

EFFECT OF DIETARY SUPPLEMENTATION WITH BIOMOS® OR T-PROTPHYT 2000 WITH AND WITHOUT HORMONE TREATMENT ON PERFORMANCE, CHEMICAL COMPOSITION, AND HORMONE RESIDUES OF MONO-SEX NILE TILAPIA

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ABSTRACT

Two experiments were carried out. In both experiments, Nile tilapia fry (0.1 g initial bodyweight) were stocked into glass aquaria at 100 fry / aquarium in duplicate aquaria / treatment. The experimental diets were offered daily at a feeding rate of 10 % of the fish biomass for 21 days (hormone-treated diets as commercially done for sex reverse) followed by 8 or 12 weeks as experimental periods in the 1st and the 2nd experiments, respectively. Bio-Mos® and T-Protphyt 2000 were added to the 1st and the 2nd experiments, respectively at graded levels. The results obtained revealed no significant ($P \geq 0.05$) differences among treatments in both experiments concerning growth performance and chemical composition of the fish (except final length and fat % in both experiments, ash % in the 1st experiment and dry matter % in the 2nd experiment). However, the diet containing testosterone alone in the 1st experiment (T₄) and that containing T-Protphyt 2000 at 4 g / Kg diet in the 2nd experiment reflected the best growth performance. The significant ($P \leq 0.05$) values in both experiments concentrated mainly in water holding capacity (WHC, where all treatments were higher than the control) and testosterone (the control was the highest). There were positive as well as negative correlations between the physico-chemical characteristics of the fish carcasses. In conclusion, we could recommend the use of 17 α -methyl testosterone at 60 mg / Kg diet for sex reverse during the early stage after the hatch and to increase the growth and survival besides decreasing fat content without any danger from the hormone residues. The dietary inclusion of Bio-Mos® was not beneficial, whereas the dietary inclusion of T-Protphyt 2000 at 4 g / Kg diet was positively effective in increasing the growth performance of the tilapia fry.

Keywords: Mono-sex Nile tilapia – Prebiotic – Testosterone – Growth.

INTRODUCTION

Probiotics and immunostimulants are good candidates to replace the use of chemicals and antibiotics in the development of sustainable shrimp farming systems. *Bacillus spp.* appears to be good probionts as well as showing potential as immunostimulants. The ongoing search for new probiotic strains and further experimentation on interactions between immunostimulant and shrimp immunity will shed more light on how to control diseases in shrimp culture (Rengpipat, 2005). The metabolic effects of the prebiotics includes the production of short – chain fatty acids, fat metabolism, and absorption of ions (Ca, Fe, Mg), besides enhancing the immunity. The increase in colonic bifidobacteria has been assumed to benefit health by producing compounds to inhibit potential pathogens, by reducing blood ammonia levels, and by

producing vitamins and digestive enzymes (Staykov *et al.*, 2005). Prebiotics are involving in the areas for the development of functional foods lies in the use of ingredients to modify the composition and metabolic activity of the gut microflora, i.e. gastrointestinal health (Allam, 2012). Therefore, the present study aimed to evaluate the effect of using the prebiotics "Bio-Mos®" and T-Protphyt 2000 in diets of Nile tilapia during hormone –sex-reversal period and 8-12 weeks thereafter on their growth, chemical composition, and testosterone residues.

MATERIALS AND METHODS

Experimental fish and management:

Newly hatched Nile tilapia (*Oreochromis niloticus*) fry (0.001 g initial body weight and 0.9 cm initial body length) were obtained from Dr. S. M. Ibrahim Hatchery at Tolombat 7, Kafr El-Sheikh governorate. The fry were transported to the wet lab. of the Experiments Station in El-Serw, National Institute of Oceanography and Fisheries, Inland Branch Water. The fry were stocked into glass aquaria (70 X 40 X 30 cm), each containing 50 l water and provided with an air stone connected with an electric compressor at 100 fry/ aquarium in duplicate aquaria/ treatment. Thereafter, the experimental diets were offered daily at a feeding rate of 10% of the fish biomass for 21 days (as commercially done for sex reverse) followed by 8-12 weeks as experimental period.

The experimental treatments (T):

The 1st experiment:

- 1- T₁: Control without any additives.
- 2- T₂: T₁+ Bio-Mos® at 2 g / Kg diet.
- 3- T₃: T₁+ Bio-Mos® at 4 g / Kg diet.
- 4- T₄: T₁+ 17 α-methyl testosterone at 60 mg / Kg diet.
- 5- T₅: T₁+ Bio-Mos® at 2 g / Kg diet + 17 α-methyl testosterone at 60 mg / Kg diet.
- 6- T₆: T₁+ Bio-Mos® at 4 g / Kg diet + 17 α-methyl testosterone at 60 mg / Kg diet.

