

Effect of Dietary Chromium Picolinate Supplementation on Growth Performance, Carcass Composition and Organs Indices of Nile Tilapia (*Oreochromis niloticus* L.) Fingerlings

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ABSTRACT

This experiment was conducted to study the effects of dietary chromium picolinate (Cr-Pic) on growth performance, carcass composition and internal organs indices of mono-sex Nile tilapia (*Oreochromis niloticus*) fingerlings in an in-door 12-weeks trial. Fish with similar body weight (28.00 ± 0.96 g) were distributed randomly into seven treatments at the same stocking density of 20 fish m^{-3} , being the control fish group (T_1) which was fed a basal diet free from supplemented Cr-Pic. Other fish groups were fed the basal diet supplemented with graded levels of Cr-Pic, being 200 ppb (T_2), 400 ppb (T_3), 600 ppb (T_4), 800 ppb (T_5), 1000 ppb (T_6) and 1200 ppb (T_7). The obtained results revealed that the dietary graded levels of Cr-Pic had no significant ($p \geq 0.05$) effects on growth performance, carcass composition and internal organs' indices of the experimental fish, but dietary Cr-Pic at 400 ppb (T_3) led to significant ($p \leq 0.05$) increase in dry matter and crude protein of fish carcass compared with the control treatment and also the same treatment (T_3) had significant increased of gonado-somatic index (GSI σ , %) among all treatments.

Key words: Nile tilapia, chromium, growth performance, carcass composition, organs indices

INTRODUCTION

Tilapias originally from Africa, have been reared for centuries (FAO, 2001). Nile tilapia (*Oreochromis niloticus*, Linnaeus 1758) is one of the most cultured fish in tropical and subtropical regions and contributes more than 85 percent to world production (FAO, 2007). In addition, world production of tilapias has increased from 1.9, 2.8, 3.6 million tones in 2000, 2005 and 2008, respectively. Furthermore, consumer demand for tilapias has been increasing at a faster rate than many other species (Feidi, 2010). Tilapia culture is believed to have originated some 4000 years ago in Egypt, where Nile tilapia *O. niloticus* are considered as the most popular fish and widely cultured species (Stickney, 2006).

According to recent statistics of the Egyptian General Authority for Fish Resources Development (GAFRD, 2009), tilapia consist 55.6% (477.458 tones) of the Egyptian production from fish culture (693.815 tones) in 2008. Also, Egypt produces 12% of the world farmed tilapia (2.121.009 tones) (FAO, 2007). Moreover, Egypt is by far the main tilapia producer in the Middle East and North Africa (MENA) region, where produced 477.458 tones in 2008 of tilapia, or 92.2% of all tilapia production in this region (Feidi, 2010). Yet, Nile tilapia is cultured in 23 African countries out of 32 countries that practiced tilapia culture in Africa. In Egypt, most of the aquaculture production of tilapia is derived from semi-intensive fish farms in earthen ponds, intensive systems, integrated intensive fish farms and cages (GAFRD, 2006).

Chromium (Cr) is an essential micro-mineral; it plays important role in nutritional and physiological responses on fish (Kucukbay *et al.*, 2006; Liu *et al.*, 2010). It is found in the environment commonly in trivalent, Cr (III) and hexavalent, Cr (VI), forms (Bagchi *et al.*, 2002). Various trivalent chromate compounds have been used as feed additives in fish diet due to its participation in carbohydrate, protein and fat metabolism (Pan *et al.*, 2002a; Liu *et al.*, 2010). Chromium picolinate (Cr-Pic) is the most popular form of trivalent Cr (III), its chemical formula is Cr (C₆H₄NO₂)₃ (NBI, 2001). It is generally accepted that organic chromium sources such as chromium picolinate, chelated Cr, chromium-amino acid complexes and yeast-incorporated Cr have more bioavailability than inorganic sources (Shiau and Liang, 1995; Shiau and Shy, 1998).

