

Effect of fungicide copper oxychloride and vitamin C on growth performance and nutrient utilization of Nile tilapia (*Oreochromis niloticus*)

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Abstract

The present study was designed to investigate the effect of four levels of copper oxychloride (0, 0.25, 0.50 and 0.75 LC₅₀) on growth performance and feed utilization of Nile tilapia. Three levels of vitamin C (0, 1 and 2/ kg diet) were used with each level of the fungicide to determine the effect of vitamin C in reducing the deleterious of fungicide, therefore twelve treatments were included in present study. At the end of the experiment after 120 days, fungicide levels used in the present study showed adverse effect on all growth performance parameters of Nile tilapia studied. Final body weight, final body length, weight gain, specific growth rate and survival rate of fish decreased (almost significantly) with increasing the fungicide level. Whereas, vit. C had mostly, an increasing trend on the previous growth performance parameters of fish with increasing dietary vit. C level, but no significant differences were observed among dietary treatments, except that survival rate was significantly ($P < 0.05$) increased with increasing dietary vit. C level. Feed utilization parameters (feed intake, feed conversion ratio and protein efficiency ratio) for Nile tilapia fish decreased with increasing fungicide level and the differences were only significant for feed intake, while dietary vit. C level had no significant effect on all feed utilization parameters of fish.

Key words: Nile tilapia, *Oreochromis* spp., copper oxychloride, fungicides, vitamin C.

Introduction

Aquaculture is one of the fastest growing food-producing sectors, supplying approximately 40% of the world's fish food (David *et al.*, 2009). Fish is one of the cheapest and promising sources of animal protein; people can easily digest 93.2% and 93.7% of fish protein and fat, respectively (Onusiriuka, 2002).

Nile tilapia is a native fish species of Egypt that has become popular species worldwide mainly as a valuable fish, easy to breed and grow in a variety of aquaculture systems (El-Sayed, 2006)

Pollution of the aquatic environments has become a more serious concern during the recent years. The loading of metals resulting from industrial and agricultural discharges into our environment creates water pollution problems due to their toxic effect on aquatic biota. Even though copper is an essential element serving many useful function in fish, it

is extremely toxic to fish when present at elevated levels (Sorensen, 1991).

Vitamin C plays an important role in growth and immunity of fish (Lin and Shiau, 2005). It is an effective antioxidant for collagen synthesis, helps to maintain various enzymes in their required reduced form, and participates in the biosynthesis of carnitine, norepinephrine and certain neuroendocrine peptides. Most teleosts are unable to synthesize ascorbic acid due to lack of L-gulonolactone oxidase that is responsible for synthesis of vitamin C (Fracalossi *et al.*, 2001). Therefore, an exogenous source of vitamin C is required in fish. The objective of the present study was to evaluate the use of vit. C as a protective agent against environmental copper toxicity for Nile tilapia, expressed in growth and feed utilization parameters.

Materials and methods

The experimental work of the present study was carried out at the Laboratory of Fish Nutrition,

Faculty of Agriculture, Benha University. The experiment started at 11 May 2009 and

continued until 11 September of the same year (120 days). The present study was designed to investigate the effect of four levels of copper oxychloride fungicide (0, 0.25, 0.50, 0.75 LC₅₀) on growth performance and feed utilization of Nile tilapia (*Oreochromis niloticus*). Three levels of vitamin C (0, 1 and 2g/ kg diet) were used with each level of the fungicide to determine the effect of vitamin C in reducing the harmful effect of this fungicide, therefore twelve treatments were included in the present study. Fish were obtained from Al-Abassa hatchery, Sharkia Governorate, Egypt. Fish were carefully transported in 50 liter plastic bags filled with water and oxygen to the laboratory, and after arrival to the laboratory fish were acclimatized to the laboratory conditions for one week in glass aquaria (180 L) filled with de-chlorinated water. Fish were fed daily a basal diet containing 30% crude protein (CP) and 3000 Kcal metabolizable energy (ME)/kg diet.

After acclimation period fish were randomly grouped into twelve groups and each group was arranged in two glass aquaria (two replicates for each treatment), therefore, twenty four

rectangular aquaria 100L ×40W ×50H, cm (180 liter of water for each) were used in this study.

The 96 h LC₅₀ value of copper oxychloride fungicide used in the present study was found to be 129.21 mg/L for red tilapia as reported by **Boock and Machado Neto (2000)**. Therefore, three sublethal concentrations of copper oxychloride were prepared (0.25, 0.50 and 0.75 LC₅₀ 96 h, i.e, 32.30, 64.60 and 96.90 mg copper oxychloride/L, respectively) and used in this study.

Stock solution of copper oxychloride was prepared by dissolving analytical grade copper oxychloride (from Merck) in distilled water. Desired concentrations of copper oxychloride were prepared from stock solution in distilled water. The level of LC₅₀ 96h of fungicide required for each aquarium (as mentioned in the experimental design) was adjusted and added carefully and well dissolved in the water at the start of the experiment. Each aquarium was stocked with 25 fish with an initial weight ranged between 1.85 and 1.94 g. Therefore, a total of 600 Nile tilapia (*Oreochromis niloticus*) fish were used in this study.

Table 1: Experimental design of different treatments of the present study.