The 2nd experiment:

- 1- T₁: Control without any additives.
- 2- T₂: T₁+ T-Protphyt 2000 at 2 g / Kg diet.
- 3- T₃: T₁+ T-Protphyt 2000 at 4 g / Kg diet.
- 4- T₄: T₁+ 17 α-methyl testosterone at 60 mg / Kg diet.
- 5- T₅: T₁+ T-Protphyt 2000 at 2 g / Kg diet + 17 α-methyl testosterone at 60 mg / Kg diet.
- 6- T₆: T₁+ T-Protphyt 2000 at 4 g / Kg diet + 17 α-methyl testosterone at 60 mg / Kg diet.

The experimental diets:

The basal diet for fish (not ≤ 25% crude protein, not ≤ 4.5% crude fat, not ≤ 3924 Kcal gross energy / Kg, not ≥ 7.1% crude fibers, from Al'Ekhawh Company – Balteem - Kafr El-Shiekh, consists of soybean meal, yellow corn, wheat bran, molasses, imported fish meal 65%, lime stone, common salt,

dicalcium phosphate, minerals mixture, and rice bran), the hormone (17 alpha methyl testosterone, 10g package, from Argent Laboratories, Inc., 235 Sacedo St., Legaspi Village, Makati City, Philippines, for aquaculture use only) and the tested prebiotics were obtained from the local market. The fish were initially weighed and total length was measured then reweighed and remeasured after the 21 days and the 8-12 weeks for both experiments, respectively.

Growth measurementd:

$$TWG = \square W_1 - W_0$$

Where:

W_1 = Average final weight

W_0 = Average initial weight

$$SGR (\%/d) = (\ln WT - \ln Wt) / (T-t) \times 100$$

Where:

SGR (%/d) = Percentage increase in body weight per fish per day.

Ln WT = Natural log of weight at time T.

Ln Wt = Natural log of initial weight.

T = time T, t = initial time, Ln = Natural Logarithm.

$$RGR \% = 100 [\text{total length (mm)} / \text{body weight (mg)}].$$

Chemical and physical analyses of fish:

At the end of the experiments, routine proximate analysis of fish for moisture, protein, lipids (ether extract) and ash contents was carried out according to the official methods of AOAC (2000). Water holding capacity (WHC %) was calculated as $100 W_2/W_1$, where W_1 and W_2 are sample's weight before and after pressing with 5 Kgs weight, respectively as cited from Abdelhamid (1983). Lean meat (LM %) was also estimated using the equation cited from Pearson (1962) and Less (1968) where $LM\% = 100 (\text{total meat nitrogen } \%) / 3.9$. Testosterone was detected in plasma and extracts of fish and gonads as well as in waste water via Appratus Immulite (Siemens USA) FDA approved using commercial kit for chemiluminescence technique for hormonal assay.

Statistical analysis:

The obtained data were statistically analyzed using general linear models procedure adapted by SPSS (2004) for Windows for user's guide. Least significant difference according to Duncan (1955) within program SPSS was done to determine the degree of significance between means. Correlations were also calculated between some measured criteria.

RESULTS

First experiment:

Bodyweight changes:

Table 1 shows that there were no significant ($P \geq 0.05$) differences among different dietary treatments in IW, FW, TWG, or DWG of Nile tilapia fry. Yet, the fry fish in treatment No. 4 gave the highest FW, TWG and DWG, but the lowest values were noticed for fry fish of the control (T_1). Anyhow, all the treatments improved these criteria in comparison with the control. Table 2

presents the changes in the total length of the experimental tilapia fry, since there were no significant ($P \geq 0.05$) differences among treatments in the IL, but there was a significant ($P \leq 0.05$) difference in the FL only between treatments No. 4 and 6.

Table (1): Averages \pm standard errors of initial bodyweight (IW), final bodyweight (FW), total bodyweight gain (TWG) and daily bodyweight gain (DWG) of the tilapia fry in mg as affected by the dietary treatments (1st experiment).

Treatment	IW	FW	TWG	DWG
1	1.0 ^a \pm 0.0	350 ^a \pm 0.00	349 ^a \pm 0.0	5.9 ^a \pm 0.0
2	1.0 ^a \pm 0.0	480 ^a \pm 109	479 ^a \pm 109	8.14 ^a \pm 1.8
3	1.0 ^a \pm 0.0	410 ^a \pm 69	409 ^a \pm 69	6.95 ^a \pm 1.1
4	1.0 ^a \pm 0.0	845 ^a \pm 424	844 ^a \pm 424	13.55 ^a \pm 6.4
5	1.0 ^a \pm 0.0	405 ^a \pm 34	404 ^a \pm 34	6.1 ^a \pm 0.2
6	1.0 ^a \pm 0.0	370 ^a \pm 29	369 ^a \pm 29	6.3 ^a \pm 0.5

a: means in the same column superscripted with the same letter are not significantly ($P \geq 0.05$) different.

