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## GROWTH PERFORMANCE, FEED EFFICIENCY AND BLOOD PARAMETERS IN NILE TILAPIA EXPOSED TO MERCURY TOXICITY AND ITS REDUCTION BY USING DIETARY SUPPLEMENTATION OF ESSENTIAL OILS

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**ABSTRACT:** A 112-day feeding trial was performed to investigate the toxic effects of mercury on Nile tilapia (*Oreochromis niloticus*) and attempt to detoxify these drastic effects by using dietary supplementation of citronella (*Cymbopogon nardus*) and geranium (*Pelargonium graveolens*) (GEO) essential oils. Fish were divided into four groups, each group was stocked into 3 aquaria, each one contains 10 fish. The first group was fed on basal diet without mercury, the second group was fed on basal diet containing 50 ppm mercury as mercuric chloride (HgCl<sub>2</sub>), each of third and fourth group was treated with 50 ppm HgCl<sub>2</sub> and supplemented with 400 mg/kg diet citronella or geranium oils, respectively. Live body weight at 16 week was significantly increased by 40.76%, 22.64 and 12.41%, respectively in each of fish group fed diets without mercury (basal diet), fish group fed a diet contaminated with mercury and supplemented with citronella oil and fish group fed a diet contaminated with mercury and supplemented with geranium oil when compared with those fed diet contaminated with mercury. Also, daily weight gain at 0-16 weeks significantly increased by 51.38, 28.44 and 15.60%, respectively. Feed conversion at 0-16 weeks significantly improved by 29.56, 16.92 and 14.27%, in fish group fed diets without mercury (basal diet), fish group fed a diet contaminated with mercury and supplemented with citronella oil and fish group fed a diet contaminated with mercury and supplemented with geranium oil, respectively, when compared with those fed diet contaminated with mercury. Serum total protein and albumin significantly increased, while serum ALT, AST, urea-N and Creatinine were decreased in fish groups fed diets without mercury. Also serum total protein increased in fish groups fed diet contaminated with mercury and treated with citronella or geranium oil than those fed diet contaminated with mercury. Residual of mercury significantly decreased (P<0.001) by 29.53 and 31.49 in liver and muscles of fish group fed a diet contaminated with mercury and supplemented with citronella oil, respectively. Also in fish group fed a diet contaminated with mercury and supplemented with geranium oil decreased by 41.45 and 47.24, respectively. Based on the obtained results it could be concluded that, growth rate and feed conversion improved by dietary essential oils supplementation.

**Key words:** Mercury, essential oils, growth rate, feed efficiency, Nile tilapia, amelioration.

### INTRODUCTION

Fish can be used as choosy bio-indicators of trace metals in freshwater reservoir since they not only accumulate some heavy metals in their bodies but also react to water contamination with alteration of various vital functions

(Dobrowolski and Skowrońska, 2006). Decrease in growth rate of fish can be caused as a result of physiological or behavioral stress during exposure to toxicants (Hansen *et al.* 2002b; Ayyat *et al.*, 2017). Fish growth depends on water quality characteristics and in polluted waters generally decreases (Hansen *et al.*, 2002a).

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Mercury (Hg) considers as one of the most dangerous toxic metals especially in the aquatic situation (Asefi and Zamani-Ahmadmoodi, 2015). Mercury can be found in 3 major forms, namely elemental Hg, inorganic Hg, and organic Hg (Looi *et al.*, 2016). There was a great attentiveness on the effects of mercury pollution on human and environmental health. However, the environmental troubles caused by the mercury contamination seems to be a continual bout since mercury-pollution has been occurring for years, and most probably will continue in the upcoming years. The agricultural drainage water containing heavy metals, inorganic anions, pesticides, fertilizers and industrial by-products in addition to sewage effluents supply the water bodies and sediment with huge quantities of heavy metals (ECDG, 2002).

Using essential oils (EOs) as feed additives in aquaculture due to its valuable role in promote the growth and increasing fish immune system (Zheng *et al.*, 2009). The main compensation of essential oils are condensed aquatic pollution, lesser toxicity, fewer toxic waste in fish tissues, compact risk of selecting resistant pathogens and less cost to the farmers (Coimbra *et al.*, 2006).

Citronella or *Cymbopogon nardus* is one of the *Cymbopogon* species with its essential oil widely used in the production of citronella essential oil, food, drink, perfumery, soap, body care products and pharmaceutical products. Billerbeck *et al.* (2011) reported that essential oil of *C. nardus* at dose of 400 mg/l could inhibit 80% of *Aspergillus niger* growth. Meanwhile, Oussalah *et al.* (2006) reported that the essential oil showed antimicrobial activity at dose of 4 mg/ml against *Pseudomonas putida*. Lee and Wendy (2013) reported that essential oil of *C. nardus* demonstrated it's possible as alternative to commercial antibacterial agent.

