*, 2001) and by stimulating the immunity (Fuller, 1992 and Gibson *et al.*, 1997). Another possible explanation for increased growth performance with adding probiotic is the improvement in digestibility, which may in turn explain the better growth and feed efficiency observed with the supplemented diets. Otherwise, probiotic influence digestive processes by enhancing the population of beneficial microorganisms, microbial enzyme activity; improving the intestinal microbial balance, consequently improving the digestibility and absorption of food and feed utilization (Bomba, *et al.*, 2002).

Mehrim (2001) found that use of 0.3% Biogen in Nile tilapia diet was significantly increased SGR compared to diets free Biogen. These results agreed with the present study which found that 0.2 - 0.3% of Biogen was the optimum addition level for Nile tilapia fry.

## 3.3. Feed Intake (FI)

The highest FI was recorded for the lowest Biogen level (1 g/kg diet) whereas the lowest (P<0.05) FI (26.33 g) was recorded by the fish in group 3 which was fed on diet supplemented with 2 g of Biogen/kg feed (Table 4). Although the lowest Biogen level (1 g/kg diet) caused the highest FI, the other increased Biogen levels 2, 3 or 4 g Biogen/kg diet in diets D3, D4 and D5 did not significantly altered feed intake compared to the control group.

The obtained results indicated that FI of Nile tilapia was significantly affected by Biogen compounds (garlic, ginger, *B. subtilis* and digestive enzymes). Results showed that Nile tilapia fry fed the diet containing 0.1% Biogen consumed more feed than those fed the other experimental diets and this may be due to increasing the palatability of diets containing Biogen by Nile tilapia through its attractive fragrant. Similar results were obtained by Saleh (2007) who found that incorporation of Biogen in the diets of *P. japonicus* and *P. semiselicatus* increased feed intake compared to control diet. This may be show the possibility that Biogen plays an important role in the exploratory and feeding behavior of Nile tilapia. This may be due to its digestive and stimulant effect (Garland, 1993), as well as fragrant and volatile oil (Abo zeid, 1998), controlling and buffering the conditions of the stomach and intestine (Boulos, 1983).

| on feed intake, feed conversion ratio and protein enciency ratio of whe thapia. |                  |            |         |        |  |  |
|---|------------------|------------|---------|--------|--|--|
| Diets   | No. <sup>+</sup> | FI         | FCR     | PER    |  |  |
| Diet1 (Control)   | 3                | 28.15 b    | 3.91 a  | 0.86 b |  |  |
| Diet2 (0.1% Biogen)   | 3                | 32.30 a    | 3.46 a  | 0.97 b |  |  |
| Diet3 (0.2% Biogen)   | 3                | 26.33 b    | 2.42 c  | 1.39 a |  |  |
| Diet4 (0.3% Biogen)   | 3                | 27.49 b    | 2.71 bc | 1.24 a |  |  |
| Diet5 (0.4% Biogen)   | 3                | 26.93 b    | 3.30 ab | 1.01 b |  |  |
| Standard error  |                  | $\pm 1.05$ | ±0.16   | 1.01 b |  |  |

Table (4): Means and standard error for the effect of increasing levels of Biogen in the diets on feed intake, feed conversion ratio and protein efficiency ratio of Nile tilapia.

Averages within each column followed by different letters are significantly different (P < 0.05) + Average of three replicates (aquaria)

#### 3.4. Feed conversion ratios (FCR)

FCR was improved with increasing Biogen level in the diet up to 2 g Biogen/kg diet whereas the highest level of Biogen 4 g Biogen/kg diet re-increased feed required for the production of the same unit of WG. In the same respect Elam (2004) found that, FCR of Nile tilapia *O. niloticus* and *Mugil cephalus* was improved when Biogen was supplemented to diets. The author indicated that, FCR for *O. niloticus* were, 2.8, 2.5 and 2.0, and were 2.4, 2.1 and 3.0, for *Mugil cephalus* fed diets contained 0, 0.1 and 0.2% Biogen, respectively.

The values of FCR recorded from the present study in group 3 and group 4 were in agreement with the value of 1.87 and 2.95 reported respectively for Nile tilapia by Bayoumi (2004). Recently, Saleh (2007) found that FCR and feed efficiency ratio for *Penaeus japonicus* was the lowest for postlarvae groups fed 0% (control diet) than those fed the experimental diets contained 0.1, 0.2 or 0.3% Biogen. Abd El-Hamied *et al.*, (2002) reported that Biogen can enhance the metabolism and energy of fish body cells. Raise the efficiency of feed utilization and balance the secretion of various secretary glands. Moreover, it increases the immune responses.