Table (2): Averages \pm standard errors of initial (IL) and final total body length (FL) of the tilapia fry in cm as affected by the dietary treatments (1st experiment).

Treatment	IL	FL
1	0.9 \pm 0.0 ^a	2.40 \pm 0.00 ^{ab}
2	0.9 \pm 0.0 ^a	2.35 \pm 0.15 ^{ab}
3	0.9 \pm 0.0 ^a	2.46 \pm 0.20 ^{ab}
4	0.9 \pm 0.0 ^a	2.85 \pm 0.35 ^a
5	0.9 \pm 0.0 ^a	2.25 \pm 0.15 ^{ab}
6	0.9 \pm 0.0 ^a	2.45 \pm 0.05 ^b

a-b: means in the same column superscripted with different letters are significantly ($P \leq 0.05$) different.

Growth and survival rates:

Table 3 presents also that there were no significant ($P \geq 0.05$) differences neither among different dietary treatments in SGR, RGR, K-factor, nor survival rate (SR) of Nile tilapia fry. Yet, the fry fish in treatment No. 4 gave the highest SGR, RGR and SR, but the lowest SGR, RGR and K-factor values were noticed for fry fish of the control (T_1). However, all the treatments had improved the SGR, RGR and K-factor in comparison with the control.

Table (3): Averages \pm standard errors of specific growth rate (SGR, %/day), relative growth rate (RGR), condition (K)-factor, and survival rate (SR) of tilapia fry as affected by the dietary treatments (1st experiment).

Treatment	SGR, %/d	RGR, %	K- factor	Survival, %
1	4.30 ^a \pm 0.00	349 ^a \pm 0.00	2.50 ^a \pm 0.00	77 ^a \pm 1.00
2	4.60 ^a \pm 0.20	479 ^a \pm 110	3.64 ^a \pm 0.14	77 ^a \pm 6.00
3	4.45 ^a \pm 0.15	409 ^a \pm 70.0	2.95 ^a \pm 0.25	76.5 ^a \pm 5.5
4	4.90 ^a \pm 0.40	844 ^a \pm 425	3.34 ^a \pm 0.65	91 ^a \pm 2.00
5	4.45 ^a \pm 0.05	404 ^a \pm 35.0	3.79 ^a \pm 1.01	81 ^a \pm 4.00
6	4.35 ^a \pm 0.05	369 ^a \pm 30.0	2.56 ^a \pm 0.00	76 ^a \pm 5.00

a: means in the same column superscripted with the same letter are not significantly ($P \geq 0.05$) differ.

Physico- chemical parameters of whole tilapia fry:

As shown in Table 4, there were significant ($P \leq 0.05$) differences among the dietary treatments in the values of water holding capacity (WHC) and fat percentages, but not ($P \geq 0.05$) in dry matter (DM) or crude protein (CP) percentages. All the dietary treatments significantly improved the WHC comparing with the control. The treatment No. 4 reflected the significantly lowest fat content and the insignificantly lowest CP content, whereas the highest fat and CP percentages were found in fry fish of the treatments No. 6 and 2, respectively. Although lean meat percentage (Table 5) did not differ significantly ($P \geq 0.05$), T_4 recorded the lowest value (1.70 %) but T_2 gave the highest one (2.03 %). Yet, Table 5 showed the significant ($P \leq 0.05$) differences among treatments in both of ash and testosterone levels in the tilapia fry carcass. Treatment No. 4 again reflected the highest ash percentage comparing with the other groups of fry fish. The testosterone concentration was the highest in the control group (T_1) followed by T_4 and lastly by each of treatments No. 5, 3, 2, and 6, respectively. Table 6 showed negative and positive correlations between some tested physico-chemical parameters in the whole carcass of the experimental tilapia fry, but the significant ones were between WHC % and testosterone hormone concentration ($r = -0.842$, $P \leq 0.001$), moisture and DM percentages, fat % and each of CP ($r = 0.631$, $P = 0.028$), ash ($r = -0.935$, $P = 0.000$), and lean meat ($r = 0.617$, $P = 0.032$), CP % and each of ash ($r = -0.855$, $P = 0.000$), and lean meat ($r = 0.994$, $P = 0.000$), and between ash and lean meat ($r = -0.953$, $P = 0.000$).

Table (4): Averages \pm standard errors of WHC, DM, fat, and CP (%) of whole tilapia fry as affected by the dietary treatments (1st experiment).