Several studies dealing with the effect of Cr on fish have been related to its role in enhancement of growth performance in hybrid tilapia (*Oreochromis niloticus*; *O. aureus*) (Pan *et al.*, 2002b; 2003), grass carp (*Ctenopharyngodon idellus*) (Liu *et al.*, 2010) and rainbow trout (*Oncorhynchus mykiss*) (Bureau *et al.*, 1995; Selcuk *et al.*, 2010); metabolism in channel catfish (*Ictalurus punctatus*) (Paripatananont and Lovell, 1997) and gilthead sea bream (*Sparus aurata*) (Fernandez *et al.*, 1999; Gatta *et al.*, 2001a); carbohydrate utilization in hybrid tilapia (*O. niloticus* X *O. aureus*) (Shiau and Shy, 1998; Pan *et al.*, 2002a, 2003), striped bass (*Morone saxatilis*) and sunshine bass (*M. chrysops* X *M. saxatilis*) (Rawles and Gatlin 1998); immune status in *O. mossambicus* (Arunkumar *et al.*, 2000) and rainbow trout (*O. mykiss*) (Gatta *et al.*, 2001b) and toxicity in largemouth bass (*Micropterus salmoides*) (Kuykendall *et al.*, 2006).

Hence, studies concerning with effects of dietary Cr on growth performance of some fish species revealed incompatible results (Shiau and Liang, 1995; Shiau and Shy, 1998; Pan *et al.*, 2002b, 2003). Furthermore, the pathway of Cr absorption is not very well understood (Paripatananont and Lovell, 1997). Yet, dietary Cr (III) is often lost during food handling and processing, as well as, Cr bioavailability is inhibited by other nutrients such as phytates, dietary fiber and other minerals (NBI, 2001). So, the present experiment was carried out to study the effects of graded levels of organic Cr (III) as Cr-Pic on growth performance, chemical analysis of carcass and internal organs' indices of mono-sex Nile tilapia (*Oreochromis niloticus*) fingerlings throughout 12-weeks as an experimental period.

MATERIALS AND METHODS

Experimental procedures and treatments: This study was conducted in Fish Research Unit, Faculty of Agriculture, Al-Mansoura University, Al-Dakahlia governorate, Egypt. Mono-sex Nile tilapia (*O. niloticus*) fingerlings were obtained from a private hatchery in Al-Reiad belonging to Kafr El-Sheikh governorate. Fish were stocked into a rearing tank for two weeks as an adaptation period, during which they were fed a basal experimental diet. Thereafter, apparently-healthy fish with similar body weight (28.00±0.96 g) were distributed randomly into seven treatments (at three replicates per treatment) at the same stocking density of 20 fish m⁻³, being the control fish group (T₁) fed the basal diet free from supplemented chromium picolinate (Cr-Pic). Other fish groups were fed the basal diet with the addition of graded levels of Cr-Pic, being 200 ppb (T₂), 400 ppb (T₃), 600 ppb (T₄), 800 ppb (T₅), 1000 ppb (T₆) and 1200 ppb (T₇). During the experimental period (12 weeks), the fish were fed the experimental diet at a rate of 3% of the live body weight daily, six days a week. The amount of feed was adjusted bi-weekly based on the actual body weight changes, where all fish were weighted in each fiberglass tank. Experimental diet was introduced by hand twice daily, at 8.00 a.m. and 14.00 p.m. Each tank (1 m³ in volume), was constructed with an air stone

connected with electric compressor. Dechlorinated tap water was used to change one third of the water in each tank every day. Wastes were removed from tanks by siphoning. Water quality parameters in each tank were measured weekly included temperature (via a thermometer), pH-value (using Jenway Ltd., Model 350-pH-meter) and dissolved oxygen (using Jenway Ltd., Model 970- dissolved oxygen meter), where water temperature was $26.0 \pm 0.8^\circ\text{C}$, pH-value 8.19 ± 0.2 and dissolved oxygen $7.21 \pm 0.3 \text{ mg L}^{-1}$, which were suitable for rearing mono-sex *O. niloticus* fingerlings according to Abdelhamid (2009). Light was controlled by a timer to provide a 14 h light: 10 h dark as a daily photoperiod. Approximately, thirty fish were kept frozen at the start of the experiment for chemical analysis.

