International Journal of Ecotoxicology and Ecobiology
2022; 7(4): 65-70
http://www.sciencepublishinggroup.com/j/ijee
doi: 10.11648/j.ijee.20220704.12
ISSN: 2575-1727 (Print); ISSN: 2575-1735 (Online)

Moringa oleifera Leaf Extract MOLE Ameliorate the Behaviour and Median Lethal Toxicity of Hetereoclarias Hybrid Fingerlings Disclosed to Cassava Wastewater CWW

Ezike Christopher Onyemaechi[1, *], Agbo Aderonke Nana[2], Uwadiegwu Nicholas Chinwe[1], Okechukwu Godwin Chukwuka Ejike[3]

[1]Department of Animal/Fisheries Science and Management, Faculty of Agriculture, Enugu State University of Science and Technology (ESUT), Enugu, Nigeria

[2]Department of Animal Science and Fisheries, Faculty of Agriculture, National Open University of Nigeria (NOUN), Kaduna, Nigeria

[3]Department of Agronomy and Ecological Management, Faculty of Agriculture, Enugu State University of Science and Technology (ESUT), Enugu, Nigeria

Email address:

Christopher.ezike@esut.edu.ng (Ezike Christopher Onyemaechi)

[*]Corresponding author

To cite this article:

Ezike Christopher Onyemaechi, Agbo Aderonke Nana, Uwadiegwu Nicholas Chinwe, Okechukwu Godwin Chukwuka Ejike. *Moringa oleifera* Leaf Extract MOLE Ameliorate the Behaviour and Median Lethal Toxicity of Hetereoclarias hybrid Fingerlings Disclosed to Cassava Wastewater CWW. *International Journal of Ecotoxicology and Ecobiology.* Vol. 7, No. 4, 2022, pp. 65-70. doi: 10.11648/j.ijee.20220704.12

Received: October 10, 2022; **Accepted**: November 3, 2022; **Published**: December 29, 2022

Abstract: The ameliorative potency of *Moringa oleifera* leaf extract MOLE 2mg/L on cassava wastewater CWW 0.01, 0.02, 0.03, 0.04, 0.05 and 0.00 mg CWW /L of clean water, disclosed to the hybrid fingerlings 2.00 ± 0.00g of the African catfish was investigated. The experiment was carried out in a semi static Fisheries Unit Laboratory bioassay, Enugu Lat. 7.4N; 8° 7'5 and long 6° 8'E. 7° 6' W for 96 hours. The acute toxicity was followed by the addition of a safety dose of MOLE to each of the acute concentration, and disclosed to the test fish of similar species and weight, for exact time of the acute test. Disclosed fish showed rapid swimming movements, mouth gaping and constraint, and loss of equilibrium and inactivity prior to demise. Dose dependent oxygenation rate and tail pulsation regularity was indicated at the onset of the experiment, but decreased towards the finish of the disclosure interval. The 96 hours LC50 was resolved to be 0.028 mg/L but was made more tolerable at 0.044 mg/L when MOLE was added. The test contaminant not surpassing the welfare utility level of 0.00028 mg/L should not be let into the aquatic environment for the farming of hybrid fingerlings of the test fish, except where 2.00 mg/L MOLE is applied to decrease the lethality of CWW to 0.00044 mg CWW/L of water.

Keywords: *Moringa oleifera* Leaf Extract, Ameliorate, Behavior Toxicity, Median Lethal Toxicity, Hetereoclarias Hybrid, Disclosure, Cassava Wastewater

1. Introduction

Cassava *Manihot exculenta* is a prime root and leaf food in Nigeria and other emerging nations [1], and its starch, support gut health and blood sugar [2]. Its flour is presently used as binder of animal meal and other industrial products [3], fuel- ethanol production, paper and textile production and as an inert carrier in pharmaceutical industries [4, 5]. The heightened usefulness has thus, introduced it to the facets of the environments and especially into the aquatics. Although reports of the use of cassava waste water has been made [6], but its damages to fish fingerlings is very huge, due to its lethality profile [7]. The toxicity of cassava waste water on different fish species abound [8, 9], but its lethality on hybrid African catfishes is rare [10]. The use of the seed extract of Moringa to better the purity of water and waste water is available in literature documentations [11-13], but has not been used to improve cassava contaminated water, exposed on hybrid catfish [14]. The aim of the present study is therefore to determine the ameliorative potency of MOLE to improve the lethality of CWW and the behavior of the disclosed hybrid fingerlings to it. The objective of the present study is therefore to investigate the 96h LC_{50}, the welfare utility level and the use of MOLE to ameliorate the median lethality of CWW on hybrid fingerlings exposed for 96 hours.