Pelargoniums are a diverse group of plants with a wide variety of growth habits and habitats. Most are native to southern Africa, but a few species occur naturally in Middle East (The Herb Society of America, 2006). Hamidpour *et al.* (2017) reported that *Pelargonium graveolens*, rose geranium, has shown multiple positive benefits. Its antibacterial and anti-fungal abilities show strong potential to replace current

therapeutic drug regimes as there is an increase in drug resistance microbes. Toxicity is a concern and should be monitored as further research progresses. However, on all fronts, *Pelargonium graveolens* shows great potential for a traditional solution in today's world as a preservative and, more eminently, a therapeutic agent.

Reasonably, contamination of the aquatic environment by mercury has been considered a major danger to the fish. Therefore, this study was carried out to investigate the toxic effects of mercury on Nile tilapia (*Oreochromis niloticus*) and attempt to detoxify these drastic effects by using dietary supplementation of essential oils; citronella (*Cymbopogon nardus*) and geranium (*Pelargonium graveolens*).

## MATERIALS AND METHODS

This study was conducted at the Department of Animal Production, Agriculture Faculty, Zagazig University and the practical work was carried out at Central Laboratory for Aquaculture Research (CLAR), Abbassa, Sharkia Governorate, Egypt.

Fingerlings Nile tilapia fish (*Oreochromis niloticus*) averaged about  $6.252 \pm 0.028$  g was used in this study. The fish were stocked in twelve glass aquaria ( $70 \times 40 \times 50$  cm) supplied with fresh aerated tap water. Fish were acclimated to laboratory conditions for 2 weeks and fed a basal diet prepared without feed additives till the beginning of the experiment. Continuous aeration was provided to each tank through an air stone connected to a central air compressor. Day after day, each aquarium was cleaned from the fish faeces, and the water was partially changed (about 30%). The photoperiod was 12-hr light: 12-hr darkness. The mean of water temperature, dissolved oxygen and pH was  $29.31 \pm 0.129^\circ\text{C}$ ,  $7.18 \pm 0.039$  mg l<sup>-1</sup> and  $6.66 \pm 0.022$ , respectively. The average of ammonia, nitrite and nitrate was  $0.17 \pm 0.014$ ,  $0.12 \pm 0.017$  and  $2.00 \pm 0.083$ , mg l<sup>-1</sup>, respectively.

Fish were divided into four groups, each group of fish was stocked into 3 aquaria and each one contains 10 fish. The first group was given basal diet without mercury, the second group was given basal diets containing 50 ppm

mercury as mercuric chloride ( $\text{HgCl}_2$ ), the third group was treated with mercury and supplemented with 400 mg citronella oil/kg diet, the fourth group was treated with mercury and supplemented with 400 mg geranium oil/kg diet. Continuous aeration was provided to each tank through an air stone connected to a central air compressor. The fish were fed on the experimental diets at the rate of 5% of body weight at the first 5 weeks of the experimental period, the remaining experimental period (5-16 weeks) fish were fed at the rate 3% of body weight and the experimental diets were offered three times daily at 9:00 am, 12:00 and 16:00 pm. Along the feeding trial, the uneaten feed was collected by siphoning. The amount of feed was readjusted every 2 weeks according to the biomass of each replicate.

All fish groups were fed on basal pelleted diet consistent of fish meal 16%, soybean meal 35.0%, corn 25%, wheat bran 17.0%, oil 2%, minerals mixture 2%, vitamin mixture 1.0% and carboxymethyl cellulose 2.0%. The chemical composition of the diet was crude protein 30.8%, ether extract 5.89%, crude fiber 4.6% and gross energy 4069 Kcal/kg diet.

All fish were weighed to the nearest 0.1 g at the beginning of the experiment and biweekly intervals throughout the experimental period. Food consumption was calculated as g/fish/day by dividing the amount of food consumed each day by the number of fish in the aquarium. Feed conversion ratio (FCR) was calculated according to **Berger and Halver (1987)** according to the following equation:  $\text{FCR} = \text{cumulative feed delivered to aquarium/fish biomass gain}$ .

Blood samples were taken from the caudal vein from five fish in each group which were randomly selected for collecting blood samples at the end of the experimental period. The blood samples were centrifuged at 3000 rpm for 20 min to separate the serum. Total protein, albumin, urea-N, creatinine, and serum transaminase enzymes (AST, aspartate aminotransferase and ALT, alanine aminotransferase) were determined in the blood serum by colorimetric methods using commercial kits (**Henry, 1974**).

Proximate chemical compositions of experimental diets were determined according to

the Association of Official Analytical Chemists (**AOAC, 1990**).

The heavy metals of both fish muscles and liver were calculated by atomic absorption apparatus according to method described by **AOAC (1990)**. Muscles and liver organs were collected separately and dried in an oven at  $105^\circ\text{C}$  for 24 hours to a constant weight, and then pieces of 5 g (dry weight) from muscle tissues and 1 g from liver tissues were ashed at  $550^\circ\text{C}$  in the muffle furnace. After cooling, the samples were digested with 2 ml concentrated  $\text{HNO}_3$ . This treatment is repeated, if necessary, to obtain clean practically C-free ash. Finally, the ash is dissolved by 10 and completed to 25 ml with 1 N HCL or distilled water and preserved in fridge till analysis in atomic absorption apparatus according to method described by **Meltem et al. (2007)**.