Experimental diets: The basal diet used was consisted of 89.19% dry matter, 27.24% crude protein, 6.42% crude lipid, 55.43% carbohydrates, 10.91% ash, 439.94 Kcal 100 g^{-1} DM gross energy (GE) and 61.91 mg CP Kcal^{-1} GE, P/E ratio. The GE was calculated according to Halver and Hardy (2002). Feed ingredients were ground and mixed manually with warm water and molasses, where Cr content in feed was not analyzed according to Kucukbay *et al.* (2006) and Selcuk *et al.* (2010). Then, graded levels of Cr-Pic (0, 200, 400, 600, 800, 1000 or 1200 $\mu\text{g kg}^{-1}$ diet) were added to prepare the experimental diets. Thereafter, the experimental diets were pressed by manufacturing machine (pellets diameter 1 mm).

Sampling procedure: At the end of the experiment, ten fish from each tank were anaesthetized by putting them in a small plastic tank containing 10 L water with 3 mL pure clove oil solved in 10 mL absolute alcohol as a natural anesthetic material, where five fish per tank were sampled and kept frozen for chemical analysis. The chemical analyses of the basal diet and whole fish body were carried out as moisture which was determined according to the AOAC (2000), crude protein by Kjeldahl according to Lakin (1978), crude lipid was extracted by petroleum ether ($40\text{-}60^\circ\text{C}$) according to Crosby (1995) and ash by dry ashing according to Petterson *et al.* (1999). Energy content in fish body ($\text{Kcal } 100 \text{ g}^{-1}$) was calculated according to Halver and Hardy (2002) as crude protein (%) $\times 5.64$ + crude lipid (%) $\times 9.44$. Body weight of individual fish was measured bi-weekly to point feed quantity and to calculate growth performance according to Halver and Hardy (2002). The liver, spleen, kidneys and gonads' indices were calculated as percentage from fish live body weight, where: Hepato-somatic index (HSI, %) = liver weight (g) $\times 100$ /fish weight (g) (Jangaard *et al.*, 1967), spleen-somatic index (SSI, %) = spleen weight (g) $\times 100$ /fish weight (g), kidney-somatic index (KSI, %) = kidneys weight (g) $\times 100$ /fish weight (g) (Alabaster and Lloyd, 1982) and gonado-somatic index (GSI σ , %) = gonads weight (g) $\times 100$ /fish weight (g) (Tseng and Chan, 1982).

Statistical analysis: Data were reported as mean values of all treatments (T_1 - T_7), replicates (tanks, $n = 3$) and standard errors of (means \pm SEM) are based on tank values (pooled values). Data were subjected to one-way analysis of variance (ANOVA), using the General Linear Model procedure (GLM) of the SAS software package (SAS, 2001) and evaluated by using the following equation:

$$Y_{ij} = \mu + A_i + e_{ij}$$

where, Y_{ij} is an observation of growth performance, chemical analysis of carcass and internal organs' indices of mono-sex Nile tilapia, μ is least square mean, A_i is the fixed effect of dietary

Cr-Pic levels (T_1 - T_7) and e_{ij} is the random error. Statistical significant ($p \leq 0.05$) differences between means were compared by using Tukey's multiple comparison analysis.

RESULTS

Growth performance: Dietary graded levels of Cr-Pic (except T_6) had no significant ($p \geq 0.05$) effects on final weight, average weight gain and specific growth rate compared with the control treatment (T_1). Also, survival rate and feed conversion ratio of the experimental fish were not affected among all treatments. Meanwhile, dietary supplemented Cr-Pic at 400 ppb (T_3) and 600 ppb (T_4) had significantly ($p \leq 0.05$) increased the feed intake compared with other graded levels of dietary Cr-Pic (Table 1).