<i>Treatment (T)</i>	<i>Fungicide, F(level of LC₅₀/96 h).</i>	<i>Vit. C, V(g /Kg diet).</i>	<i>Experimental diet.</i>
T ₁ (F1× V1)	0.00	0	D1
T ₂ (F1× V2)	0.00	1	D2
T ₃ (F1× V3)	0.00	2	D3
T ₄ (F2× V1)	0.25	0	D1
T ₅ (F2× V2)	0.25	1	D2
T ₆ (F2× V3)	0.25	2	D3
T ₇ (F3× V1)	0.50	0	D1
T ₈ (F3× V2)	0.50	1	D2
T ₉ (F3× V3)	0.50	2	D3
T ₁₀ (F4× V1)	0.75	0	D1
T ₁₁ (F4× V2)	0.75	1	D2
T ₁₂ (F4× V3)	0.75	2	D3

Fish were taken from each aquarium every week and the toxicant was renewed completely with fresh solution of the same concentration after cleaning and removing the accumulated excreta, finally fish were stocked again in the aquarium. Water temperature, pH and dissolved oxygen were measured daily at 2.00

pm, while total ammonia was weekly measured. Water quality parameters measured were found to be within acceptable limits for fish growth and health (**Boyd, 1979**).

The basal diet used in this study was formulated to contain 30%CP and 3000 Kcal ME/kg diet. Feed ingredients and proximate analysis

of the basal diet are presented in (Table 2). The basal diet was divided into three parts, the first was used as control (D1) while the second and third

parts were supplied by 1 and 2g vitamin C/kg diet representing diets D2 and D3, respectively.

Table 2: Feed ingredients and proximate analysis of the basal diet.

Ingredients	%
Fish meal (60%)	28
Soybean meal (44%)	18
Yellow corn	24
Wheat flour	13
Wheat bran	9
Corn oil	4
Vi t and Min.mix ¹	4
Total	100
Proximate analysis	
Dry matter (DM)	95.00
Crude protein (CP)	30.12
Ether extract (EE)	5.32
Crud fiber (CF)	5.21
Ash	8.45
Nitrogen free extract (NFE) ²	50.9
Metabolizable energy (ME),Kcal/Kg diet ³	3019
Protein /energy ratio (P/E ratio) ⁴	99.78

¹Each 1Kgf of vitamins and minerals mixture contains : Vitamins, D3, 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.

²NFE = 100 - (CP + EE + CF + Ash)

³Based on Kilo calorie values of 4.50/g protein, 8.51/g lipid and 3.9/g NFE (Jauncey, 1982).

⁴P/E ratio = mg protein/Kcal ME.

Therefore, fish of T₁, T₄, T₇ and T₁₀ were fed on the control diet (D1) and those of T₂, T₅, T₈ and T₁₁ were fed on (D2), whereas, fish of T₃, T₆, T₉ and T₁₂ were fed on (D3). Fish were given the experimental diets at a daily rate of 3% of total biomass six days/ week (twice daily) till the end of experimental period. Every two weeks, fish were taken from each aquarium, weighed and the amount of feed was adjusted according to the changes in body weight through the experimental period.

Records of body weight (BW) and body length (BL) of individual fish were measured for all fish of each aquarium at the start and the end of the experimental period. Other growth and feed utilization parameters were calculated as follows:

Weight gain (WG) = Final weight (g) - Initial weight (g).

Condition factor (K) = BW/(BL)³, Where: BW = Live body weight (g) and BL = body length (cm)

Specific growth rate (SGR) = [(lnW₂ - lnW₁)/T] × 100, Where: ln = The natural log; W₂ = Final fish weight at certain period (g); W₁ = Initial fish weight at the same period (g); T = Period (days).

Survival rate (SR%) = [(total number - dead number) / total number] × 100.

Feed conversion ratio (FCR) = Feed intake (g) / Weight gain (g).

Protein efficiency ratio (PER) = Weight gain (g) / CP intake (g).

Statistical analysis of data obtained was carried out by applying the computer program, SAS (2004). 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:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + e_{ijk}$$

Where: Y_{ijk} = the observation of the ith fish treated by the ith fungicide level and the jth vitamin level; μ = overall mean; α_i = the effect of ith fungicide level; β_j = the effect of jth vitamin level; $(\alpha\beta)_{ij}$ = the effect of the interaction between ith level of fungicide and jth

vitamin level; e_{ijk} = random error assumed to be independently and randomly distributed

$(0, \delta^2 e)$.

Results and discussion

Effect of fungicide (copperoxchloride) and vitamin levels and their interactions on growth performance of Nile Tilapia:

Body weight:

Results of the effect of fungicide (F) and vitamin C (vit. C) levels and their interactions on body weight (BW) of Nile tilapia are presented in Table (3). At the start of the experiment, the averages initial BW of fish ranged between 1.85 and 1.94 g with no significant differences among treatments groups. At the end of experiment (after 120 days), the highest final BW (27.33 g) was recorded by fish of T₁, while the lowest one (18.81 g) was shown by fish of T₁₂.

Results in Table (3) indicated that final BW of fish decreased with increasing the F level from 0 to 0.75 LC₅₀. Fish exposed to 0 F level recorded the highest final BW (25.70 g), while those treated with 0.75 LC₅₀ 96 h F level showed the lowest ($P < 0.05$) one being 20.26 g. However, there were no significant differences in final BW of fish treated with either 0.25 or 0.50 LC₅₀ F level and those of the control group 0 F as shown in (Table 3). Similar results were reported by (Sweilum, 2006) who found that final BW of Nile tilapia exhausted by sublethal doses of dimethoate (20, 10 and 5 mg/l) and malathion (2, 1 and 0.5 mg/l) decreased below the control values with increasing each pesticide concentration. Also, Mousa *et al.* (2005) reported that, Nile tilapia exposed to basegran herbicide at field concentration (0.004 mg/l = 1/100 LC₅₀) and ½ of field concentration showed significant ($P < 0.05$) reduction in final BW with increasing the level of pesticide compared with control fish. Similarly, Sweilum (2000) showed that increasing the concentration of pollution sewage water in fish ponds lead to decreased final BW of Nile tilapia.