2. Materials and Methods

2.1. Experimental Fish, MOLE and CWW

Three hundred and sixty (360) hybrid fingerlings of African catfish (mean weight 2.00 ± 0.0 g, mean length 2.40 ±0.12cm) were obtained from a Private Farm in Enugu Nigeria and conveyed to the Fisheries Wet Laboratory of the Department of Animal/Fisheries Science and Management, Enugu State University of Science and Technology ESUT Lat. 7.4N; 8° 7'5 and long 6° 8'E. 7° 6' W, Enugu Nigeria. They were held in four fiber reinforced plastic (FRP) tanks, containing 300 L of de-chlorinated tap water. Ventilation was provided to all tanks round the clock in order to keep dissolved oxygen contents. Before the start of the study, the fish were adjusted for 14 days, and were fed with trade fish diet composed of 40% crude protein. The faecal matter and other waste materials were drained off daily to reduce ammonium content in the water. CWW was collected from a local cassava factory, while the leaf extract of moringa was prepared according to standard procedures [7], and was dissolved in distilled water to make a stock solution, used during the study. Ethical clearance from the Enugu State University of Science and Technology Committee on Experimental Animal Care was obtained and followed.

2.2. Acute Toxicity Test

The lethality of CWW on C. gariepinus was carried out following the OECD recommendation for testing of substances that has defined composition No. 203 in a semi-static restoration system by using 200L capacity glass aquaria. Five different concentrations (0.01, 0.02, 0.03, 0.04, and 0.05) and control 0.00 mgL^{-1} were selected and prepared in triplicates for acute exposures after a random- variable test, and ten (10) fish were exposed to each replicate. A group was exposed to clean freshwater which served as control. Feed was not offered 12 h prior to and during the 96 h of test period. Dead fish were immediately removed to prevent worsening of water quality. The exposure solution was renewed each day and was also The experiment was conducted under the natural daylight of 12:12 light-dark cycle and the physico-chemical parameters of the test water were examined daily, using standard methods [15], and were recorded (dissolved oxygen 4.40 ±0.20 - 7.00 ± 0.40 mg L^{-1} temperature 27.70 ± 0.5°C, pH 4.5 ±0.3-7.7 ± 0.12 and free carbon dioxide 4.24 ± 0.6 mg L^{-1}). The test fish were checked on hours 24, 48, 72 and 96 in each replicate to determine the dose response effects of CWW on the fish. 2mg/L of MOLE [14] was added to each lethal dose and exposed for the same period to estimate its possible use to enhance CWW acute toxicity level on test fish. The behavioral responses in exposed and control fish were observed and recorded daily. The LC_{50} was determined by Probit analysis [16]. The welfare utility dose WUD was estimated by 'applying the safety application factor (AF) suggested by CCREM [17].'

2.3. Statistical Analysis

Data was indicated as mean ± standard error and was analyzed using the 20.0 statistical package SPSS computer program (SPSS Inc. Chicago, Illinois, USA). Differences in the test concentrations and control were subjected to one-way analysis of variance (ANOVA), followed by Duncan range tests to determine significant mean differences.

3. Results

The dose response behavioral toxicity variation of the test fish exposed to acute doses of CWW is shown in Table 1, while Tables 2 and 3 represent the mortality rates followed by their probit 96h LC_{50} values during acute and amelioration levels in Figures 1 and 2 respectively.

Table 1. *Behavioral toxicity of hybrid catfish fingerlings disclosed to CWW.*

Parameters	Concentration Mg/L	Period (hours)			
		24	48	72	96
Rapid swimming movements	0.01	++	+	-	-
	0.02	++	++	-	-
	0.03	+++	++	+	-
	0.04	++++	+++	++	+
	0.05	-	-	++	+++
Mouth gaping and constraint	0.01	-	-	+	+
	0.02	-	-	+	+
	0.03	-	-	+	+
	0.04	-	-	++	+++
	0.05	-	-	++	+++
Loss of balance and inactivity	0.01	-	-	+	+
	0.02	-	-	+	+
	0.03	-	-	+	+
	0.04	-	-	++	+++
	0.05	-	-	++	+++

