GROWTH PERFORMANCE AND SERUM BIOCHEMISTRY OF AFRICAN CATFISH (*Clarias gariepinus*) JUVENILES SUBJECTED TO SUB-LETHAL LEAD ACETATE EXPOSURE

Uzochukwu, I.E. [+,*], Iheukwumere U. [#], Amaefule, B.C. [+], Ogbonna, J.C. [+], Eze, S.C. [+], Ogwuegbu, M.C. [+], and Ezeasor, C.K.[$]

[+]Department of Animal Science, University of Nigeria, Nsukka, Nigeria

[*]Department of Hydrobiology, Faculty of Science and Technology, University of Debrecen, Debrecen, Hungary

[#]Animal Care and Veterinary Service, University of Ottawa, Ottawa Canada

[$]Department of Veterinary Pathology and Microbiology, University of Nigeria, Nsukka, Nigeria

Corresponding Author: Uzochukwu, I.E., ifeanyi.uzochukwu@unn.edu.ng; +2348060484743

ABSTRACT

A 56-day study was designed to assess the effect of sub-lethal lead acetate exposure on the growth performance, and serum biochemistry and anti-oxidative status of African catfish. Forty-eight (48) juveniles catfish of similar age and weight were procured and used for the study in a completely randomized design. The fish were randomly divided into 2 treatments of Control and Lead (Pb) acetate exposed (11.02mg/l) groups. At the end of the study, weight and length measurements were taken and blood samples were collected from the fish for studies of serum biochemistry and anti-oxidative status. The results showed a significant (p< 0.05) decrease in the final body weight (FBW), total weight gain (TWG), total feed intake (TFI), specific growth rate (SGR) and standard-length gain (SLG) in the Pb-group compared to the control. On the other hand, serum albumin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), and superoxide dismutase (SOD) levels significantly differed (p< 0.05) between the groups, while serum glucose, globulin, total protein, catalase, and malondialdehyde were not affected. The Pb-exposed group showed increased serum AST and SOD levels and decreased albumin and ALT levels compared to the control. It was concluded that exposure of catfish juveniles to sub-lethal lead acetate in water negatively affected growth performance, stimulated increased free radical generation, and caused liver damage.

Keywords: Heavy metal, pollution, African catfish, growth performance, oxidative stress

INTRODUCTION

The fisheries industry represents an important food sector with an annual fish production of about 1 million tons which amounts to more than 60% of the world supply of animal protein, especially in developing countries like Nigeria (FAO, 2007). In addition to improving food insecurity, the fishery industry is also recognized for its role in poverty alleviation, as well as the improvement of the socioeconomic status and livelihoods particularly, in rural communities (Graaf and Garibaldi, 2014). African catfish represents a choice fish species in Nigeria due to several reasons including its fast growth rate, high fecundity, and the market preference for its meat’s taste. Although aquaculture represents the major source of catfish in the country, there is still heavy reliance on the wild (rivers, lakes and dams) for seedlings and adults of various catfish species including *Clarias gariepinus*, *Heterobranchus bidorsalis*, Clarias X Heterobranchus hybrid (Heteroclarias) and *Clarias nigro-digitatus* (Banrie, 2012).

The issue of freshwater contamination with heavy metals such as lead (Pb) continues to pose serious concerns globally, due to their public health concerns. Like many other heavy metals, Pb is highly persistent, toxic, and tends to bio-accumulate and bio-magnify in organisms, and along the food chain (Jean-Marc, 2011). Following their rising application in industrial productions and activities, their ubiquitous occurrences, and reports of increasing freshwater contaminations in Nigeria, the need to investigate their impacts on the growth and health of aquatic lives can never be overemphasized. The current study sought to investigate the effects of sub-lethal Pb contamination on the growth performance and serum biochemistry of African catfish juveniles in an aquaculture setting. The results of this study can serve as a guide on the will go a long way to provide an idea of the impact of the increasing water contamination and pollution on aquatic lives, and possibly help to guide governments and other agencies in the development of policies for the protection of aquatic health, as well as the health of humans who depends on these water systems for their source of quality proteins.