Final BW of Nile tilapia fish varied slightly with vit. C level. Fish fed 1g vit.C/Kg diet surpassed slightly those fed either 0 or 2g vit.C/Kg diet. The differences in final BW of fish due to vit.C level effect were not significant (Table 3). In agreement with the previous results, Henrique *et al.* (1998) reported that, after 12 weeks of feeding, the mean final BW of the gilthead seabream fish

fed graded amounts of ascorbyl polyphosphate equivalent to 0, 25, 100, and 200mg of L-ascorbate /Kg diet were not significantly different. Similarly Ai *et al.*, (2006) reported that the final BW of large yellow coroher (*Pseudosciaena crocea*) increased with increasing dietary vit. C level 0.1 to 12.2, 23.8, 47.6 89.7, 188.5 and 489.0 mg ascorbic acid /Kg diet, but the differences were not significant. On contrast Adewolu and Aro (2009) reported that the final BW of (*Claris gariepinus*) significantly ($P < 0.05$) increased with increasing dietary vit.C level from 0 to 50, 100, 500, 1000 and 1500 mg L- ascorbic acid /Kg diet

Body length:

Results presented in (Table 3) showed that the differences in initial body length (BL) of fish among different treatments were insignificant indicating that the experimental groups at the start of the experiment were randomly distributed. At the end of the experiment, fish of T₉ showed the longest final BL (12.48cm), whereas those of T₁₂ recorded the shortest one (9.46 cm).

At the end of the experiment, fish exposed to 0.25 LC₅₀ F level showed the longest final BL (11.24 cm) while those treated with 0.75 LC₅₀ F level recorded the shortest one (9.74 cm). The differences in final BL of fish exposed to either 0 or 0.50 LC₅₀ F level and those of fish treated with 0.75 LC₅₀ F level were significant ($P < 0.05$). Whereas, no significant differences were detected in final BL between fish exposed to 0 F level and those of fish treated with either 0.25 or 0.50 LC₅₀ F level. Also, there were no significant differences in final BL between fish treated with 0.25 LC₅₀ F level and those of fish exposed to 0.75 LC₅₀ F level (Table 3). In accordance with the previous results Zaghoul (2001) cleared that BL of African catfish exposed to 0.35mg copper at 100 ppm CaCO₃ hardness was significantly ($P < 0.05$) decreased compared with the control. Also, Fivelstad *et al.* (2007) reported that the mean length for Atlantic salmon (*salmo salar* L.) exposed of 47 days to high carbon oxide level (12mm Hg) was significantly reduced ($P < 0.05$) compared to their respective control group (0.3mm Hg). Results in (Table 3) revealed that fish fed the diet supplemented with 2g vit.C/Kg diet

(V3) had somewhat longest final BL (11.06cm) than those of fish fed diets either with 0 vit. C or supplemented with 1g vit.C

/Kg diet (V2) being 10.39 and 10.48 cm, respectively and the differences were not significant.

Condition factor:

Condition factor (K) is frequently assumed to reflect not only characteristics of fish such as health, reproductive state and growth but also characteristics of environment such as habitat quality, water quality and prey availability (Liao *et al.*, 1995).

The highest K value (2.14) at the end of the experiment was shown by fish of T₁₀ and the lowest one (1.54) was recorded by fish of T₄ as shown in (Table 3). The previous results disagreed with those reported by Zaghoul (2001) who reported that, *Clarias gariepinus* fish exposed to 0.35mg Cu/l at 100 ppm Ca Co₃ hardness revealed a high significant decrease in K value compared with the control group. Also Ali *et al.*, (2003) indicated that sublethal concentrations of copper (0.15, 0.30 and 0.50 ppm) significantly (P<0.05) decreased as compared with the control and the decreased was linearly correlated with the increase of copper concentration. However, Fivelstad *et al.* 2007 claimed that at the end of exposure (47 days), K value of Atlantic salmon (*salmo salar L.*) of the high carbon oxide group (12mm Hg) was reduced compared to the control group (0.3mm Hg). Reduced K value observed in the high carbon oxide group to seems to be a typical long-term effect of carbon oxide on Atlantic salmon (Fivelstad *et al.*, 1999 and 2003). However, in studies performed at a lower carbon dioxide partial pressure compared to the present investigation, fish length was not significantly affected.

Results of the final K values presented in (Table 3) indicated that the both F and vit.C levels used in this study had no significant effect on final K value of Nile tilapia. Concerning the effect of vit.C on K value of Nile tilapia. Results obtained in the present study disagreed with those reported by Wang *et al.* (2003) who found that k value of Korean rockfish fed the vitamin C free diet was significantly (P<0.05) lower than that of fish fed vit.C supplemented containing equivalents of 50, 100 and 200 mg ascorbic acid /Kg diet for 12 weeks. Also, Kim *et al.* (2003) revealed that, after 10 weeks Nile tilapia fish fed the diet supplemented with 2000 mg ascorbic acid /Kg diet had significantly (P<0.05) higher K value than those fed the control diet.