The best FCR values observed with probiotic supplemented diets suggest that addition of probiotic improved feed utilization. Similar results have been reported for probiotic use in diets of tilapia (El-Haroun *et al.*, 2006). In practical terms, this means that Biogen use can decrease the amount of feed necessary for fish growth, which could result in reduction of production costs. In addition to the above mentioned modality of action, the improvement in growth performance parameters and feed conversion ratio, due to Biogen addition may be explained on the basis that it supply the fish intestine by *Bacillus subtilis*, the pH shift of the intestine would increase the growth rate of these beneficial commercial bacterium which is also a component of Biogen.

# 3.5. Protein efficiency ratio (PER)

Supplementation of tilapia diet by each of the examined levels 1, 2, 3, and 4 g Biogen/kg diet significantly (P<0.001) improved PER specially fish groups fed the diets enriched by 2 or 3 g Biogen/kg diet. The benefit effect achieved by using Biogen may be attributed to the presence of enzymes as one of the Biogen components which assisting in the utilization of proteins.

Results of PER indicated that supplementing diets with Biogen significantly improves dietary protein. This contributes to an optimized protein use for growth of Nile tilapia. Ringo and Gatesoupe (1998) and Saleh (2007) showed similar improvement in the biological value of the diets supplemented with probiotics. Flores *et al.*, (2003) observed a similar trend where the addition of 0.1% probiotics in tilapia fry diets improved growth and apparent protein digestibility and mitigated the effects of stress factors.

From nutritional point of view and in agreement with the data of Shelby *et al.*, (2006) and Saleh (2007), the present results revealed that the use of the probiotic Biogen as a feed additive for Nile tilapia is recommended to stimulate productive growth performance and nutrient utilization (FCR and PER). Feed utilization was the highest in Nile tilapia fed the Biogen supplemented diets, meaning that the nutrients were more efficiently used for growth performance.

#### 3.6. Chemical composition of fish

Results of Table (5) indicated that, fish group fed control diet had the lowest dry matter content (DM) compared to the other groups. Crude protein content in whole fish body was significantly (P<0.05) decreased with the increasing levels of Biogen (50.39, 44.48, 47.54 and 48.72% with 0.1, 0.2, 0.3 and 0.4% Biogen level, respectively) and the highest protein content was obtained in control group (55.45%).

With respect to ether extract, results of Table (5) indicated that values of ether extract lie in two groups, the first group include fish fed the first and the second diets (Diet 1 and Diets 2) and the second group included fish fed the diets D3, D4 and D5. Statistical analysis indicated that the differences between the two groups were significant while the differences between diets within each group were not significant and the indicated that the low Biogen level (0.1%) in the diet did not affected the ether extract content of whole fish compared to fish fed the basal diet while the other levels of Biogen (0.2, 0.3 or 0.4%) significantly increased the ether extract of whole fish. The lowest EE content was obtained in the control fish group (34.29%). Ash content in the whole fish body was significantly increased with diets containing 0.2 and 0.4% Biogen (15.05 and 13.90%, respectively) where the least one was observed in control group (10.14%).

Soltan et al.

The obtained results did not in agreement with those obtained by Mehrim (2001) and Abdelhamied *et al.*, (2002) who found that the dietary inclusion of Biogen increased the protein content of fish and lowered the fat content of the whole fish body, without significant differences in ash content of Nile tilapia. Also, Khattab *et al.*, (2004) found that, protein in whole fish body was significantly increased and fat and ash contents of whole fish body weight of Nile tilapia were decreased with Biogen levels 0.1, 0.2 and 0.3%.

Table (5): Means and standard error for the effect of increasing levels of Biogen in the diets on chemical composition of Nile tilapia.

|                     | p = 2= |           |          |             |          |
|---------------------|--------|-----------|----------|-------------|----------|
| Diets               | No.    | DM        | СР       | EE          | Ash      |
| Diet1 (Control)     | 9      | 29.49 b   | 55.45 a  | 34.29 b     | 10.14 b  |
| Diet2 (0.1% Biogen) | 9      | 32.21 a   | 50.39 ab | 35.53 b     | 12.26 ab |
| Diet3 (0.2% Biogen) | 9      | 30.50 ab  | 44.48 b  | 38.08 a     | 15.05 a  |
| Diet4 (0.3% Biogen) | 9      | 31.41 ab  | 47.54 b  | 38.82 a     | 12.16 ab |
| Diet5 (0.4% Biogen) | 9      | 30.05 b   | 48.72 b  | 37.36 a     | 13.90 b  |
| Standard error      |        | ±0.63     | ±2.23    | ±1.34       | ±0.96    |
|                     | 0.11 1 | 1 11 00 1 |          | 1 11:00 (7) | 0 0 =    |

Averages within each column followed by different letters are significantly different (P<0.05)

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