Parameters	Concentration Mg/L	Period (hours)			
		24	48	72	96
Opercula oxygenation	0.01	+	-	-	-
	0.02	++	+	-	-
	0.03	++	++	+	-
	0.04	+++	++	++	+
	0.05	+++	++	++	+
Tail pulsation regularity	0.01	+	+	-	-
	0.02	+	+	-	-
	0.03	++	+	-	-
	0.04	+++	++	++	+
	0.05	+++	++	++	+
Rapid swimming movements, Mouth gaping and constraint, Loss of balance and inactivity, Opercula oxygenation, Tail pulsation regularity	0.00	-	-	-	-
	0.00	-	-	-	-
	0.00	-	-	-	-

Key: -none, + mild, ++ moderate, +++strong, ++++ very strong.

3.1. Behavioral Toxicity

Exposed fish to the acute concentrations of CWW for 96 h showed differing grade of behavioral responses prior to death, such as period declined rapid swimming movements, dose dependent mouth gaping and constraint, Loss of balance and inactivity, before death (table 1). The Opercula oxygenation and Tail pulsation regularity declined with period and indicated dose dependent response but no clear unusual behavioral disarray was observed in the control group of fish during the study (table 1).

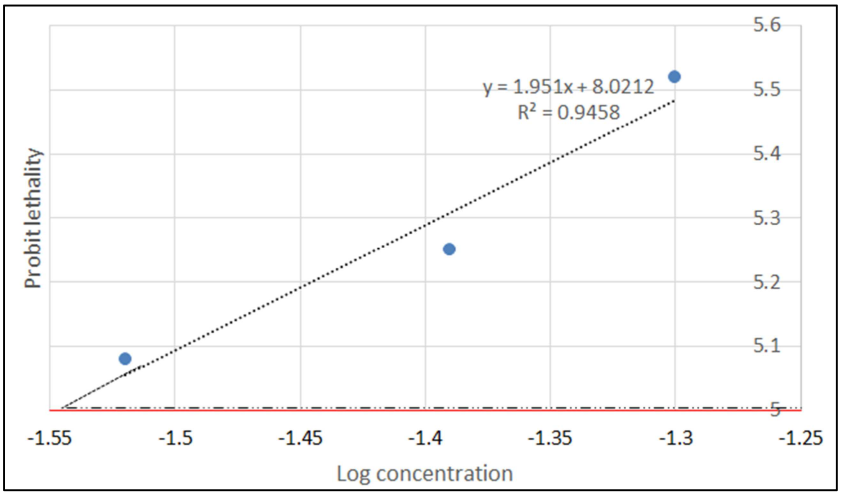

Figure 1. *Logarithmic probit line of 96h LC_{50} of CIWW disclosed to hybrid catfish fingerlings.*

Table 2. *Mortality rate of hybrid catfish fingerlings disclosed to CWW.*

conc. Mg/L	24h	48h	72h	96h
0.00	0	0	0	0
0.00	0	0	0	0
0.00	0	0	0	0
0.01	0	1	1	1
0.01	1	1	1	1
0.01	1	1	1	1
0.02	1	1	1	1
0.02	1	1	1	2
0.02	1	1	1	2
0.03	1	1	1	2
0.03	1	1	1	2
0.03	1	1	2	2
0.04	1	1	2	2
0.04	1	1	1	2
0.04	1	2	2	2
0.05	1	2	2	2
0.05	1	2	2	2
0.05	1	3	1	2

Concentration mg/L	Log concentration	Replicate 1	Replicate 2	Replicate 3	cumulative Mortality	% mortality	Probit
0.01	-2	3	4	4	11	36.66	4.64
0.02	-1.69	4	4	5	13	43.33	4,82
0.03	-1.52	5	5	6	16	53.33	5.08
0.04	-1.39	6	5	7	18	60.00	5.25
0.05	-1.3	7	7	7	21	70.00	5.52

3.2. Median Lethal Concentration and Safety Level Amelioration of Lethal with MOLE

The 96 hours LC_{50} was determined to be 0.028 mg/L (y= 1.951x + 8.0212, R^2=0.9458) Figure 1 but was ameliorated to be 0.044 mg/L (y= 7.2479x + 15.102, R^2 = 0.9061) when MOLE was added (Figure 2).