Treatment	WHC	Dry Matter	Fat	C P
1	69.00 ^b \pm 0.00	25.1 ^a \pm 0.00	24.80 ^{ab} \pm 0.00	47.50 ^a \pm 0.0
2	95.85 ^a \pm 0.44	26.4 ^a \pm 1.20	26.65 ^{ab} \pm 0.44	49.15 ^a \pm 1.7
3	95.14 ^a \pm 1.80	26.2 ^a \pm 0.89	19.25 ^{bc} \pm 3.10	45.50 ^a \pm 2.4
4	93.85 ^a \pm 2.40	26.4 ^a \pm 0.09	12.00 ^c \pm 1.30	41.10 ^a \pm 4.9
5	94.85 ^a \pm 2.90	26.3 ^a \pm 0.79	20.45 ^{ab} \pm 3.90	47.60 ^a \pm 2.3
6	94.35 ^a \pm 0.44	25.75 ^a \pm 1.1	27.50 ^a \pm 1.70	47.80 ^a \pm 4.4

a-c: means in the same column superscripted with different letters are significantly ($P \leq 0.05$) different.

Table (5): Averages \pm standard errors of ash (%), lean meat (%), and testosterone hormone concentrations (ng / g fresh weight) of whole tilapia fry carcass as affected by the dietary treatments (1st experiment).

Treatment	Ash	Lean Meat	Hormone
1	27.70 ^b \pm 0.0	1.95 ^a \pm 0.00	88.98 ^a \pm 0.0
2	24.20 ^b \pm 1.2	2.03 ^a \pm 0.07	30.51 ^c \pm 2.6
3	35.25 ^{ab} \pm 5.4	1.87 ^a \pm 0.09	31.51 ^c \pm 9.8
4	46.90 ^a \pm 6.3	1.70 ^a \pm 0.20	59.80 ^b \pm 9.2
5	31.95 ^{ab} \pm 1.7	2.00 ^a \pm 0.09	32.75 ^c \pm 5.6
6	24.70 ^b \pm 6.1	2.00 ^a \pm 0.20	30.42 ^c \pm 2.9

a –c: means in the same column superscripted with different letters are significantly ($P \leq 0.05$) different.

Table (6): Pearson correlation coefficients and probability values between the determined physico- chemical parameters of whole tilapia fry carcass (1st experiment).

Criteria	WHC %	Moisture %	Dry matter %	Fat %	Crude Protein %	Ash %	%Lean Meat
Moisture %	0.445-0.147						
Dry matter %	0.438-0.178	-1.000*					
%Fat	0.188-0.558	0.102-0.753	0.062-0.856				
Crude protein %	0.171-0.594	0.174-0.588	0.167-0.624	0.631-0.028			
Ash %	0.200-0.533	0.145-0.652	0.119-0.728	0.935-0.000	-0.855-0.000		
%Lean Meat	0.122-0.707	0.173-0.591	0.160-0.638	0.617-0.032	0.994-0.000	-0.953-0.000	
Hormone Concentration	0.842-0.001	0.353-0.261	0.361-0.276	0.136-0.674	-0.119-0.713	0.142-0.660	0.162-0.615

*: $P \leq 0.05$.

Second experiment:

Bodyweight changes:

Table 7 shows that there were no significant ($P \geq 0.05$) differences neither among different dietary treatments in IW, FW, TWG, nor DWG of Nile tilapia fry. Yet, the fry fish in treatment No. 3 gave the highest FW, TWG and DWG, but the lowest values were noticed for fry fish of the control (T_6). Table 8 gave a light on the changes of the total body length of the fry in the 2nd experiment, where no significant ($P \geq 0.05$) differences among different dietary treatments in IL were found, but there were significant ($P \leq 0.05$) differences in FL between T_5 and each of T_2 and T_3 .

Table (7): Averages \pm standard errors of initial bodyweight (IW), final bodyweight (FW), total bodyweight gain (TWG) and daily bodyweight gain (DWG) of the tilapia fry in mg as affected by the dietary treatments (2nd experiment).

Treatment	IW	FW	TWG	DWG
1	1.0 ^a \pm 0.0	2820 ^a \pm 0.00	2819 ^a \pm 0.00	30.0 ^a \pm 0.00
2	1.0 ^a \pm 0.0	1878 ^a \pm 121.0	1877 ^a \pm 12.1	20.0a \pm 0.00
3	1.0 ^a \pm 0.0	3063 ^a \pm 1904	3062 ^a \pm 19.4	36.5 ^a \pm 230
4	1.0 ^a \pm 0.0	2536 ^a \pm 0.00	2535 ^a \pm 0.00	30.0 ^a \pm 0.00
5	1.0 ^a \pm 0.0	1652.5 ^a \pm 67	1651.5 ^a \pm 6.7	20.0 ^a \pm 0.00
6	1.0 ^a \pm 0.0	1630 ^a \pm 599	1629 ^a \pm 5.99	20.0 ^a \pm 10.0

a: means in the same column superscripted with the same letter are not significantly ($P \geq 0.05$) different.