Weight gain:

The highest WG (25.44 g) was recorded by fish of T₁ while the lowest ones (16.82 and

16.97 g) were shown by fish of T₁₁ and T₁₂, respectively as illustrated in (Table 4).

Fish exposed to 0 F showed the highest WG (24.01g) and those treated with 0.75 LC₅₀ recorded the lowest one (17.97g), indicating that WG decreased as the F level increased (Table 4). Differences in WG between fish exposed to 0.75 LC₅₀ F level and those for fish treated with all other F levels were significant (P<0.05). Whereas, no significant differences were observed in WG between fish exposed to other F levels. Similar results were reported by Zaghoul (2001) who found that (*Clarias gariepinus*) fish exposed to 0.35mg copper /L at 100 ppm Ca Co₃ hardness showed a highly significant decrease in body WG compared to the control group. Also, Mousa (2004), found significant reduction in WG of tilapia (monosex and mixed sex) and common carp exposed to 1/10 and 1/2 LC₅₀ of machete herbicide. Also, Mousa *et al.* (2005) indicated that, Nile tilapia fish exposed to basegran herbicide field concentration (0.004 mg/L= 1/100 LC₅₀) and 1/2 field concentration (0.002mg/L) showed significant (P<0.05) reduction in WG compared with control fish and the effect was dose depended.

Results in Table 4 showed that WG of fish fed 2g vit.C/Kg diet (V3) was slightly higher than that of fish fed either 0 or 1g vit.C /Kg diet V1 and V2 respectively and the differences were not significant. In partial agreement with the previous results, Chen *et al.* (2004) reported that there were no significant differences in average WG of golden shiners fish fed diets differing in ascorbic acid (23, 43, 98 or 222 mg /Kg diet) after 14 weeks. Also, Elizabeth *et al.* (2007) indicated that Juvenile matrinxa (*Brycon amazonicus*) fish fed 800 mg ascorbic acid /Kg diet recorded greater WG compared with that of fish fed 350, 400, 500, 600 or 800 mg ascorbic acid/Kg diet after 2 months of feeding. Similarly, Adewolu and Aro (2009) claimed that WG of (*Clarias gariepinus*), fingerlings fed diets with 0 of vit.C were significantly (P<0.05) lower than those fed on supplemented vit.C 50, 100, 150, 500, 1000 and 1500 mg /Kg diet.

Specific growth rate:

Fish of T₆ and T₂ recorded the highest SGR values being 2.34 and 2.32, respectively, while those of T₁, T₁₁ and T₁₂ showed the lowest ones (2.00, 2.03 and 2.03), respectively (Table 4).

As presented in Table(4) SGR values of fish exposed to 0, 0.25 and 0.50 LC₅₀ F levels were

nearly similar being 2.20, 2.23 and 2.24, respectively with no significant differences, whereas, fish treated with 0.75 LC₅₀ F level recorded the lowest (P<0.05) SGR value (2.08). The previous results are in agreement with those reported by **Abdel-Tawab et al. (2004)** who reported that SGR of Nile tilapia (*Oreochromis niloticus*) exposed to ¼ and ½ LC₅₀ of mercuric chloride was significantly (P<0.05) lower in comparison with the control. Similar results were obtained by **Mousa (2004)** who indicated that Nile tilapia (monosex and mixed sex) and common carp exposed to field concentration (0.004ppm) and 1/10 LC₅₀ of machete herbicide for 3 months showed significant (P<0.05) reduction in SGR values. In accordance with the previous results, **Mousa et al. (2005)** found that Nile tilapia exposed to field concentration (0.004 mg/L= 1/100 LC₅₀) and ½ field concentration (0.002mg /L) of basegran herbicide showed significant (P<0.05) reduction in SGR compared with control fish. Also, **Sweilum (2006)** showed that the exposure of Nile tilapia to sublethal dose of dimethoate (20,10 and 5 mg/ l) and malathion (2, 1 and 0.5 mg/l) decreased SGR value below levels for the control fish.

Results in (Table 4) showed that SGR values of Nile tilapia fish varied slightly according to vitamin C level applied being 2.13, 2.22 and 2.22 for fish fed 0, 1 and 2 g vit.C /Kg diet, respectively. The differences in SGR values attributed to vit.C level effect were not significant. Similar results were reported by **Thompson et al. (1993)** with Atlantic (*salmo salar L*). Similarly, **Henrique et al. (1998)** indicated that SGR of the Gilthead seabream, *Saparus aurata*, fed a basal diet supplemented with graded amounts of vit.C (25, 50, 100, and 200 mg of ascorbic acid /Kg diet for 12 weeks were not significant compared to fish fed vit. C free diet. Also, **Ai et al. (2006)** found that SGR of large yellow croaker (*Pseudosciaena crocea*) increased with increasing dietary vit. C level from 0.1 to 12.2, 23.8, 47.6, 89.7, 188.5 and 489 mg /Kg diet but the differences were not significant. In Contrarily, **Kim et al. (2003)** reported that fingerlings of Nile tilapia fed diet with 2000mg ascorbic acid had significantly (P<0.05) higher SGR values after 10 weeks of feeding than fish fed the control diets. Also, **Tewary and Patra (2008)** found that Indian major carp, rohu (*Labeo rohita*) fed vit C supplemented diets up to 1000 mg /Kg diet showed higher SGR compared with control fish.

Table (4): Effect of fungicide (Copperoxychloride) and vitamin C levels and their interactions on weight gain, specific growth rate and survival rate of Nile tilapia (*Oreochromis niloticus*).