Carcass composition of the experimental fish: Data in Table 2 showed that dietary supplementation of Cr-Pic at 400 ppb (T_3) lead to significant ($p \leq 0.05$) increase in dry matter and

Table 1: Effect of graded levels of dietary chromium picolinate supplementation on growth performance parameters of mono-sex *Oreochromis niloticus* (Means \pm SEM)

Treatment	Body weight (g fish ⁻¹)						
	Initial	Final	AWG	SGR (% day ⁻¹)	FI (g)	FCR	SR (%)
T_1	28.0 ^A	91.0 ^{ab}	63.0 ^{ab}	1.373 ^a	96.2 ^a	1.553 ^A	100.0 ^A
T_2	28.0 ^A	83.0 ^{bc}	55.0 ^{bc}	1.276 ^{ab}	88.8 ^b	1.633 ^A	100.0 ^A
T_3	28.0 ^A	99.0 ^a	71.0 ^a	1.480 ^a	100.7 ^a	1.436 ^A	100.0 ^A
T_4	28.0 ^A	99.0 ^a	70.0 ^a	1.470 ^a	100.3 ^a	1.440 ^A	100.0 ^A
T_5	28.0 ^A	93.0 ^{ab}	64.0 ^{ab}	1.393 ^a	87.6 ^b	1.390 ^A	100.0 ^A
T_6	28.0 ^A	76.0 ^c	47.0 ^c	1.153 ^b	76.9 ^c	1.643 ^A	100.0 ^A
T_7	28.0 ^A	93.0 ^{ab}	64.0 ^{ab}	1.393 ^a	87.4 ^b	1.390 ^A	100.0 ^A
SEM	± 0.96	± 4.24	± 4.20	± 0.06	± 2.43	± 0.09	± 0.00

AWG: Average Weight Gain; SGR: Specific Growth Rate; FI: Feed Intake; FCR: Feed Conversion Ratio; SR: Survival rate; SEM: Standard error of means which are pooled values. Means having different small letters were significantly different ($p \leq 0.05$), but means having capital letter are not significantly different ($p \geq 0.05$)

Table 2: Effect of graded levels of dietary chromium picolinate supplementation on carcass composition of mono-sex *Oreochromis niloticus* (Means \pm SEM)

Treatment	Dry matter (%)	% on dry matter basis			
		Crude protein	Crude lipid	Ash	Energy content (Kcal 100 g ⁻¹)
At the start of the experiment	20.5	55.2	24.4	20.4	541.5
At the end of the experiment					
T_1	24.5 ^c	61.0 ^{bc}	24.2 ^b	14.8 ^{bc}	572.6 ^c
T_2	24.1 ^c	59.5 ^c	27.0 ^a	13.5 ^{ab}	590.7 ^a
T_3	28.4 ^a	63.7 ^a	20.7 ^c	15.6 ^{ab}	554.9 ^d
T_4	27.9 ^a	63.2 ^{ab}	23.7 ^b	13.1 ^c	580.7 ^{bc}
T_5	24.7 ^c	61.5 ^{abc}	24.2 ^b	14.3 ^{cd}	575.4 ^c
T_6	26.1 ^b	56.6 ^d	27.5 ^a	15.9 ^{ab}	578.6 ^{bc}
T_7	22.0 ^d	62.2 ^{ab}	21.4 ^{bc}	16.3 ^a	553.5 ^d
SEM	± 0.24	± 0.70	± 0.86	± 0.30	± 4.61

SEM: Standard error of means which are pooled values. Means having different letters were significantly different ($p \leq 0.05$)