The data were statistically analyzed by completely randomized design with **SAS (2002)** in relation to the following model:  $Y_{ij} = \mu + T_i + E_{ij}$ . Where  $\mu$  is the overall mean,  $T_i$  is the fixed effect of  $i^{\text{th}}$  treatments, and  $E_{ij}$  is the random error. Means were tested for significant differences using Duncan's Multiple Range test (**Duncan, 1955**).

## RESULTS AND DISCUSSION

### Growth Performance

The insignificant differences among the experimental groups for initial live body weight showed that the groups at the start of the experiment were homogenous.

Live body weight at 16 week were significantly ( $P < 0.01$ ) affected with mercury contamination, where it significantly increased by 40.76%, 22.64 and 12.41%, respectively in fish group fed diet without mercury (basal diet), fish group fed diet contaminated with mercury and supplemented with citronella oil and fish group fed diet contaminated with mercury and supplemented with geranium oil when compared with those fed diet contaminated with mercury. On the other hand, live body weight of fish at 8 week of the experimental period was insignificantly affected by the treatments (Table 1).

**Table 1. Live body weight (g) and daily weight gain (g/day) of Nile tilapia fish as affected by dietary mercury contamination and their amelioration**

Treatment	Body weight (g)			Daily gain (g/day)		
	0 Week	8 Week	16 Week	0-8 Week	8-16 Week	0-16 Week
T1	6.225±0.017	18.480±0.449	43.133±2.194 <sup>a</sup>	0.219±0.008	0.440±0.038 <sup>a</sup>	0.330±0.020 <sup>a</sup>
T2	6.268±0.010	17.787±0.283	30.642±0.655 <sup>c</sup>	0.206±0.005	0.230±0.008 <sup>c</sup>	0.218±0.006 <sup>c</sup>
T3	6.271±0.013	18.732±0.254	37.580±1.224 <sup>b</sup>	0.222±0.004	0.337±0.017 <sup>b</sup>	0.280±0.011 <sup>b</sup>
T4	6.249±0.018	17.710±0.41	34.446±0.734 <sup>bc</sup>	0.205±0.007	0.299±0.008 <sup>bc</sup>	0.252±0.006 <sup>bc</sup>
<b>Significance</b>	NS	NS	**	NS	***	**

NS = Not significant, \*\*\* P<0.001 and \*\* P<0.01.

Means in the same column with different letters differ significantly (P<0.05).

T<sub>1</sub> Control group; fish were fed the basal diet.

T<sub>2</sub> Negative control group; fish were fed a diet contaminated with 50 ppm mercury.

T<sub>3</sub> Fish were fed a diet contaminated with 50 ppm mercury and supplemented with 400 mg citronella oil/kg diet.

T<sub>4</sub> Fish were fed a diet contaminated with 50 ppm mercury and supplemented with 400 mg geranium oil/kg diet.

Daily weight gain at 8-16 and 0-16 week of the experimental period were significantly (P < 0.001 and 0.01, respectively) affected with mercury contamination. Daily weight gain at 8-16 week increased by 91.30, 46.52 and 30.00% in fish group fed diets without mercury (basal diet), fish group fed diet contaminated with mercury and supplemented with citronella oil and fish group fed diet contaminated with mercury and supplemented with geranium oil, respectively, when compared with those fed diet contaminated with mercury. The same figures for 0-16 weeks were 51.38, 28.44 and 15.60%, respectively (Table 1). On the other hand, daily body gain of fish at 0-8 week of the experimental period insignificantly affected with treatments.

The present results were in good agreement with those obtained by **Sivakami *et al.* (1995)** who reported that exposure fish to mercury significantly decreased growth rate. Moreover, the growth depression in fish in the present study may be due to the toxicity of Hg through the production of superoxide radicals and glutathione enzyme depletion (**Miura *et al.*, 1995; Agarwal *et al.*, 2010**). Decreased growth rate may be due to the use of body energy for repairing damaged cells which may lower the somatic and reproductive growth (**Houck and Cech, 2004**).

Relative growth rate at 8-16 and 0-16 week of the experimental period were significantly (P<0.001 and 0.01, respectively) affected with mercury contamination, while, relative growth rate at 0-8 week of the experimental period was insignificantly affected (Table 2). Relative growth rate at 0-16 week increased by 13.12, 8.09 and 4.93% in fish group fed diets without mercury (basal diet), fish group fed a diet contaminated with mercury and supplemented with citronella oil and fish group fed a diet contaminated with mercury and supplemented with geranium oil, respectively, when compared with those fed diet contaminated with mercury.