MATERIAL AND METHOD

Location and Duration of the Study

The experiment was conducted at the Fishery Unit of the Department of Animal Science Teaching and Research Farm, the University of Nigeria, Nsukka in Enugu state, Nigeria. The experiment lasted for 60 days (2 months)

Procurement of Test Materials

The lead acetate ($Pb(C_2H_3O_2)_2$) used for this study was analytical grade and was procured from a reputable laboratory-chemicals store (Joechem chemicals) at Nsukka, Enugu State.

Experimental Design and Fish Management

A total of forty-eight (48) African catfish (*Clarias gariepinus*) juveniles of similar age and sizes were procured from a reputable fish farm in Nsukka, Enugu State, and used for the study. After procurement, the fish were stocked in a concrete pond (3.6 m × 2.7 m × 2.7 m) containing dechlorinated water and acclimatized for two weeks. Thereafter, the fish were weighed and randomly sorted into two treatments groups, A and B in a Completely Randomized Design. Treatment A represents the control group with no administration of lead acetate, while fish in Treatment B was exposed to sub-lethal concentrations of lead acetate. Each group was replicated thrice with 8 fish per replicate, with each replicate stcked in and reared in individual plastic containers (20 L) for eight weeks. The present study considered the LC_{50} values of Lead (Pb) on *Clarias gariepinus* to be 55.12 mg/L, as studied and reported by (Awoyemi *et al*., 2014). For this, a 20% sub-lethal concentration of the 96-h LC_{50} values of Pb (11.02 mg/L) was applied and exposed to the Treatment -B fish as a toxicant. The fish were fed with a basal diet containing about 30% crude protein, 6% crude fibre, and 3.0 mcal/kg of metabolizable energy at 3% (1.5% fed twice) of their biomass daily for a total period of 8 weeks.

Data Collection and Statistical Analysis

After the acclimatization period, before the commencement of the study, and at the end of the study, the wet-weight and standard-length measurements of the fish were taken and recorded and used to deduce the various growth performance indices. At the end of the eight weeks duration also, fish samples were randomly selected from each treatment group, and blood samples were obtained from the caudal vein of the fish using a 1-mL heparinized syringe for serum biochemistry and oxidative stress status as described (Qadir et al., 2014). Data generated were subjected to independent sample t-test analysis using the R-statistical software.

RESULTS AND DISCUSSION

Growth Performance

The results of the effect of lead acetate exposure on the growth performance of African catfish juveniles are presented in Table 1. The results showed that there were no significant differences ($p \geq 0.05$) between the lead-treated fish group and the control in the initial body weight, t (4) = 0.16, p = .08, feed conversion ratio, t (4) = 1.95, p = .12, Initial standard length, t (4) = 1.33, p = .26, final standard length, t (4) = 2.23, p = .08, and the condition factor, t (4) = 0.06, p = .95. However, the treatments groups differed ($p<0.05$) in the growth indices of final body weight (FBW), t (4) = 3.32, p = .03, total weight gained (TWG), t (4) = 3.44, p = .03, total feed intake (TFI), t (4) = 3.42, p = .03, specific growth rate (SGR), t (4) = 3.46, p = .03, and standard-length gained (SLG), t (4) = 4.99, p < .001. The results indicateed that exposure of catfish juveniles to lead acetate treatments negatively affected the growth performance indices of FBW, TWG, SLG, TFI, and SGR.

The lowered FBW, TWG, SLG, TFI, and SGR of fish in the Treatment B may be attributed to the lead acetate contaminant. When exposed to Pb or other heavy metals polluted water, aquatic organisms have been reported to lower their feed intake and expend available energy on detoxification processes, rather than metabolic and growth processes (Florian *et al*, 2016). Therefore, Pb pollution in an aquatic system can adversely affect the feed intake and growth performance of fish. This may explain the lower TFI, TWG, SLG and SGR observed for the Pb-group compared to the control. This is in alignment with the studies of Ayegbusi *et al*. (2018) that recorded adverse effects of lead chloride on the growth performance of *Clarias gariepinus*. It is therefore clear from the above that exposure of fish to heavy metals such as Pb causes a reduction in fish growth.