Table (8): Averages \pm standard errors of initial body length (IL) and final body length (FL) of the tilapia fry in cm as affected by the dietary treatments (2nd experiment).

Treatment	IL	FL
1	0.9 \pm 0.0 ^a	4.30 \pm 0.00 ^a
2	0.9 \pm 0.0 ^a	4.05 \pm 0.35 ^{ab}
3	0.9 \pm 0.0 ^a	4.30 \pm 1.00 ^a
4	0.9 \pm 0.0 ^a	4.32 \pm 0.00 ^a
5	0.9 \pm 0.0 ^a	3.93 \pm 0.03 ^b
6	0.9 \pm 0.0 ^a	3.80 \pm 0.80 ^b

a-b: means in the same column superscripted with different letters are significantly ($P \leq 0.05$) different.

Growth and survival rates:

Table 9 shows also that there were no significant ($P \geq 0.05$) differences among different dietary treatments in SGR, RGR, K-factor, nor survival rate (SR) of Nile tilapia fry. Yet, the fry fish in treatment No. 3 gave the highest RGR.

Table (9): Averages \pm standard errors of specific growth rate (SGR, %/day), relative growth rate (RGR, %), condition (K)-factor, and survival rate (SR, %) of tilapia fry as affected by the dietary treatments (2nd experiment).

Treatment	SGR	RGR	K-Factor	Survival rate
1	3.88 ^a \pm 0.00	281.9 ^a \pm 0.00	3.60 ^a \pm 0.00	93.0 ^a \pm 0.0
2	3.68 ^a \pm 0.03	187.7 ^a \pm 120	2.95 ^a \pm 0.60	90.5 ^a \pm 0.5
3	3.82 ^a \pm 0.40	306.2 ^a \pm 190	3.25 ^a \pm 0.05	90.5 ^a \pm 5.0
4	3.80 ^a \pm 0.00	253.5 ^a \pm 0.00	3.20 ^a \pm 0.00	95.0 ^a \pm 0.0
5	3.62 ^a \pm 0.02	165.2 ^a \pm 6.00	2.75 ^a \pm 0.20	91.5 ^a \pm 2.0
6	3.60 ^a \pm 0.20	162.9 ^a \pm 59.0	3.05 ^a \pm 0.80	84.0 ^a \pm 7.0

a: means in the same column superscripted with the same letter are not significantly ($P \geq 0.05$) different.

Physico- chemical parameters of whole tilapia fry:

Table 10 presents significant ($P \leq 0.05$) differences among treatments in each of WHC, DM, and fat percentages, but non significant ($P \geq 0.05$) in CP %. Treatment No. 3 gave the highest WHC %, but T₄ reflected the highest DM and fat percentages. Table 11 illustrates also non significant ($P \geq 0.05$) differences among treatments in ash and lean meat percentages; yet, testosterone hormone concentration in the tilapia fry carcass differed significantly ($P \leq 0.05$) in favor of T₁ against all the other treatments, while the lowest concentration was registered for T₅. Table 12 showed negative and positive correlations between some tested physico-chemical parameters in the whole carcass of the experimented tilapia fry, but the significant ones were between moisture and each of DM and fat percentages, DM and fat %, CP and each of ash and lean meat, as well as between ash and lean meat. When considering both experiments together, these physico-chemical parameters of the whole carcasses of tilapia fry gave also negative and positive correlation coefficients as shown in Table 13. Particular significant ones were between WHC % and each of fat %, ash %, and testosterone

concentration, between moisture and DM percentages, fat and ash %, CP % and each of ash and lean meat percentages, and between ash and lean meat percentages.

Table (10): Averages \pm standard errors of WHC, DM, fat, and CP (%) of whole tilapia fry as affected by the dietary treatments (2nd experiment).