Mercury is a sculpture-seeking metal that bind to –SCH<sub>3</sub> and –SH groups present in methionine and cysteine. These amino acids are part of the enzyme structure. Often, the sulphhydryl (–SH) groups are found on enzyme active site. In such circumstance, attachment of Hg<sup>2+</sup> on the –SH group would indeed be detrimental to the activities of the enzyme (**Manahan, 1979**). Metal ions have been found to interact with cell components such as DNA and nuclear proteins, causing DNA damage and conformational changes that may lead to cell cycle modulation, carcinogenesis or apoptosis (**Wang and Shi, 2001; Beyersmann and Hartwig, 2008**).

**Table 2. Relative growth rate (g gain/100 g body weight) and mortality rate (%) of Nile tilapia fish as affected by dietary mercury contamination and their amelioration**

Treatment	Relative growth rate			Mortality rate		
	0-8 Week	8-16 Week	0-16 Week	0-8 Week	8-16 Week	0-16 Week
T1	99.152±1.725	79.783±4.097 <sup>a</sup>	149.358±2.223 <sup>a</sup>	0±0	0±0	0±0
T2	95.749±1.182	53.065±1.199 <sup>c</sup>	132.036±1.124 <sup>c</sup>	6.667±3.333	3.333±3.333	10±0
T3	99.663±0.909	66.853±1.673 <sup>b</sup>	142.716±1.484 <sup>b</sup>	0±0	3.333±3.333	3.333±3.333
T4	95.614±1.721	64.183±1.060 <sup>b</sup>	138.544±0.981 <sup>b</sup>	3.333±3.333	0±0	3.333±3.333
Significance	NS	***	***	NS	NS	NS

NS = Not significant and \*\*\* P<0.001.

Means in the same column with different letters differ significantly (P<0.05).

Essential oils have several modes of actions as antioxidant, such as prevention of chain initiation, free radical scavengers, reducing agents, termination of peroxides, prevention of continued hydrogen abstraction as well as quenchers of singlet oxygen formation and binding of transition metal ion catalysts (Yildirim *et al.*, 2000; Mao *et al.*, 2006). The antioxidant capability of phenolic compounds is mainly due to their redox properties, which permit them to act as hydrogen donors, reducing agents, singlet oxygen quenchers as well as metal chelators (Kumar *et al.*, 2005).

Geranium essential oil (GEO) is obtained from the scented leaves of rose-scented geranium and has been known for diverse biological and pharmacological properties such as antimicrobial (Lis-Balchin and Deans, 1996), anti-inflammatory (Boukhatem *et al.*, 2013), hypoglycaemic and antioxidant (Boukhris *et al.*, 2012).

### Mortality Rate

Results presented in Table 2 show that mortality rate insignificantly differed among treatments. Fish group fed diet contaminated with mercury recorded higher mortality rate during the whole experimental period, on the other hand, fish fed diet without mercury (basal diet) recorded lower mortality rate.

### Feed Efficiency

Results presented in Table 3 show that feed intake caused insignificant differences between treatments. Daily feed intake during 0-16 weeks increased by 6.09 and 6.49% in fish group fed

basal diet and fish group fed a diet contaminated with mercury and supplemented with citronella oil, when compared with those fed diet contaminated with mercury. On the other hand fish group fed a diet contaminated with mercury and supplemented with geranium oil recorded lower feed intake.

Feed conversion at each of 8-16 and 0-16 week of the experimental period was significantly (P<0.001) affected with mercury contamination, while, at 0-8 week of the experimental period was insignificantly affected (Table 3). Feed conversion at 0-16 week improved by 29.56, 16.92 and 14.27% in fish group fed diet without mercury (basal diet), fish group fed diet contaminated with mercury and supplemented with citronella oil and fish group fed diet contaminated with mercury and supplemented with geranium oil, respectively, when compared with those fed diet contaminated with mercury.

The present results were in good agreement with those obtained by Sivakami *et al.* (1995) who reported that exposure of fish to mercury significantly caused deterioration of feed conversion.

### Serum Biochemical Parameters

Serum biochemical parameters were significantly (P<0.001, 0.01 or 0.05) affected with mercury contamination (Tables 4 and 5). Serum total protein, albumin and globulin were increased in fish group fed control diet (without mercury contamination) and fish group fed diet

**Table 3. Daily feed intake (g) and feed conversion (g food/g gain) of Nile tilapia fish as affected by dietary mercury contamination and their amelioration**

Treatment	Daily feed intake (g)			Feed conversion (g food/g gain)		
	0-8 Week	8-16 Week	0-16 Week	0-8 Week	8-16 Week	0-16 Week
T1	0.296±0.004	0.296±0.004	0.523±0.0095	1.356±0.040	1.722±0.119 <sup>c</sup>	1.594±0.074 <sup>c</sup>
T2	0.289±0.004	0.698±0.016	0.493 ±0.010	1.404±0.014	3.042±0.088 <sup>a</sup>	2.263±0.032 <sup>a</sup>
T3	0.300±0.004	0.749±0.011	0.525 ±0.007	1.350±0.008	2.234 ±0.089 <sup>b</sup>	1.880±0.049 <sup>b</sup>
T4	0.289±0.006	0.687±0.040	0.488± 0.022	1.414±0.026	2.300±0.129 <sup>b</sup>	1.940± 0.061 <sup>b</sup>
Significance	NS	NS	NS	NS	***	***

NS = Not significant and \*\*\* P<0.001.