Table 1: Effect of lead acetate exposure on the growth performance of African catfish juveniles

Parameters	**(T1) (Control)**	**(T2) (Lead acetate)**	**P-Values**
1BW (g)	9.83±0.15	9.87±0.32	0.08^{NS}
FBW (g)	48.90±3.10	36.10±5.89	0.03^{*}
TWG (g)	39.07±3.19	26.23±5.98	0.03^{*}

TF1 (g)	53.10±3.81	39.11±5.98	0.03*
FCR	1.36±0.07	1.51±0.11	0.12NS
SGR	2.86±0.13	2.30±0.25	0.03*
ISL (cm)	11.27±0.64	11.90±0.52	0.26NS
FSL (cm)	20.30±0.92	18.4 ±1.15	0.08NS
SLG (cm)	9.03±0.50	6.50 ±0.72	0.00**
K	0.59±0.07	0.60±0.18	0.95NS

*: Significant ($p < 0.05$); **: highly Significant; NS: Non-significance; BW: body weight; WG: weight gain; TFI: Total feed intake; FCR: Feed conversion ratio; SGR: Specific growth rate; ISL: Initial standard length; FSL: Final standard length; SLG: Standard length gain; K: Condition factor.

Serum Biochemistry and Oxidative Stress Status

The results of the effect of lead acetate exposure on the serum biochemistry of African catfish juveniles are shown in Table 2.

Table 2. Effect of lead acetate exposure on the serum biochemistry of African catfish juveniles

Parameters	**T1 (Control)**	**T2 (Lead Acetate)**	**P-value**
Glucose (mmol/l)	88.75±4.12	95.65± 8.16	0.13NS
Globulin (g/dl)	19.25± 8.19	17.55 ± 7.09	0.73NS
Albumin (g/dl)	32.86± 0.93a	29.55 ± 1.38b	0.002*
Total protein (g/dl)	52.11± 8.17	47.10 ± 7.86	0.35NS
ALT (u/l)	116.07± 8.72 a	97.11± 6.04 b	0.004*
AST (u/dl)	144.73± 8.52 b	158.56 ± 7.16 a	0.02*
CAT (Mm/l)	153.29 ± 4.25	180.48 ± 22.95	0.06NS
SOD (u/dl)	9.27± 2.38 b	26.17 ± 6.50 a	< 0.001*
MDA (Mm/l)	3.59± 0.40	3.36 ± 0.33	0.37NS

Significant: ($p < 0.05$); NS: Non-significance; SOD: Superoxide dismutase; MDA: Malondialdehyde.

Results (Table 2) showed that there were no significant differences between fish in the lead treated and the control in the serum indices of serum glucose, t (8) = -1.69, p = 0.13, globulin, t (8) = 0.35, p = 0.73, total protein, t (8) = 0.99, p = 0.35, catalase, t (4) = - 2.61, p= 0.056, and Malondialdehyde, t (8) = 0.96, p = 0.37. However, the groups differed in the serum albumin, t (8) = 4.44, p = 0.002, Alanine aminotransferase (ALT), t (8) = 3.10, p = 0.001, Aspartate aminotransferase (AST), t (8) = -2.78, p = 0.02, Superoxide dismutase (SOD), t (8) = -5.46, p = < 0.001. The results showed that lead acetate administration resulted in a lowered serum ALT and albumin, but increased levels of AST, and SOD in the fish.

The lowered serum albumin and ALT values may be attributed to the Pb pollutant administered to the fish. Exposure of fish to heavy metals contaminated water can lead to liver damage which is evidenced by a marked reduction in serum albumin levels (Samuel et *al*., 2021). Similar marked decreases in serum albumin have been noted in *Channa punctatus* fish collected from polluted canals (Javed and Usmani, 2015). On the other hand, serum aminotransferases, ALT, AST, and ALP are the most used indicators of liver damage in animals (Oztopuz et al., 2019). While higher activities of these enzymes are suggestive of a prolonged heavy metals exposure period, thus indicating liver damage, lowered activities may connote lesser damage (Tabrez et al., 2021). The marked decrease in serum ALT and increase in AST as reported in the present study may suggest varying degrees of liver damage in the fish due to the administered Pb pollutant. Similar reports of elevated serum AST, ALT, and ALP activities have been previously reported in various fish species (Zorriehzahra *et al.*, 2010, Parvathi *et al.*, 2011). Furthermore, according to Farombi et al. (2007), bioaccumulation of heavy metals (as Pb) may lead to increased tissue generation of superoxide anions which causes elevated SOD production that scavenges and converts superoxide radicals to H_2O_2. This may explain the elevated levels of SOD reported for the Pb-exposed group in this study.