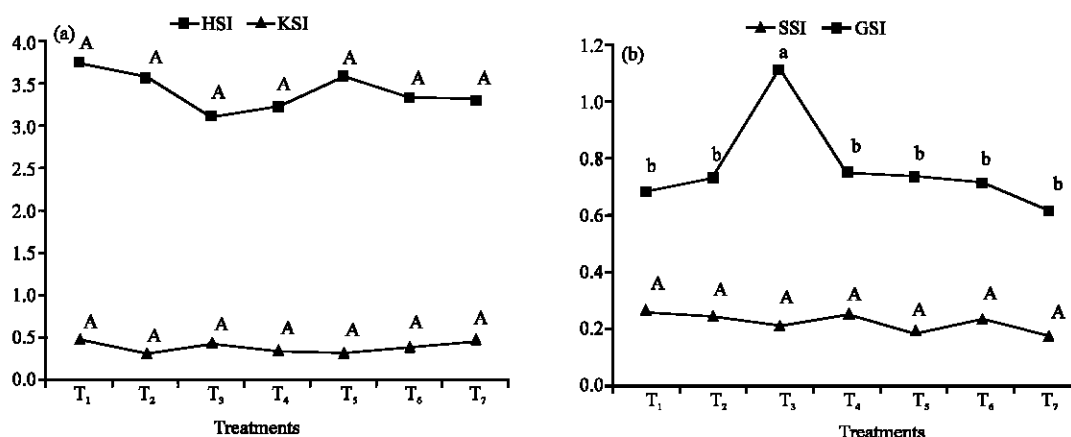


Fig. 1(a-b): Effect of graded levels of dietary chromium picolinate supplementation on internal organs indices of mono-sex *Oreochromis niloticus*, (a) HSI: Hepato-somatic index (%); KSI: Kidney-somatic index (%); (b) SSI: Spleen-somatic index (%); GSI: Gonado-somatic index (%). Means having different small letters were significantly different ($p \leq 0.05$), but means having capital letter are not significantly different ($p \geq 0.05$)

crude protein, but ash did not differ significantly ($p \geq 0.05$) compared with the control treatment. While, both of crude lipid and energy content were decreased significantly ($p \leq 0.05$) in the same treatment (T₃) compared with the control treatment. However, neutral effects on carcass composition parameters in the same treatment (T₃) were observed when compared with the other graded levels of dietary Cr-Pic. Also, the similar neutral effects of the graded levels of dietary Cr-Pic could be noted on carcass composition when compared with the control treatment.

Internal organs' indices: No significant ($p \geq 0.05$) effects were recorded in hepato-somatic index (HSI, %), kidney-somatic index (KSI, %) and spleen-somatic index (SSI, %) of fish fed dietary graded levels of Cr-Pic compared with the control treatment. Meanwhile, mono-sex *O. niloticus* fed on dietary Cr-Pic inclusion at level of 400 ppb (T₃) revealed significant ($p \leq 0.05$) increase of gonado-somatic index (GSI[♂], %) among all treatments (Fig. 1a, b).

DISCUSSION

Results in the present study, revealed that the graded levels of dietary supplemented Cr-Pic (except T₀) had no significant ($p \geq 0.05$) effects on growth performance parameters compared with the control treatment (T₁). Although, dietary supplemented Cr-Pic at 400 ppb (T₃) and 600 ppb (T₄) had significantly ($p \leq 0.05$) increased the feed intake. In this respect, no significant ($p > 0.05$) differences in weight gain were found among groups of hybrid tilapia (*Oreochromis niloticus* X *O. aureus*) fed diets without or with 2 mg Cr-Pic kg⁻¹ (Pan *et al.*, 2003) or fed chromium nicotinate (Cr-Nic) (Pan *et al.*, 2002a). Yet, studies on gilthead sea bream (*S. aurata*) (Gatta *et al.*, 2001a) and rainbow trout (*O. mykiss*) (Bureau *et al.*, 1995; Selcuk *et al.*, 2010) showed that dietary supplementation of Cr-yeast or Cr-Pic did not improve growth performance. Moreover, El-Sayed *et al.* (2010) found that dietary supplementation of Cr-Pic (up to 1200 ppb) has not any significant effect on growth performance of Nile tilapia (*O. niloticus*). In contrary with the obtained results, it has been reported that dietary Cr supplements as chromic oxides (Shiau and Liang, 1995; Shiau and Shy, 1998) and chromium chloride (Shiau and Lin, 1993) significantly increased weight