Means in the same column with different letters differ significantly (P<0.05).

**Table 4. Blood total protein and its fractions of Nile tilapia fish as affected by dietary mercury contamination and their amelioration.**

Treatment	Total protein (g/100ml)	Albumin (g/100ml)	Globulin (g/100l)	Albumin : Globuin Ratio
T1	5.680±0.012 <sup>a</sup>	3.510±0.006 <sup>a</sup>	2.170±0.006 <sup>a</sup>	1.618±0.001 <sup>b</sup>
T2	4.713±0.152 <sup>c</sup>	3.140±0.012 <sup>d</sup>	1.573±0.155 <sup>b</sup>	2.033±0.193 <sup>a</sup>
T3	5.350±0.029 <sup>b</sup>	3.240±0.023 <sup>b</sup>	2.110±0.006 <sup>a</sup>	1.536±0.007 <sup>b</sup>
T4	5.280±0.017 <sup>b</sup>	3.190± 0.006 <sup>c</sup>	2.090±0.023 <sup>a</sup>	1.527±0.020 <sup>b</sup>
Significance	***	***	**	*

\*\*\* P<0.001, \*\* P<0.01 and \* P<0.05

Means in the same column with different letters differ significantly (P<0.05).

**Table 5. Serum transaminase enzymes (AST and ALT), alkaline phosphatase (ALP), creatinine and urea-N of Nile tilapia fish as affected by dietary mercury contamination and their amelioration**

Treatment	AST(IU)	ALT(IU)	Creatinine (mg/100ml)	Urea-N (mg/100ml)	ALP
T1	20.133±0.291 <sup>c</sup>	14.600±0.346 <sup>c</sup>	0.217±0.007 <sup>b</sup>	12.267±0.088 <sup>c</sup>	33.167±0.713 <sup>a</sup>
T2	24.233±0.233 <sup>a</sup>	24.733±0.296 <sup>a</sup>	0.540±0.025 <sup>a</sup>	18.767±0.333 <sup>a</sup>	17.733±1.087 <sup>d</sup>
T3	22.400±0.115 <sup>b</sup>	17.667±0.371 <sup>b</sup>	0.300±0.038 <sup>b</sup>	15.967±0.463 <sup>b</sup>	22.833±0.524 <sup>c</sup>
T4	21.467±0.536 <sup>b</sup>	16.967±0.120 <sup>b</sup>	0.280± 0.017 <sup>b</sup>	15.433 ±0.203 <sup>b</sup>	27.000 ±0.404 <sup>b</sup>
Significance	***	***	***	***	***

\*\*\* P<0.001.

Means in the same column with different letters differ significantly (P<0.05).

contaminated with mercury and treated with citronella oil followed by fish group fed diet contaminated with mercury and treated with geranium oil, respectively while serum albumin/globulin ratio decreased when compared with fish groups fed diets contaminated with mercury (Table 4).

Serum ALT, AST, urea-N and creatinine significant increased in fish group fed diet contaminated with mercury (Table 5). These findings agreed with those found by **Abdel-Tawwab et al. (2004)** who found that AST, ALT and ALP activities were decreased on Nile tilapia exposed to inorganic mercury.

Also **Gill et al. (1990)** who found a marked reduction in hepatic, branchial and renal AST and ALT in rosy barb (*Puntius conchoni*) after toxication with mercuric chloride. They mentioned that, the reduced levels of aminotransferase in various organs may result from tissue damage and consequently the reduction of enzyme turnover causally related to the presence of toxic mercury.

### Mercury Residual in Fish Body

The bioaccumulation of mercury in liver and muscles of fish were measured at the end of the experimental period. Residual of mercury significantly decreased ( $P < 0.001$ ) by 29.53 and 31.49 in the liver and muscles of fish group fed diet contaminated with mercury and supplemented with citronella oil, respectively. Also in fish group fed diet contaminated with mercury and supplemented with geranium oil decreased by 41.45 and 47.24, respectively (Table 6).

Similar finding was also demonstrated in Hg contaminated fish *Gymnotus carapo*, after acute

exposure to  $Hg^{+2}$ ; the highest mercury level was found in the liver, followed by the gills and lowest concentration was observed in the muscles (**Vergilio et al., 2012**). Muscles was found to accumulate small amounts of all the heavy metals and might have received them through circulation. It was suggested that, the low accumulation of metals in muscles may be due to lack of binding affinity of these metals with the proteins of muscles (**Osman, 2012**).

Liver plays multifunctional role in detoxification mechanism and storage process and may be due to their strong binding with cystine residues of metallothionein, where the lower molecular weight protein has high affinities for heavy metals and its storage as a constituent of hepatic cytoplasm, trigger increased accumulation of metal in the liver (**Ashraf et al., 2011; Montaser et al., 2010**).