Treatment	WHC	Dry Matter	Fat	C P
1	97.90 ^{ab} \pm 0.0	27.20 ^{ab} \pm 0.0	7.30 ^b \pm 0.00	47.4 ^a \pm 0.0
2	98.05 ^{ab} \pm 0.3	26.29 ^{ab} \pm 2.9	6.90 ^{bc} \pm 0.20	560 ^a \pm 0.6
3	101.6 ^a \pm 2.7	26.05 ^{ab} \pm 2.2	5.50 ^{bc} \pm 0.09	540 ^a \pm 7.5
4	96.80 ^b \pm 0.0	31.30 ^a \pm 0.0	10.2 ^a \pm 0.00	44.7 ^a \pm 0.0
5	99.20 ^{ab} \pm 0.4	23.35 ^b \pm 0.8	4.25 ^d \pm 1.00	44.9 ^a \pm 3.7
6	98.00 ^{ab} \pm 0.0	24.50 ^b \pm 0.2	4.80 ^{dc} \pm 1.10	47.8 ^a \pm 1.6

a-d: means in the same column superscripted with different letters are significantly ($P \leq 0.05$) different.

Table (11): Averages \pm standard errors of ash (%), lean meat (%), and testosterone hormone concentration (ng / g fresh weight) of whole tilapia fry carcass as affected by the dietary treatments (2nd experiment).

Treatment	Ash	Lean Meat	Hormone
1	45.30 ^a \pm 0.0	1.95 ^a \pm 0.00	88.98 ^a \pm 0.00
2	37.1.0 ^a \pm 0.4	2.30 ^a \pm 0.00	49.35 ^b \pm 19.5
3	40.50 ^a \pm 7.4	2.21 ^a \pm 0.30	38.69 ^b \pm 12.4
4	45.10 ^a \pm 0.0	1.90 ^a \pm 0.00	50.58 ^b \pm 0.00
5	50.85 ^a \pm 4.7	1.85 ^a \pm 0.10	16.69 ^b \pm 3.10
6	47.4.0 ^a \pm 2.7	1.95 ^a \pm 0.04	27.40 ^b \pm 6.70

a – b: means in the same column superscripted with different letters are significantly ($P \leq 0.05$) different.

Table (12): Pearson correlation coefficients and probability values between the determined physico- chemical parameters of whole tilapia fry carcass (2nd experiment).

Criteria	WHC %	Moisture %	Dry matter%	Fat %	Crude protein %	Ash %	%Lean Meat
Moisture %	0.459 0.182						
Dry matter %	0.459– 0.182	-1.000 *					
Fat %	0.326– 0.358	0.769– 0.009	0.769 0.009				
Crude protein %	0.098– 0.788	0.263– 0.463	0.263 0.463	0.126 0.728			
Ash %	0.185 0.608	0.468 0.173	0.468– 0.173	0.410– 0.240	-0.957 0.000		
%Lean Meat	0.131– 0.719	0.344– 0.331	0.344 0.331	0.195 0.589	0.995 0.000	-0.972 0.000	
Hormone Concentration	0.005– 0.990	0.175– 0.629	0.175 0.629	0.521 0.122	0.038 0.917	0.188– 0.604	0.049 0.894

*: $P \leq 0.05$.

Table (13): Pearson correlation coefficients and probability values between the determined physico- chemical parameters of whole tilapia fry carcass (1st and 2nd experiments together).

Criteria	WHC %	Moisture %	Dry matter %	Fat %	Crude protein %	Ash %	%Lean Meat
Moisture %	- 0.048 0.831						
Dry matter %	0.046 0.842	-1.000 *					
Fat %	0.511- 0.015	0.114- 0.613	0.118 0.610				
Crude protein %	0.059 0.795	0.141- 0.531	0.183 0.426	0.089- 0.693			
Ash %	0.429 0.046	0.177 0.431	0.202- 0.380	0.853- 0.000	-0.443 0.039		
%Lean Meat	0.063 0.780	0.191- 0.394	0.242 0.291	0.053- 0.816	0.994 0.000	-0.473 0.026	
Hormone Concentration	0.585- 0.004	0.024- 0.914	0.022 0.925	0.101 0.653	- 0.066 0.769	0.057- 0.803	0.081- 0.720

*: $P \leq 0.05$.

DISCUSSION

For interpretation of the obtained results, Hanley *et al.* (1995) found that dietary inclusion of Aqua-Mos may greatly improve the survival, the biomass recovered and subsequent the economics of the production of the hatchery-reared tilapia. Also, Attia *et al.* (2007) confirmed the economical – environmental benefits of using the dietary supplemental microbial – phytase. It increases ($P < 0.05$) body weight, feed efficiency, carcass %, and serum contents of P, Ca, Mg and Zn.

Generally, the probiotics actively inhibit the colonization of potential pathogens in the digestive tract by antibiosis or by competition for nutrients and/or space, alteration of microbial metabolism, and/or by the stimulation of host immunity. Probiotics may stimulate appetite and improve nutrition by the production of vitamins, detoxification of compounds in the diet, and by breakdown of indigestible components (Irianto and Austin, 2002). So, Eid and Mohamed (2000) proved that Biogen® and Prmifer® improved the growth performance, feed conversion, protein efficiency ratio and apparent protein digestibility for monosex tilapia fingerlings compared to fish fed the control diet. No significant differences were detected in plasma proteins. Feed cost required to produce 1 Kg weight gain compared to fish fed the control diet was reduced by using commercial feed additives (Biogen® and Prmifer®). Also, Sittipun *et al.* (2000) found that shrimp fed a crude β -glucan had enhanced immunity, higher hemocytes count, higher bacterial clearance rates and better resistance to bacteria.