Items	No.	Weight gain(g)	Specific growth rate	Survival rate%
Fungicide level (F):				
0.00 (F1)	6	24.01±0.85 a	2.20±0.03 a	94.67±2.17 a
0.25 (F2)	6	21.65±0.85 a	2.23±0.03 a	82.00±2.17 b
0.50 (F3)	6	21.37±0.85 a	2.24±0.03 a	74.00±2.17 c
0.75 (F4)	6	17.97±0.85b	2.08±0.03 b	74.00±2.17 c
Vitamin C level (V):				
0 (V1)	8	21.50±0.73	2.13±0.03	76.00±1.88 b
1 g/Kg (V2)	8	21.41±0.73	2.22±0.03	83.00±1.88 a
2 g/Kg (V3)	8	21.84±0.73	2.22±0.03	84.50±1.88 a
F × V:				
T ₁ (F1×V1)	2	25.44 ±1.47a	2.00±0.06 c	92.00±3.76
T ₂ (F1×V2)	2	24.00 ±1.47 ab	2.32±0.06 a	96.00±3.76
T ₃ (F1×V3)	2	22.60 ±1.47 ab	2.28±0.06 ab	96.00±3.76
T ₄ (F2×V1)	2	17.19 ±1.47 c	2.08±0.06 bc	80.00±3.76
T ₅ (F2×V2)	2	23.05 ±1.47 ab	2.28±0.06 ab	82.00±3.76
T ₆ (F2×V3)	2	24.17 ±1.47 ab	2.34±0.06 a	84.00±3.76
T ₇ (F3×V1)	2	20.61 ±1.47 abc	2.24±0.06 ab	68.00±3.76
T ₈ (F3×V2)	2	21.78 ±1.47 abc	2.25±0.06 ab	78.00±3.76
T ₉ (F3×V3)	2	21.73 ±1.47 abc	2.25±0.06 ab	76.00±3.76
T ₁₀ (F4×V1)	2	20.12 ±1.47 c	2.18±0.06 abc	64.00±3.76
T ₁₁ (F4×V2)	2	16.82 ±1.47 c	2.03±0.06 c	76.00±3.76
T ₁₂ (F4×V3)	2	16.97 ±1.47 c	2.03±0.06 c	82.00±3.76

Data are presented as means ±SE

Means followed by different letters in each column are significantly ($p < 0.05$) different

Survival rate:

Survival rate (SR) of Nile tilapia fish as affected by F and vit.C levels and their interactions are presented in (Table 4). Fish of T₂ and T₃ recorded the highest SR values (96.00 for both) whereas, those of T₁₀ showed the lowest one being 64.00. Results in (Table 4) showed that values of SR decreased with increasing the F level from 0 to 0.75 LC₅₀ regardless of vit.C level. Fish exposed to 0 fungicide showed the highest SR value (94.67 %), whereas, those treated with either 0.50 or 0.75 LC₅₀ F level recorded the lowest values being 74.00 % for both. Differences in SR values between fish exposed to 0 F level and those of fish treated with different F levels were significant ($P < 0.05$). Also the differences in SR values between fish treated with 0.25 LC₅₀ F level and those of fish exposed to either 0.50 or 0.75 LC₅₀ F levels were significant ($P < 0.05$). While, no significant differences were detected in SR values between fish treated with 0.50 LC₅₀ F level and those exposed to 0.75 LC₅₀ F level. The previous results are in partial agreement with those reported by **Abdel-Tawab (2004)** who reported that SR of Nile tilapia exposed to $\frac{1}{4}$ and $\frac{1}{2}$ LC₅₀ mercuric chloride was significantly ($P < 0.05$) decreased in comparison to the control. **Mousa et al. (2005)** indicated that the SR of Nile tilapia decreased significantly ($P < 0.05$) in the two group treated with basegran, field concentration (0.004 ppm) and 1/12 field concentration. Also, **Sweilum (2006)** concluded that SR of Nile tilapia was highly affected by sublethal pesticide concentration. The SR of fish gradually decreased with increased doses of dimethoate (20, 10 and 5 mg/l) and malathion (2, 1 and 0.5 mg/l)

The effect of increasing dietary vit.C on SR values of Nile tilapia fish irrespective of F treatment is presented in Table (4). Fish fed the diet free from vitamin C (V1) showed the lowest ($P < 0.05$) S R (76 %) value, whereas those fed 2 g vit.C /Kg diet (V3) had relatively higher SR value than those fed 1g vit.C /Kg diet (V2) being 84.50 and 83.00% respectively, but with no significant differences. In agreement with previous results, **Wang et al. (2003)** claimed that SR for juvenile Korean rock fish, *Sebastes schegeli* (Higendorf) fed the vit.C free diet for 12 weeks were significantly (0.05) lower than that

of fish fed vit.C supplemented diets with 0, 50, 100 and 200 mg ascorbic acid /Kg diet in either form of L-ascorbyl-2 glucose or L-ascorbyl-2-monophosphate-Na/Ca. Also, **Ai et al. (2004)** found that, after 8 weeks of feeding, SR of Japanese seabass, (*Lateolabrax japonicus*), was significantly ($P < 0.05$) with increasing dietary ascorbic acid (0.0, 12.2, 23.8, 47.6, 188.5 and 489.0 mg ascorbic acid /Kg diet) when supplementation of ascorbic acid was equal to more than 23.8 mg ascorbic acid /Kg diet, the SR was significantly ($P < 0.05$) higher than that of the control group, and no significant differences were observed among fish fed the diet with equal to or higher than 23.8 mg ascorbic acid /Kg diet. Similarly, **Ibrahim et al. (2010)** concluded that, SR % of Nile tilapia was significantly ($P < 0.05$) higher in fish fed the basal diet supplemented with vit.C (500 mg /Kg diet for one month than those fed the basal diet only (control) for one and two months. On the other hand, **Henrique et al. (1998)** reported that there were no significant differences in SR of seabream, (*Sparus aurata*), fed diets supplemented with graded amounts of ascorbic (0, 25, 50, 100 and 200 mg /Kg diet.).