Mercuric ion, gradually ionic mercury forms complexes with SH group and other ligands in the tissues of the body and only a very small fraction exists in the free form (**Ashraf et al., 2012**).

### Conclusion

Based on the obtained results it could be concluded that, growth rate and feed conversion improved by dietary essential oils supplementation.

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**Table 6. Mercury residual (ppm) of Nile tilapia as affected by dietary mercury contamination and their amelioration.**

Treatment	Muscles	Liver
T1	0.000±0.000 <sup>c</sup>	0.000±0.000 <sup>d</sup>
T2	0.127±0.012 <sup>a</sup>	0.193±0.009 <sup>a</sup>
T3	0.040±0.006 <sup>b</sup>	0.057±0.009 <sup>c</sup>
T4	0.060±0.006 <sup>b</sup>	0.080±0.000 <sup>b</sup>
Significance	***	***

\*\*\*  $P < 0.001$ . Means in the same column with different letters differ significantly ( $P < 0.05$ ).

## REFERENCES

- Abdel-Tawwab, M., A.M. Shalaby, M.H. Ahmad and Y.A. Khattab (2004). Effect of supplementary dietary L-ascorbic acid (vitamin C) on mercury detoxification, physiological aspects and growth performance of Nile tilapia (*Oreochromis niloticus* L.). Proc. 6<sup>th</sup> Int. Symposium on Tilapia in Aquac. Int. Conv. Cent. Roxas Boulevard Manila, Philippines, 159-171.
- Agarwal, R., S.K. Goel, R. Chandra and J.R. Behari (2010). Role of vitamin E in preventing acute mercury toxicity in rat. Environ. Toxicol. Pharmacol., 29: 70-80.
- AOAC (1990). Association of Official Analytical Chemists. The Official Methods of Analyses Association of Official Analytical Chemists International. 5<sup>th</sup> Ed., Arlington, VA, USA.
- Asefi, M. and R. Zamani-Ahmadm Mahmoodi (2015). Mercury concentrations and health risk assessment for two fish species, *Barbus grypus* and *Barbus luteus*, from the Maroon River, Khuzestan province, Iran. Environ. Monitoring and Assessment, 187: 653-663.
- Ashraf, M.A., M.J. Maah and I. Yusoff (2011). Assessment of heavy metals in the fish samples of mined out ponds Bestari Jaya, Peninsular Malaysia. Proc. Indian Nat. Sci. Acad., 77 (1): 57-67.
- Ashraf, M.A., M.J. Maah and I. Yusoff (2012). Bioaccumulation of heavy metals in fish species collected from former tin mining catchment. Int. J. Environ. Res., 6 (1): 209-218.
- Ayyat, M.S., K.M. Hemat, A.E. El-Hais and K.M. Abd El-Latif (2017). The role of some feed additives in fish fed on diets contaminated with cadmium. Environ. Sci. and Pollution Res., 24 (30): 23636-23645.
- Berger, A. and J.E. Halver (1987). Effect of dietary protein, lipid and carbohydrate content on the growth, feed efficiency and carcass composition of striped bass, (*Morone saxatilis* W.) fingerlings. Aquac., 18: 345-356.
- Beyersmann, D. and A. Hartwig (2008). Carcinogenic metal compounds: recent insight into molecular and cellular mechanisms. Arch Toxicol., 82 (8): 493-512.
- Billerbeck, V.G., C.G. Roques, J.M. Bessiere, J.L. Fonvieille and R. Dargent (2011). Effects of *Cymbopogon nardus* (L.) W. Watson essential oil on the growth and morphogenesis of *Aspergillus niger*. Canadian J. Microbiol., 47: 9-17.
- Boukhatem, M.N., A. Kameli, M.A. Ferhat, F. Saidi and M. Mekarnia (2013). Rose geranium essential oil as a source of new and safe anti-inflammatory drugs. The Libyan J. Med., 8: 225-232.
- Boukhris, M., M. Bouaziz, I. Feki, H. Jemai, A. El-Feki and S. Sayadi (2012). Hypoglycemic and antioxidant effects of leaf essential oil of *Pelargonium graveolens* L'Hér. in alloxan induced diabetic rats. Lipids in Health and Disease, 11: 81.
- Coimbra, J.L., A.C. Soares, M.S. Garrido, C.S. Souza and F.L. Ribeiro (2006). Toxicidade de extratos vegetais *Scutellonema bradys*. Pesquisa Agropecuária Brasileira., 41(7): 1209 -1211.
- Dobrowolski, R. and M. Skowrońska (2006). The study of trace metal levels in select environmental components of the Zemborzyce reservoir. Polish J. Environ. Studies, 4: 537-542.
- Duncan, D.B. (1955). Multiple Ranges and Multiple F-test. Biometrics, 11: 1-42.
- ECDG (2002). European Commission DG ENV. E3 Project ENV. E.3/ETU/0058. Heavy metals in waste. Final report. COWI A/S, Denmark.
- Gill, T.S., H. Tewari and J. Pande (1990). Use of the fish enzyme system in monitoring water quality: Effects of main tissue enzyme. Comp. Biochem. Physiol., 97: 287-292.
- Hamidpour, R., S. Hamidpour, M. Hamidpour, V. Marshall and R. Hamidpour (2017). *Pelargonium graveolens* (Rose Geranium) - A Novel Therapeutic Agent for Antibacterial, Antioxidant, Antifungal and Diabetics. Archives in Cancer Res., 5: 1-5.
- Hansen, J.A., J. Lipton, P.G. Welsh, J. Morris, D. Cacula and M.J. Suedkamp (2002a).