CONCLUSION

This study sort to investigate the biological impacts of the increasing contamination and pollution of the natural aquatic systems with heavy metals. It is clear from this work that exposure of African catfish juveniles to sub-lethal Pb dosages doses in water can have adverse biological and physiological effects including stimulation of free radical production and liver damage. Further studies may be required to assess the bioaccumulation rates of this metal, its correlation with the various biological impacts, and the possible implication for human consumers.

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[+]Department of Animal Science, University of Nigeria, Nsukka, Nigeria

[*]Department of Hydrobiology, Faculty of Science and Technology, University of Debrecen, Debrecen, Hungary

[#]Animal Care and Veterinary Service, University of Ottawa, Ottawa Canada

[$]Department of Veterinary Pathology and Microbiology, University of Nigeria, Nsukka, Nigeria

Corresponding Author: Uzochukwu, I.E., ifeanyi.uzochukwu@unn.edu.ng; +2348060484743

Abstract

A 56-day study was designed to assess the effect of sub-lethal lead acetate exposure on the growth performance, and serum biochemistry and anti-oxidative status of African catfish. Forty-eight (48) juveniles catfish of similar age and weight were procured and used for the study in a completely randomized design. The fish were randomly divided into 2 treatments of Control and Lead (Pb) acetate exposed (11.02mg/l) groups. At the end of the study, weight and length measurements were taken and blood samples were collected from the fish for studies of serum biochemistry and anti-oxidative status. The results showed a significant ($p< 0.05$) decrease in the final body weight (FBW), total weight gain (TWG), total feed intake (TFI), specific growth rate (SGR) and standard-length gain (SLG) in the Pb-group compared to the control. On the other hand, serum albumin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), and superoxide dismutase (SOD) levels significantly differed ($p< 0.05$) between the groups, while serum glucose, globulin, total protein, catalase, and malondialdehyde were not affected. The Pb-exposed group showed increased serum AST and SOD levels and decreased albumin and ALT levels compared to the control. It was concluded that exposure of catfish juveniles to sub-lethal lead acetate in water negatively affected growth performance, stimulated increased free radical generation, and caused liver damage.

Keywords: Heavy metal, pollution, African catfish, growth performance, oxidative stress

Introduction

The fisheries industry represents an important food sector with an annual fish production of about 1 million tons which amounts to more than 60% of the world supply of animal protein, especially in developing countries like Nigeria (FAO, 2007). In addition to improving food insecurity, the fishery industry is also recognized for its role in poverty alleviation, as well as the improvement of the socioeconomic status and livelihoods particularly, in rural communities (Graaf and Garibaldi, 2014). African catfish represents a choice fish species in Nigeria due to several reasons including its fast growth rate, high fecundity, and the market preference for its meat's taste. Although aquaculture represents the major source of catfish in the country, there is still heavy reliance on the wild (rivers, lakes and dams) for seedlings and adults of various catfish species including *Clarias gariepinus*, *Heterobranchus bidorsalis*, Clarias X Heterobranchus hybrid (Heteroclarias) and *Clarias nigro-digitatus* (Banrie, 2012).

The issue of freshwater contamination with heavy metals such as lead (Pb) continues to pose serious concerns globally, due to their public health concerns. Like many other heavy metals, Pb is highly persistent, toxic, and tends to bio-accumulate and bio-magnify in organisms, and along the food chain (Jean-Marc, 2011). Following their rising application in industrial productions and activities, their ubiquitous occurrences, and reports of increasing freshwater contaminations in Nigeria, the need to investigate their impacts on the growth and health of aquatic lives can never be overemphasized. The current study sought to investigate the effects of sub-lethal Pb contamination on the growth performance and serum biochemistry of African catfish juveniles in an aquaculture setting. The results of this study can serve as a guide on the will go a long way to provide an idea of the impact of the increasing water contamination and pollution on aquatic lives, and possibly help to guide governments and other agencies in the development of policies for the protection of aquatic health, as well as the health of humans who depends on these water systems for their source of quality proteins.