Feed conversion ratio, body weight, fish production and survival of rainbow trout and common carp were significantly improved by supplementing diets with 2 g/kg Bio-Mos®. Adding 2 g/kg Bio-Mos® to the feed significantly improved immune capacity of rainbow trout and common carp as indicated by serum lysozyme concentration and complement activity

(triggered both through alternative and classical pathways). Bio-Mos® supplementation improved performance of rainbow trout and common carp and modified immune response, which could result in improved fish health (Staykov *et al.*, 2005).

Koeleman (2007) reported that different products based on mannan oligosaccharides, better known as prebiotics, have been shown to benefit animal health and performance. A new product derived from yeast found on fruit trees is the latest promising edition to the prebiotic portfolio. Biobuds® and yeast or Biogen® as probiotics led to improving the growth performance, feed conversion, protein efficiency ratio and feed costs for tilapia fingerlings (Mohamed, 2007 and Mohamed *et al.*, 2007, respectively). Moreover, El-Ashram *et al.* (2008) concluded that, super Biobuds® can improve body gain, survival and enhance resistance to challenge infection. Yet, Abdelhamid and Elkatan (2006) found that dietary supplementation of Biobuds® slightly improved body weight gain but reduced the survival rate of tilapia fingerlings. El-Haroun *et al.* (2006) and El-Haroun (2007) reported that Biogen® dietary supplementation improved growth performance and feed utilization, carcass protein and fat percentages as well as economical profit in Nile tilapia and catfish culture, respectively.

Barbu *et al.* (2008) mentioned that Bio-Mos (a prebiotic or a pro-nutrient) is a mannan oligosaccharide derived from the outer cell wall of a specific strain of *Saccharomyces cerevisiae*. They added that the functional foods are a group of foods that contain in their structure biologically active components that may improve the health status and that can have an impact on some physiological effects in addition to their nutritional function. Recently, its effectiveness in aquaculture has been established. Improvements in the growth performance and health status of several species of fish are being seen as a result of supplementing the fish feeds with mannan oligosaccharide. Since, 0.2 – 0.6 % dietary inclusion of Bio-Mos for carp, rainbow trout and European catfish led to a superior weight gain (11.6 – 24.0 %), the improvement of feed conversion ratio (from 2.0 to 1.6) and to reduction of mortalities percent, 1.92 vs. 3.59 % for carp. Bio-Mos® also improved anterior gut morphology and the immune status. Also, dietary beta glucans are natural feed ingredients that can modulate the inflammatory response and therefore provide, in combination with extra vitamins and minerals, a promising tool for fighting chronic enteritis (Star *et al.*, 2009). Algedawy *et al.* (2011) concluded that the probiotic Biogen® is superior to the multienzyme mixture Natuzyme® as a feed additive for improving the growth performance and cellular and humoral immune responses.

There was a negative relationship between crude proteins and crude fats in the chemical composition of Nile tilapia fish on one hand (El-Ebiary and Zaki, 2003 and Abdelhamid *et al.*, 2007b&c), and a positive correlation between crude protein and crude ash contents of Nile tilapia fish, on the other hand (Abdelhamid *et al.*, 2007a). A negative relationship was noticed between CP and EE contents of fish body but a positive relationship between CP and ash contents was recorded too (Abdelhamid *et al.*, 2000 and El-Saidy and Gaber, 2002). El-Saidy and Gaber (2002) found that there was a positive correlation between crude protein and fat contents of the fish, but Abdelhamid

et al. (2005a&b) found a negative correlation between protein and fat contents of the fish.