- Relationship between exposure duration, tissue residues, growth, and mortality in rainbow trout (*Oncorhynchus mykiss*) juveniles sub-chronically exposed to copper. *Aquatic Toxicol.*, 58:175-188.
- Hansen, J.A., P.G. Welsh, J. Lipton and M.J. Suedkamp (2002b). The effects of long-term cadmium exposure on the growth and survival of juvenile bull trout (*Salvelinus confluentus*). *Aquatic Toxicol.*, 58:165-174.
- Henry, R.J. (1974). *Clinical Chemistry, Principle and Technics*, 2<sup>nd</sup> Edition. Harper and Row, Page, 525.
- Houck, A. and J.J. Cech (2004). Effects of dietary methylmercury on juvenile Sacramento black fish bioenergetics. *Aquat Toxicol.*, 69: 107-23.
- Kumar, R.S., T. Sivakumar, R.S. Sunderam, M. Gupta, U.K. Mazumdar, P. Gomathi, Y. Rajeshwar, S. Saravanan, M.S. Kumar, K. Muruges and K.A. Kumar (2005). Antioxidant and antimicrobial activities of *Bauhinia racemosa* L. stem bark. *Brazil J. Med. Biol. Res.*, 38: 1015-1024.
- Lee, S.W. and W. Wendy (2013). Chemical composition and antimicrobial activity of *Cymbopogon nardus* citronella essential oil against systemic bacteria of aquatic animals. *Iranian J. Microbiol.*, 5 (2): 147-152.
- Lis-Balchin, M. and S. Deans (1996). Antimicrobial effects of hydrophilic extracts of *Pelargonium* species (Geraniaceae). *Letters in Appl. Microbiol.*, 23 (4): 205-207.
- Looi, L.J., Z.A. Ahmad, H. Hazzeman, M.Y. Fatimah and H. Zailina (2016). The levels of mercury, methylmercury and selenium and the selenium health benefit value in grey-eel catfish (*Plotosus canius*) and giant mudskipper (*Periophthalmodon schlosseri*) from the Strait of Malacca. *Chemosphere*, 152: 265-273.
- Manahan, S.E. (1979). *Environmental Chemistry*, 3<sup>rd</sup> Ed. Willand grant press, Boston, Massachusetts, 453-455.
- Mao, L.C., X. Pan, F. Que and X-H. Fang (2006). Antioxidant properties of water and ethanol extracts from hot air-dried and freeze-dried daylily flowers. *Eur. Food Res. Technol.*, 222: 236-241.
- Meltem, D., L.G. Mziya and A.O. Argun (2007). Investigation of heavy metals levels in economically important fish species captured from the tuzla lagoon. *Food Chem.*, 102 (1): 415-421.
- Miura, K., A. Naganuma, S. Himeno and N. Imura (1995). Mercury toxicity: biochemical aspects. In: Goyer RA, Cherian G, editors. *Toxicol. Metals*. Berlin: Springer-Verlag, 163-187.
- Montaser, M., M. Mahfouz, S. El-Shazly, G. Abdel-Rahman and S. Bakry (2010). Toxicity of heavy metals on fish at Jeddah Coast KSA: Metallothionein expression as a biomarker and histopathological study on liver and gills. *World J. Fish and Marine Sci.*, 2 (3): 174-185.
- Osman, A.G. (2012). Biomarkers in Nile tilapia *Oreochromis niloticus niloticus* (Linnaeus, 1758) to assess the impacts of river Nile pollution: Bioaccumulation, biochemical and tissues biomarkers. *J. Environ. Prot.*, 3: 966-977.
- Oussalah, M., S. Caillet, L. Saucier and M. Lacroix (2006). Antimicrobial effects of selected plant essential oils on the growth of a *Pseudomonas putida* strain isolated from meat. *Meat Sci.*, 73: 236-244.
- SAS (2002). SAS Institute Inc., Cary, NC, USA. In: NOTE: SAS Proprietary Software Version 9.00 (TS M0).
- Sivakami, R., G. Premkishore and M.R. Chandran (1995). Sublethal effects of mercury on feeding energetics and body composition of the freshwater catfish *Mystus vittatus* (Bloch). *J. Aquac. Trop.*, 10 (2): 109-117.
- The Herb Society of America (2006). *Pelargoniums: An Herb Society of America Guide* The Herb Society of America - 9019 Kirtland Chardon Rd., Kirtland, OH, 44094 - (440) 256-0514.
- Vergilio, C.S., C.E. Carvalho and E.J. Melo (2012). Accumulation and histopathological effects of mercury chloride after acute exposure in tropical fish *Gymnotus carapo*. *J. Chem. Health Risks*, 2 (4): 1-8.