Material and Method

Location and Duration of the Study

The experiment was conducted at the Fishery Unit of the Department of Animal Science Teaching and Research Farm, the University of Nigeria, Nsukka in Enugu state, Nigeria. The experiment lasted for 60 days (2 months)

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Experimental Design and Fish Management

A total of forty-eight (48) African catfish (*Clarias gariepinus*) juveniles of similar age and sizes were procured from a reputable fish farm in Nsukka, Enugu State, and used for the study. After procurement, the fish were stocked in a concrete pond (3.6 m × 2.7 m × 2.7 m) containing dechlorinated water and acclimatized for two weeks. Thereafter, the fish were weighed and randomly sorted into two treatments groups, A and B in a Completely Randomized Design. Treatment A represents the control group with no administration of lead acetate, while fish in Treatment B was exposed to sub-lethal concentrations of lead acetate. Each group was replicated thrice with 8 fish per replicate, with each replicate stcked in and reared in individual plastic containers (20 L) for eight weeks. The present study considered the LC_{50} values of Lead (Pb) on *Clarias gariepinus* to be 55.12 mg/L, as studied and reported by (Awoyemi *et al*., 2014). For this, a 20% sub-lethal concentration of the 96-h LC_{50} values of Pb (11.02 mg/L) was applied and exposed to the Treatment -B fish as a toxicant. The fish were fed with a basal diet containing about 30% crude protein, 6% crude fibre, and 3.0 mcal/kg of metabolizable energy at 3% (1.5% fed twice) of their biomass daily for a total period of 8 weeks.

Data Collection and Statistical Analysis

After the acclimatization period, before the commencement of the study, and at the end of the study, the wet-weight and standard-length measurements of the fish were taken and recorded and used to deduce the various growth performance indices. At the end of the eight weeks duration also, fish samples were randomly selected from each treatment group, and blood samples were obtained from the caudal vein of the fish using a 1-mL heparinized syringe for serum biochemistry and oxidative stress status as described (Qadir et al., 2014). Data generated were subjected to independent sample t-test analysis using the R-statistical software.

Results and Discussion

Growth Performance

The results of the effect of lead acetate exposure on the growth performance of African catfish juveniles are presented in Table 1. The results showed that there were no significant differences ($p \geq 0.05$) between the lead-treated fish group and the control in the initial body weight, t (4) = 0.16, p = .08, feed conversion ratio, t (4) = 1.95, p = .12, Initial standard length, t (4) = 1.33, p = .26, final standard length, t (4) = 2.23, p = .08, and the condition factor, t (4) = 0.06, p = .95. However, the treatments groups differed ($p<0.05$) in the growth indices of final body weight (FBW), t (4) = 3.32, p = .03, total weight gained (TWG), t (4) = 3.44, p = .03, total feed intake (TFI), t (4) = 3.42, p = .03, specific growth rate (SGR), t (4) = 3.46, p = .03, and standard-length gained (SLG), t (4) = 4.99, p < .001. The results indicateed that exposure of catfish juveniles to lead acetate treatments negatively affected the growth performance indices of FBW, TWG, SLG, TFI, and SGR.

The lowered FBW, TWG, SLG, TFI, and SGR of fish in the Treatment B may be attributed to the lead acetate contaminant. When exposed to Pb or other heavy metals polluted water, aquatic organisms have been reported to lower their feed intake and expend available energy on detoxification processes, rather than metabolic and growth processes (Florian *et al*, 2016). Therefore, Pb pollution in an aquatic system can adversely affect the feed intake and growth performance of fish. This may explain the lower TFI, TWG, SLG and SGR observed for the Pb-group compared to the control. This is in alignment with the studies of Ayegbusi *et al*. (2018) that recorded adverse effects of lead chloride on the growth performance of *Clarias gariepinus*. It is therefore clear from the above that exposure of fish to heavy metals such as Pb causes a reduction in fish growth.

Table 1: Effect of lead acetate exposure on the growth performance of African catfish juveniles

Parameters	(T1) (Control)	(T2) (Lead acetate)	P-Values
1BW (g)	9.83±0.15	9.87±0.32	0.08^{NS}
FBW (g)	48.90±3.10	36.10±5.89	0.03^{*}
TWG (g)	39.07±3.19	26.23±5.98	0.03^{*}
TF1 (g)	53.10±3.81	39.11±5.98	0.03^{*}
FCR	1.36±0.07	1.51±0.11	0.12^{NS}
SGR	2.86±0.13	2.30±0.25	0.03^{*}

ISL (cm)	11.27±0.64	11.90±0.52	0.26^{NS}
FSL (cm)	20.30±0.92	18.4 ±1.15	0.08^{NS}
SLG (cm)	9.03±0.50	6.50 ±0.72	0.00^{**}
K	0.59±0.07	0.60±0.18	0.95^{NS}

*: Significant (p < 0.05); **: highly Significant; NS: Non-significance; BW: body weight; WG: weight gain; TFI: Total feed intake; FCR: Feed conversion ratio; SGR: Specific growth rate; ISL: Initial standard length; FSL: Final standard length; SLG: Standard length gain; K: Condition factor.