Effect of Fungicide and Vitamin C Levels and Their Interactions on Feed utilization of Nile Tilapia.

Feed intake.

Feed intake (FI) values of the experimental fish as affected by F and vit.C levels and their interactions are presented in (Table 5). As shown in this table, the highest FI values were obtained by fish of T₁, T₃ and T₂ being 45.73, 45.60 and 45.42 g/fish, respectively, while the lowest value (28.44g) was recorded by fish of T₁₁.

Obtained data in Table(5) revealed that FI decreased almost in a linear manner with each increase in the F level being 45.46, 36.65, 35.39 and 32.98g for fish exposed to 0, 0.25, 0.50 and 0.75 LC₅₀ F levels, respectively. Difference in FI between fish exposed to 0 F level and those of fish treated with different F levels were significant ($P < 0.05$). Also, differences in FI between fish treated with 0.25 LC₅₀ F level and those of fish exposed to 0.75 LC₅₀ F level were significant ($P < 0.05$). Whereas, no significant difference were observed between fish exposed to 0.25 and 0.50 LC₅₀ F levels and between those treated with 0.50 and 0.75 LC₅₀ F levels. Results of

the present study are in full agreement with the previous results, **Ali et al. (2003)** who indicated that exposure of *Oreochromis niloticus* fish to different copper concentration (0.15, 0.30 and 0.50 ppm) significantly ($P<0.05$) reduced their FI as compared with the control fish. Similar results were obtained by **Sweilum (2006)** who reported that FI of Nile tilapia decreased below levels for the control fish with exposure to both dimethoate (20, 10 and 5 mg /L) and malathion (2.0, 1.0 and 0.5 mg/L) pesticide. Also, **Abdel-Tawab et al. (2007)** reported that FI of Nile tilapia was linearly decreased ($P<0.05$) with increasing copper concentration (0, 0.503 or 1.25 mg/L.).

With respect of dietary vit.C levels effect on FI of Nile tilapia fish, results of (Table 5) showed that FI varied slightly according to vit.C level, being 36.70, 37.30 and 36.70g for fish fed 0, 1 and 2g vit.C /Kg diet, respectively regardless of F levels, with no significant differences. These results are in agreement with those reported by **Chen et al. (2004)** who reported that there were no significant differences in FI of Golden shiners fish fed diets with different ascorbic acid levels (23, 43, 98 and 222 mg /Kg diet) after 14 weeks of the dietary trail. On contrary, **Adewolu and Aro (2009)** reported that FI of (*Clarias garpienus*) fingerlings linearly increased ($P<0.05$) with increasing dietary vit.C levels (0, 50, 100, 150, 500, 1000 and 1500 mg ascorbic acid /Kg diet.) in a 14 weeks feeding trial. Whereas, **Henrique et al. (1998)** found that voluntary FI of Seabream, (*Sparus aurata*), was significantly ($P<0.05$) lower for fish fed 200 mg ascorbic acid /Kg diet, when compared with the group fed 0 mg ascorbic acid /Kg diet.

Feed conversion ratio.

Results of (Table 5) show that the best (lowest) FCR (1.60) was recorded by fish of T₆ and the poorest (highest) ratio (2.10) was shown by fish of T₃.

Regardless of vit.C effect, fish exposed to 0.50 CL₅₀ F level showed the best FCR (1.66), followed by those treated by 0.25 and 0.75 LC₅₀ F levels being 1.70 and 1.72, respectively, whereas the poorest FCR (1.89) was recorded by fish exposed to 0 F level. It is clear that the fungicide level had no clear trend on FCR values of Nile tilapia. This may due to that F supplementation reduced feed intake with relatively sufficient total gain to record better FCR. Moreover the differences in FCR

values among all F levels used were not significant. In this concern **Ali et al. (2003)** reported that the exposure of *Oreochromis niloticus* fish to different copper concentrations (0.15, 0.30 and 5.0 ppm) increased ($P<0.05$) their FCR values in comparison with their respective controls. Also, **Abdel-Tawab (2004)** reported that FCR of Nile tilapia was linearly increased in fish exposed to $\frac{1}{4}$ and $\frac{1}{2}$ LC₅₀ of mercuric chloride. Fish exposed to the high dose of mercuric chloride ($\frac{1}{2}$ LC₅₀ recorded the highest (poorest) FCR value, while the best FCR one was obtained with control fish. Similarly, **Sweilum (2006)** reported that FCR increased above the optimum value of the control with chronic exposure to different concentrations of dimethoate (20,10 and 5 mg/ l) and malathion (2.0, 1.0 and 0.5 mg/l.).