Table 3. *Mortality rate of hybrid fingerlings disclosed to CWW, and ameliorated with MOLE.*

0.00+2mg/L MLE	0	0	0	0
0.00+2mg/L MLE	0	0	0	0
0.00+2mg/L MLE	0	0	0	0
0.01+2mg/L MLE	0	0	0	0
0.01+2mg/L MLE	0	0	0	0
0.01+2mg/L MLE	0	0	0	0
0.02+2mg/L MLE	0	0	0	1
0.02+2mg/L MLE	0	0	0	1
0.02+2mg/L MLE	0	0	0	1
0.03+2mg/L MLE	0	0	1	2
0.03+2mg/L MLE	0	0	1	2
0.03+2mg/L MLE	0	0	2	2
0.04+2mg/L MLE	0	1	2	2
0.04+2mg/L MLE	0	1	1	2
0.04+2mg/L MLE	0	1	1	2
0.05+2mg/L MLE	0	2	2	2
0.05+2mg/L MLE	0	1	2	2
0.05+2mg/L MLE	0	3	1	2

Concentration. mg/L	Log concentration	Replicate 1	Replicate 2	Replicate 3	cumulative Mortality	% mortality	Probit
0.01	-2	0	0	0	0	0.00	0
0.02	-1.69	1	1	1	3	10.00	3.72
0.03	-1.52	3	3	4	10	33.33	4.56
0.04	-1.39	5	4	4	13	43.33	4.82
0.05	-1.3	6	5	6	17	56.66	5.15

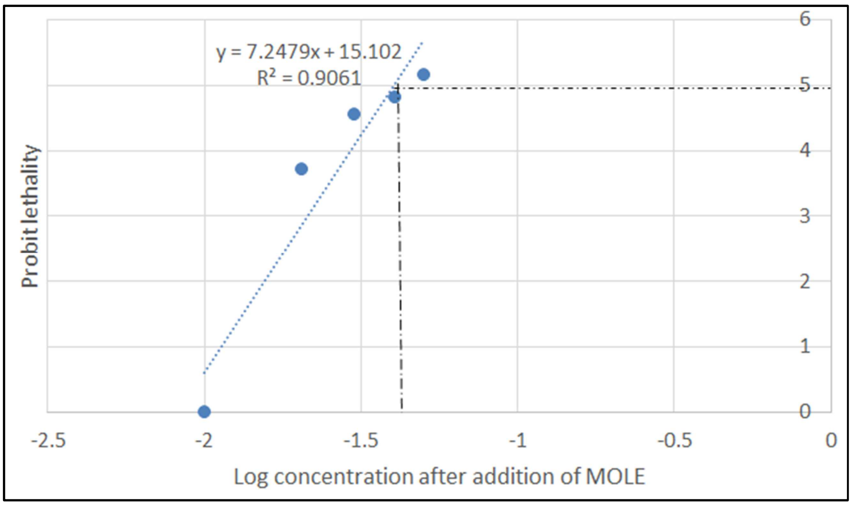

Figure 2. *Logarithmic probit line of 96h LC_{50} of CIWW +MOLE, disclosed to hybrid catfish fingerlings.*

4. Discussion

The initiation of CWW to the aquatic domain of the disclosed hybrid fish, commenced a dose and period behavioral responses to counter the effects of the contaminant [18]. These reaction imply that the CWW brought about some stress to the fish because the control group did not make any effort to move out of their resident. The various behavioural responses observed in this study confirm with previous reports [7, 19]. Noticed dose and time conditioned bearing in opercula oxygenation and tail pulsation regularity was a sign that breathing and vitality of the fish was impeded by the contaminant [20]. The water standard pH and DO could be due the toxicant absorption and use in water, in consonant with reports [21]. The dose dependent lethality frequency of the disclosed fish might be due to abnormal alterations in water quality parameters as well as neural disturbances of the fish living structure. Alterations in phospholipid composition of plasma membrane may have contributed to neural injury correlated with cyanide induced histologic hypoxia [22, 23]. The 96 LC_{50} value in this study is in harmony with [7], who noted a value of 0.024 mg/L.

Moringa seed was proposed to be a satisfactory element of natural water treatment [24, 25]. It has been shown to be a good bio- coagulant and efficient water purifier [26], owing to its water soluble proteins polymers, functional groups, such as hydroxyl and carboxyl [27], and the elimination of heavy metals and microbes from waste water [28-30].

5. Conclusion

2mg/L of MOLE ameliorate the lethal toxicity of CWW on hybrid catfish fingerlings, from 0.028 to 0.044 mg/L, and it is recommended for use in tropical waters for fisheries and aquaculture.