Serum Biochemistry and Oxidative Stress Status

The results of the effect of lead acetate exposure on the serum biochemistry of African catfish juveniles are shown in Table 2.

Table 2. Effect of lead acetate exposure on the serum biochemistry of African catfish juveniles

Parameters	T1 (Control)	T2 (Lead Acetate)	P-value
Glucose (mmol/l)	88.75±4.12	95.65± 8.16	0.13^{NS}
Globulin (g/dl)	19.25± 8.19	17.55 ± 7.09	0.73^{NS}
Albumin (g/dl)	32.86± 0.93a	29.55 ± 1.38b	0.002^{*}
Total protein (g/dl)	52.11± 8.17	47.10 ± 7.86	0.35^{NS}
ALT (u/l)	116.07± 8.72 a	97.11± 6.04 b	0.004^{*}
AST (u/dl)	144.73± 8.52 b	158.56 ± 7.16 a	0.02^{*}
CAT (Mm/l)	153.29 ± 4.25	180.48 ± 22.95	0.06^{NS}
SOD (u/dl)	9.27± 2.38 b	26.17 ± 6.50 a	$< 0.001^{*}$
MDA (Mm/l)	3.59± 0.40	3.36 ± 0.33	0.37^{NS}

Significant: (p < 0.05); NS: Non-significance; SOD: Superoxide dismutase; MDA: Malondialdehyde.

Results (Table 2) showed that there were no significant differences between fish in the lead treated and the control in the serum indices of serum glucose, t (8) = -1.69, p = 0.13, globulin, t (8) = 0.35, p = 0.73, total protein, t (8) = 0.99, p = 0.35, catalase, t (4) = - 2.61, p= 0.056, and Malondialdehyde, t (8) = 0.96, p = 0.37. However, the groups differed in the serum albumin, t (8) = 4.44, p = 0.002, Alanine aminotransferase (ALT), t (8) = 3.10, p = 0.001, Aspartate aminotransferase (AST), t (8) = -2.78, p = 0.02, Superoxide dismutase (SOD), t (8) = -5.46, p = < 0.001. The results showed that lead acetate administration resulted in a lowered serum ALT and albumin, but increased levels of AST, and SOD in the fish.

The lowered serum albumin and ALT values may be attributed to the Pb pollutant administered to the fish. Exposure of fish to heavy metals contaminated water can lead to liver damage which is evidenced by a marked reduction in serum albumin levels (Samuel et *al*., 2021). Similar marked decreases in serum albumin have been noted in *Channa punctatus* fish collected from polluted canals (Javed and Usmani, 2015). On the other hand, serum aminotransferases, ALT, AST, and ALP are the most used indicators of liver damage in animals (Oztopuz et al., 2019). While higher activities of these enzymes are suggestive of a prolonged heavy metals exposure period, thus indicating liver damage, lowered activities may connote lesser damage (Tabrez et al., 2021). The marked decrease in serum ALT and increase in AST as reported in the present study may suggest varying degrees of liver damage in the fish due to the administered Pb pollutant. Similar reports of elevated serum AST, ALT, and ALP activities have been previously reported in various fish species (Zorriehzahra *et al*., 2010, Parvathi *et al*., 2011). Furthermore, according to Farombi et al. (2007), bioaccumulation of heavy metals (as Pb) may lead to increased tissue generation of superoxide anions which causes elevated SOD production that scavenges and converts superoxide radicals to H_2O_2. This may explain the elevated levels of SOD reported for the Pb-exposed group in this study.

Conclusion

This study sort to investigate the biological impacts of the increasing contamination and pollution of the natural aquatic systems with heavy metals. It is clear from this work that exposure of African catfish juveniles to sub-lethal Pb dosages doses in water can have adverse biological and physiological effects including stimulation of free radical production and liver damage. Further studies may be required to assess the bioaccumulation rates of this metal, its correlation with the various biological impacts, and the possible implication for human consumers.

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