FCR values varied slightly with dietary vit.C levels applied being 1.75, 1.74 and 1.73 for fish fed 0, 1 and 2 g vit.C/Kg diet, respectively regardless F level tested and the differences in FCR values due to vit.C level effect were not significant. In accordance with the pervious results, **Chen et al.(2004)** found that there were no significant ($P<0.05$) in FCR of Golden shiner fish fed diets different in ascorbic acid levels 23, 43, 98 and 222 mg /Kg diet) after 14 weeks. Whereas, **Adewolu and Aro (2009)** concluded that (*Calarias gariepinus*) fingerlings fed the diet without vit.C supplementation had higher FCR ($P<0.0$) compared with those fed diet containing vit.C supplements (50, 1200, 150, 500, 1000 and 1500 mg L- ascorbic acid/Kg diet). On the other hand, **Kim et al. (2003)** reported that, after 10 weeks of feeding, fish fed 2000 mg ascorbic acid /Kg diet had significantly ($P<0.05$) higher feed efficiency ratio than fish fed the control diet..

Protein efficiency ratio:

The effect of F and vit.C levels and their interactions on protein efficiency ratio (PER) values are illustrated in (Table 5). As shown in this table, fish of T₉ recorded the highest (best) PER value (2.04) while fish of T₃ showed the lowest (poorest) one being 1.65.

Results of PER values as affected by F level showed that PER values decreased with increasing the F level from 0 to 0.75 LC₅₀. Fish exposed to 0 F level showed the highest PER value (1.99), whereas, those treated with 0.75 LC₅₀ recorded the lowest one being 1.76 and the

differences in PER values due to F level effect were not significant as presented in Table (5). In partial agreement with the previous results, **Ali et al., (2003)** revealed that the PER value of Nile tilapia fish treated with different copper concentration (0.15, 0.30 and 5.0 ppm) were significantly ($P<0.05$) decreased as compared with the control. Similarly, **Abdel-Tawab et al. (2004)** indicated that PER was higher with the control fish, while the best one was obtained with fish exposed to high dose of mercuric chloride $\frac{1}{2}$ LC₅₀. Also, **Sweilum (2006)** indicated that PER values for Nile tilapia exposed to sublethal doses of dimethoate (20, 10 and 5 mg /l) and malathion (2.0, 1.0 and 0.5 mg /l) decreased below levels for the control.

With the respect the effect of dietary vit.C level on PER values of Nile tilapia fish, obtained results

show that fish fed 0, 1or 2g vit C /Kg diet recorded the same PER value being 1.91 indicating that dietary vit. C level had no effect on PER values. In partial agreement with the previous results, **Chen et al. (2004)** indicated that the dietary vit.C levels (23, 43, 98 or 222 mg ascorbic acid had no significant effect on PER values of juvenile Golden shiners. Also, **Wang et al. (2003)** claimed that the PER of juvenile Korean rock fish fed vit.C free diet were significantly ($P<0.05$) lower than those of fish fed diets supplemented with 50, 100 and 200 mg ascorbic acid /Kg diet either in the form L- ascorbyl-2- glucose or L- ascorbyl-2- monophosphate-Na Ca. Similarly, **Adewolu and Aro (2009)**, reported that PER values for (*Clarias gariepinus*) fingerlings fed the diets without vit.C supplementation were the lowest compared with other diets supplemented with vit. C (50, 100, 150, 500, 1000 and 1500 mg ascorbic acid /Kg feed).

Table (5): Effect of increasing levels of fungicide (Copperoxychloride) and vitamin C and their interactions on feed intake, feed conversion ratio and protein efficiency ratio of Nile tilapia.

Items	No.	Feed intake (g)	Feed conversion ratio (g feed /g gain)	Protein efficiency ratio(g gain /g cp intake)
Fungicide (F):				
0.00 (F1)	6	45.46±1.11 a	1.89±0.06	1.99±0.07
0.25 (F2)	6	36.65±1.11 b	1.70±0.06	1.92±0.07
0.50 (F3)	6	35.39±1.11 bc	1.66±0.06	1.90±0.07
0.75 (F4)	6	32.98±1.11 c	1.72±0.06	1.76±0.07
Vitamin C level(V):				
0 (V1)	8	36.70±0.96	1.75±0.05	1.91±0.06
1 g/Kg (V2)	8	37.30±0.96	1.74±0.05	1.91±0.06
2 g/Kg (V3)	8	36.70±0.96	1.73±0.05	1.91±0.06
F × V:				
T ₁ (F1×V1)	2	45.73±1.93 a	1.78±0.10	1.87±0.12
T ₂ (F1×V2)	2	45.42±1.93 a	1.89±0.10	1.76±0.12
T ₃ (F1×V3)	2	45.60±1.93 a	2.10±0.10	1.65±0.12
T ₄ (F2×V1)	2	30.90±1.93 cd	1.79±0.10	1.83±0.12
T ₅ (F2×V2)	2	39.28±1.93 ab	1.71±0.10	1.93±0.12
T ₆ (F2×V3)	2	39.78±1.93 ab	1.60±0.10	2.02±0.12
T ₇ (F3×V1)	2	34.68±1.93 bcd	1.68±0.10	1.95±0.12
T ₈ (F3×V2)	2	36.09±1.93 bc	1.67±0.10	2.00±0.12
T ₉ (F3×V3)	2	35.34±1.93 bc	1.64±0.10	2.04±0.12
T ₁₀ (F4×V1)	2	35.85±1.93 bc	1.78±0.10	1.84±0.12
T ₁₁ (F4×V2)	2	28.44±1.93 d	1.70±0.10	1.93±0.12
T ₁₂ (F4×V3)	2	34.60±1.93 cd	1.69±0.10	1.93±0.12

Data are presented as means ± SE.