- Wang, S. and X. Shi (2001). Molecular mechanisms of metal toxicity and carcinogenesis. *Mol Cell Biochem.*, 222: 3-9.
- Yildirim, A., A. Mavi, M. Oktay, A.A. Kara, O. Algur and V. Bilaloglu (2000). Comparison of antioxidant and antimicrobial activities of *Tilia (Tiliaargentea Desf ex DC)*, sage (*Salvia triloba L.*), and black tea (*Camellia sinensis*) extracts. *J Agric Food Chem.*, 48: 5030-5034.
- Zheng, Z., J.Y. Tan, H. Liu, X. Zhou, X. Xiang and K. Wang (2009). Evaluation of oregano essential oil (*Origanum heracleoticum L.*) on growth, antioxidant effect and resistance against *Aeromonas hydrophila* in channel catfish (*Ictalurus punctatus*). *Aquac.*, 292 (3): 214-218.

## أداء النمو وكفاءة الاستفادة من الغذاء ومقاييس الدم في البلطي النيلي المعرض لسمية الزئبق وتقليل سميته باستخدام إضافات غذائية من الزيوت الأساسية

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الهدف من هذه الدراسة هو معرفة التأثيرات السامة للزئبق ومحاولة إزالتها بواسطة استخدام المعاملة بزيت السترونيلا وزيت العنبر في العلائق المستخدمة خلال (١١٢) يوم تغذية، قسمت الأسماك إلى أربع مجموعات وكل مجموعة من الأسماك تم تخزينها في ثلاث أحواض كل واحد منها يحتوى على (١٠) أسماك، غذيت المجموعة الأولى على عليقة أساسية بدون زئبق والمجموعة الثانية غذيت على عليقة أساسية بالإضافة إلى ٥٠ جزء في المليون زئبق (كلوريد الزئبق) وكلا من المجموعة الثالثة والرابعة عوملت بالزئبق وأضيفت لها ٤٠٠ ملليجرام لكل كجم عليقة من زيت سترونيلا والعنبر على التوالي وتأثر وزن الجسم الحى معنويا عند عمر ١٦ أسبوع حيث زاد بنسبة ٤٠,٧٦%، ٢٢,٦٤% و ١٢,٤١% على التوالي في مجموعة الأسماك التي تغذت على عليقة بدون الزئبق (عليقة أساسية)، مجموعة الأسماك التي تغذت على عليقة ملوثة بالزئبق وأضيفت لها زيت السترونيلا و مجموعة الأسماك الملوثة بالزئبق ومضاف لها زيت العنبر بالمقارنة مع مجموعة الاسماك التي غذيت على عليقة ملوثة بالزئبق، وأيضا تحسنت الزيادة اليومية في الوزن عند (صفر-١٦) أسبوع بنسبة ٥١,٣٨، ٢٨,٤٤ و ١٥,٦٠% على التوالي وتحسن معامل التحويل الغذائى عند (صفر-١٦) أسبوع بنسبة ٢٩,٥٦، ١٦,٩٢ و ١٤,٢٧% في مجموعة الأسماك التي تغذت على عليقة بدون الزئبق (عليقة أساسية)، ومجموعة الأسماك التي غذيت على عليقة ملوثة بالزئبق ومضاف إليها زيت السترونيلا ومجموعة الأسماك التي غذيت على عليقة ملوثة بالزئبق ومضاف إليها زيت العنبر، على التوالي، عند مقارنتها بمجموعة الأسماك التي غذيت على عليقة ملوثة بالزئبق، وازداد البروتين الكلى والالبيومين فى سيرم الدم فى مجموعة الأسماك التي غذيت على عليقة بدون زئبق وانخفض urea-N، ALT، AST والكرياتين وايضا زاد البروتين الكلى فى سيرم الدم فى مجموعة الأسماك التي تغذت على عليقة ملوثة بالزئبق ومضاف لها زيت السترونيلا أو العنبر بالمقارنة مع مجموعة الأسماك التي غذيت على عليقة ملوثة بالزئبق، انخفضت متبقيات الزئبق معنويا بنسبة ٢٩,٥٣ و ٣١,٤٩ في كبد وعضلات مجموعة الأسماك التي غذيت على عليقة ملوثة بالزئبق ومضاف إليها زيت السترونيلا، على التوالي، وانخفض أيضا بمجموعة الأسماك التي غذيت على عليقة ملوثة بالزئبق ومضاف لها زيت العنبر بنسبة ٤١,٤٥ و ٤٧,٢٤، على التوالي، وبناءً على النتائج المتحصل عليها نستنتج أن إضافة الزيوت الأساسية فى علائق أسماك البلطى النيلي حسنت من معدل النمو، ومعامل التحويل الغذائى.

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