gain, feed intake, protein and energy retention in hybrid tilapia fed diets containing high levels of glucose. Similar results were obtained on the same species when either Cr-Nic or Cr-Pic was included in a high glucose diet (Pan *et al.*, 2002b). In addition, dietary Cr-Pic has also been reported to decrease mortality rate in poultry, pigs and fish (NRC, 1997). Furthermore, the results of studies on rainbow trout (Tacon and Beveridge, 1982) and channel catfish (Ng and Wilson, 1997) showed that dietary trivalent Cr supplementation did not improve growth performance or feed efficiency. Also, in gilthead sea bream (Gatta *et al.*, 2001a) and rainbow trout (Bureau *et al.*, 1995), chromium yeast incorporated in practical diets did not affect the growth performance. In this trend, the variation obtained between the different studies is not surprising, because a variety of factors such as the form of chromium used, dose and duration of treatment were not standardized among the different trials.

Yet, the obtained results of fish carcass composition showed that dietary supplementation of Cr-Pic at 400 ppb (T_3) had increased significantly ($p \leq 0.05$) dry matter and crude protein percentage compared with the control; but in the same treatment, carcass lipid content was lowest although its highest fish weights which may be due to the highest crude protein content in fish carcass (Table 2) since there is a negative relationship between protein and lipid content, Abdelhamid *et al.* (2007), as well as due to Cr which is necessary for metabolism of carbohydrates, lipids and many aspects of fish metabolism (Pan *et al.*, 2002a, 2003). The obtained results also are supported by that of El-Sayed *et al.* (2010) who found that dietary supplementation of Cr-Pic (up to 1200 ppb) to Nile tilapia (*O. niloticus*) diets has significantly increased crude protein content and decreased ether extract content but ash content was not significantly ($p > 0.05$) affected. Moreover, Liu *et al.* (2010) found that dietary Cr-Pic significantly lowered carcass lipid content in grass carp (*C. idellus*). However, it has been reported that no significant effect on crude protein and ash content of fish fed dietary Cr-Pic.

No significant ($p \geq 0.05$) differences in internal organs' indices (HSI, KSI and SSI %) of mono-sex *O. niloticus* were related to the graded levels of dietary Cr-Pic. This may be explained as Cr-Pic did not cause any adverse effects on internal organs associated with absent of fish mortality among all treatments (Table 1) which confirming the role of chromium for enhancement the immune responses of hybrid tilapia (*O. niloticus* X *O. aureus*) (Magzoub *et al.*, 2009). In addition, El-Afifi (2008) reported that the liver weight percentage of broiler chicks was not affected by dietary Cr-Pic or chromium yeast. Moreover, El-Hommosany (2008) confirmed the same result, whereas he did not find any effect on relative weights of liver and spleen due to adding chromium chloride into quail diets. However, fish fed dietary Cr-Pic at 400 ppb (T_3) had increased significantly ($p \leq 0.05$) of gonado-somatic index (GSI σ , %) among all treatments, which may be related an indicators for reproductive physiological effect of Cr-Pic on tilapia fish. Hence, it appears that there are no attempts were done related with its reproductive effects in fish. In particular, it has been shown to have a positive influence of Cr (III) on the reproductive efficiency of pigs and cattle (Mallard and Borgs, 1997).

CONCLUSIONS

Generally, the obtained results revealed that the experimental dietary graded levels of Cr-Pic had no significant effects on growth performance, carcass composition and internal organs' indices parameters, but dietary Cr-Pic at 400 ppb (T_3) led to significant ($p \leq 0.05$) increase in dry matter and crude protein of mono-sex Nile tilapia *Oreochromis niloticus* carcass compared with the control treatment. Also, the same treatment (T_3) had significant increased of gonado-somatic index

(GSI[♂], %) among all treatments. So, it could be concluded that the experimental dietary graded levels of organic chromium as Cr-Pic are the safety levels for the experimental fish.

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