Means followed by different letters in each column are significantly ($P<0.05$) different

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تأثير المبيد الفطري أوكسي كلورو النحاس وفيتامين ج على أداء النمو والاستفادة من مركبات الغذاء لأسماك البلطي النيلي
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الملخص العربي

صممت هذه الدراسة لبيان تأثير أربع مستويات من المبيد الفطري أكسي كلورو النحاس (٠.٢٥ و ٠.٥٠ و ٠.٧٥ و ٠.٩٥ للجرعة النصف المميطة من المبيد الفطري) على صفات النمو والاستفادة من الغذاء لأسماك البلطي النيلي، كذلك اختبرت ثلاث مستويات من فيتامين ج (٠ و ١ جم /كجم عليقه) مع كل مستوى من المبيد الفطري لدراسة تأثير فيتامين ج على تخفيف التأثير الضار للمبيد الفطري، ولذلك تم استخدام ١٢ معاملة في هذه الدراسة والتي استمرت لمدة ١٢٠ يوماً. عند نهاية التجربة أظهرت النتائج أن مستويات المبيد الفطري المستخدمة في هذه الدراسة كان لها تأثير عكسي على كل صفات النمو المدروسة في سمك البلطي النيلي فقد أوضحت النتائج أن كل من وزن الجسم النهائي، وطول الجسم النهائي، الوزن المكتسب، ومعدل النمو النسبي وكذلك معدل البقاء انخفضت (غالباً معنوياً) بزيادة مستوى المبيد الفطري المستخدم، بينما أظهر استخدام المستويات المختلفة من فيتامين ج اتجاهها إلى زيادة قيم كل الصفات السابقة ولكن لم تكن هذه الزيادة معنوية إلا بالنسبة لصفة معدل البقاء الذى زاد معنوياً بزيادة مستويات فيتامين ج المستخدم انخفضت كل صفات الاستفادة من الغذاء (كمية الغذاء المأكل، معدل تحويل الغذاء، كفاءة استخدام البروتين) لأسماك البلطي النيلي بزيادة مستوى المبيد الفطري المستخدم، ولم يكن هذا التأثير معنوياً إلا في حالة كمية الغذاء المأكل، بينما أظهرت النتائج أن المستويات المختلفة لفيتامين ج المستخدم في هذه الدراسة لم يكن لها أى تأثير معنوي على كل صفات الاستفادة من الغذاء بالنسبة لأسماك البلطي النيلي.

Table (3): Effect of fungicide(Copper oxychloride) and vitamin C levels and their interactions on body weight, body length and condition factor of Nile tilapia (*Oreochromis niloticus*).

No.	Body weight (g)		Body length (cm)				Condition factor			
	Initial	No.	Final	No.	Initial	No.	Final	No.	Initial	No.
150	1.90±0.05	142	25.70±0.92 a	150	3.75±0.05	142	10.87±0.32 a	150	4.09±0.10	142
150	1.87±0.05	123	23.66±1.03 a	150	3.77±0.05	123	10.61±0.29 ab	150	3.65±0.10	123
150	1.88±0.05	114	24.89±0.99 a	150	3.86±0.05	114	11.24±0.3 a	150	3.46±0.10	114
150	1.92±0.05	111	20.26±1.04 b	150	3.76±0.05	111	9.74±0.33 b	150	3.52±0.10	111
200	1.90±0.04	152	22.98±0.84	200	3.73±0.04	152	10.39±0.28	200	3.94±0.08	152
200	1.90±0.04	169	24.36±0.84	200	3.81±0.04	169	10.48±0.27	200	3.56±0.08	169
200	1.89±0.04	169	23.89±0.89	200	3.81±0.04	169	11.06±0.27	200	3.53±0.08	169
50	1.92±0.11	46	27.33±1.61 a	50	3.72±0.09	46	10.97±0.51	50	4.06±0.17	46
50	1.90±0.11	48	25.32±1.59 ab	50	3.77±0.09	48	10.46±0.50	50	4.10±0.17	48
50	1.88±0.11	48	24.53±1.59 ab	50	3.74±0.09	48	11.20±0.50	50	4.10±0.17	48
50	1.85±0.11	40	18.99±1.73 c	50	3.67±0.09	40	10.08±0.05	50	4.10±0.17	40
50	1.88±0.11	41	25.12±1.71 ab	50	3.84±0.09	41	10.73±0.54	50	3.40±0.17	41
50	1.88±0.11	42	26.00±1.69 a	50	3.81±0.09	42	11.00±0.54	50	3.46±0.17	42
50	1.88±0.11	34	22.43±1.88 abc	50	3.85±0.09	34	10.25±0.6	50	3.90±0.17	34
50	1.88±0.11	42	25.36±1.69 a	50	3.85±0.09	42	10.90±0.54	50	3.20±0.17	42
50	1.89±0.11	38	25.47±1.77 ab	50	3.88±0.09	38	12.48±0.56	50	3.80±0.17	38
50	1.94±0.11	32	22.31±1.93 abc	50	3.68±0.09	32	10.09±0.61	50	3.70±0.17	32
50	1.91±0.11	38	20.10±1.77 bc	50	3.79±0.09	38	9.74±0.56	50	3.55±0.17	38
50	1.90±0.11	41	18.81±1.71 c	50	3.82±0.09	41	9.46±0.56	50	3.31±0.17	41

Data are presented as means \pm SE.

Means followed by different letters in each column are significantly ($P < 0.05$) different.