From the obtained results in the present study, testosterone level in fish carcass was lower in all treatments comparing with control of both experiments. Moreover, it was lower in T₄ of the 2nd experiment than in the same treatment (T₄) of the 1st experiment. Since the high level in the control is due to natural secretion whereas the low levels in the treatments are due to that the external application of the hormone depresses the natural secretion of the gonads. Also, by age advance, its level decreases due to its half live time. The obtained results are in accordance with those given by Nemat Allah (2002), where the obtained levels are of the natural produced (endogenous) hormone and there is no residual effect of the synthetic (exogenous) hormone used for sex reversal. Nemat Allah (2002) mentioned also that during the treating fry with the low level of the male sex hormone, more than 90% of the hormone quantity is concentrated in the viscera, whereas 10% of the hormone goes to the muscles. He added that, after the 28 day-treatment period, the hormone is quickly disappeared. After 4 days (0.5 g body weight), 1% only is found in the viscera. So, after one month of the hormonized diet withdrawal, it seems obviously that there is no any trace of the synthetic hormone in the fish body. After 4 months of the withdrawal (catch and marketable size), the level of normal (endogenous) male sex hormone in the mono-sex Nile tilapia was 11 ng/g comparing with 37 ng/g in the normal male (not sex reversed) male. For knowledge, the effective treating dose of the male sex hormone for human beings is 80 mg (1333 times the dose required for fish sex reversal). He attributed the lower male sex hormone level in mono-sex than the normal male to the depression of hormone production from pituitary gland, which stimulates the sex hormone production from the gonads. This means that the hormone found in the mono-sex is the natural (endogenous) hormone only.

In conclusion, we could recommend the use of 17 α -methyl testosterone at 60 mg / Kg diet for sex reverse during the early stage after the hatch and to increase the growth and survival besides decreasing fat content without any danger from the hormone residues. The dietary inclusion of Bio-Mos® was not beneficial, whereas the dietary inclusion of T-Protophyt 2000 at 4 g / Kg diet was positively effective in increasing the growth performance of the tilapia fry.

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تأثير الإضافات الغذائية "البيوموس" و "البروتفيت 2000" مع أو بدون المعاملة الهرمونية على أداء النمو، التركيب الكيماوى، ومتبقيات الهرمون فى البلطي النيلي وحيد الجنس

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كان هدف الدراسة هو تقييم تأثيرات احتواء علائق ذريعة أسماك البلطي النيلي حديثة الفقس (في عمر معاملتها الهرمونية لقلب الجنس لإنتاج أسماك وحيدة الجنس ذكور) على مستويات متدرجة من البريبيوتيك (Bio-Mos®) في تجربة أولى و البريبيوتك (T-Protphyt 2000) في تجربة ثانية، مع أو بدون هرمون 17- ألفا - ميثايل - تستوستيرون في كلى التجريبتين، وكانت التغذية التجريبية لمدة 21 يوما أعقبها 8 أو 12 أسبوع (فى التجريبتين على التوالي) بمعدل 10% من وزن جسم الذريعة يوميا، وذلك في أحواض زجاج.

هذا ويمكن تلخيص أهم النتائج المتحصل عليها من التجريبتين في أنه لم تكن هناك فروق معنوية على مستوى 0.05 بين المعاملات المختلفة في كلى التجريبتين في قيم مقاييس أداء النمو، رغم تفوق المعاملة الرابعة (المحتوية على 60 مجم 17 ألفا ميثايل تستوستيرون / كجم عليقة) بشكل ملحوظ (الضعف تقريبا) في التجربة الأولى والمعاملة الثالثة (المحتوية على 4 جم T-Protphyt 2000 / كجم عليقة) في التجربة الثانية على باقي المعاملات الخمسة المختلفة الأخرى. تركزت الفروق المعنوية على مستوى 0.05 بين المعاملات المختلفة في التجريبتين في بعض قيم خصائص جسم السمك، خاصة في كل من نسبة قدرة الاحتفاظ بالماء (فكانت كل المعاملات أعلى معنويا عن المقارنة) وتركيز هرمون التستوستيرون في جسم السمك (فكان المستوى الأقصى في أسماك المقارنة). كانت هناك ارتباطات سالبة وأخرى موجبة بين الخصائص الفيزيوكيماوية لجسم الأسماك. ونخلص من هذه الدراسة إلى أهمية استخدام هرمون الجنس الثانوي الذكري (17 ألفا ميثايل تستوستيرون) بالمعدل المستخدم تجاريا (60 مجم / كجم عليقة، والمُختبر وفي هذه الدراسة) في تغذية الذريعة حديثة الفقس (لمدة 21 يوما لعكس الجنس لإنتاج وحيد جنس كله ذكور من البلطي النيلي، كما هو متبع تجاريا واختبر في هذه الدراسة) لزيادة نموها وحياتها، ولخفض دهون أجسامها، دون أي خطورة من احتوائها على متبقيات الهرمون، وأن إضافة البريبيوتيك Bio-

Mos® لم يكن له جدوى، بل قد يزيد من تكاليف الإنتاج، بينما استخدام الـ T-Protyt 2000 بتركيز 4 جم/كجم علف قد يكون مثمرا في زيادة الوزن ومعدلات النمو، مما قد يكون لاستخدامه أهمية اقتصادية في إنتاج الذريعة من أسماك البلطي النيلي.

قام بتحكيم البحث

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