References

[1] Parmar, A., Sturm, B., & Hensel, O. (2017). Crops that feed the world: Production and improvement of cassava for food, feed, and industrial uses. Food Security, 9 (5), 907-927.

[2] Udoro, E. O., Anyasi, T. A., & Jideani, A. I. O. (2021). Process-induced modifications on quality attributes of cassava (Manihot esculenta Crantz) flour. Processes, 9 (11), 1891.

[3] Olukunle, O. J. (2005, October). Development of a cassava peeling machine. In Conference on International Agricultural Research for Development, Tropentag Stuttgart-Hohenheim. October (pp. 11-13).

[4] Kamal, A. R., & Oyelade, O. A. (2010). Present status of cassava peeling in Nigeria. Journal of agricultural Engineering and technology, 18 (2), 7-13.

[5] Saikeaw, N., Rungsardthong, V., Pornwongthong, P., Vatanyoopaisarn, S., Thumthanaruk, B., Pattharaprachayakul, N., & Uttapap, D. (2021). Preparation and properties of biodegradable cat litter produced from cassava (Manihot esculenta L. Crantz) trunk. In E3S Web of Conferences (Vol. 302). EDP Sciences.

[6] Beakou, B. H., El Hassani, K., Houssaini, M. A., Belbahloul, M., Oukani, E., & Anouar, A. (2017). A novel biochar from Manihot esculenta Crantz waste: application for the removal of Malachite Green from wastewater and optimization of the adsorption process. Water Science and Technology, 76 (6), 1447-1456.

[7] Adewoye, S. O., Fawole, O. O., Owolabi, O. D., & Omotosho, J. S. (2005). Toxicity of cassava wastewater effluents to African catfish: Clarias gariepinus (Burchell, 1822). SINET: Ethiopian Journal of Science, 28 (2), 189-194.

[8] Oghenejoboh, K. M. (2015). Effects of cassava wastewater on the quality of receiving water body intended for fish farming. British Journal of Applied Science & Technology, 6 (2), 164.

[9] Wade, J. W., Omoregie, E., & Ezenwaka, I. (2002). Toxicity of cassava (Manihot esculenta Crantz) effluent on the Nile tilapia, Oreochromis niloticus (L.) under laboratory conditions. Journal of Aquatic sciences, 17 (2), 89-94.

[10] Oyewo, O. A., Elemike, E. E., Onwudiwe, D. C., & Onyango, M. S. (2020). Metal oxide-cellulose nanocomposites for the removal of toxic metals and dyes from wastewater. International Journal of Biological Macromolecules, 164, 2477-2496.

[11] Sharma, K., Kumar, M., Waghmare, R., Suhag, R., Gupta, O. P., Lorenzo, J. M., & Kennedy, J. F. (2022). Moringa (*Moringa oleifera* Lam.) polysaccharides: Extraction, characterization, bioactivities, and industrial application. International Journal of Biological Macromolecules.

[12] Kebede, T. G., Mengistie, A. A., Dube, S., Nkambule, T. T., & Nindi, M. M. (2018). Study on adsorption of some common metal ions present in industrial effluents by Moringa stenopetala seed powder. Journal of environmental chemical engineering, 6 (1), 1378-1389.

[13] Suhartini, S., Hidayat, N., & Rosaliana, E. (2013). Influence of powdered *Moringa oleifera* seeds and natural filter media on the characteristics of tapioca starch wastewater. International journal of recycling of organic waste in agriculture, 2 (1), 1-11.

[14] Owodunni, A. A., & Ismail, S. (2021). Revolutionary technique for sustainable plant-based green coagulants in industrial wastewater treatment—A review. Journal of Water Process Engineering, 42, 102096.

[15] APHA (American Public Health Association, American Water works Association and Water Environmental Federation), 2005. Standard Methods of Examination of water and Wastewater. 21st ed. APHA, Washington DC, pp 20001-23710.

[16] Finney, D. J., 1971. Probit Analysis. Cambridge University Press, London. Pp 123-125.

[17] CCREM (Canadian Council of Resources and Environmental Ministry). 1991. Canadian Water Quality Guidelines: Canadian Council of Resouces and Environmental Ministry, Ottawa: Inland Waters Directorate, Environment, Canada.

[18] Gautam, R. K., Banerjee, S., Gautam, P. K., & Chattopadhyaya, M. C. (2014). Remediation technologies for phosphate removal from wastewater: an overview. Adv. Environ. Res, 36 (1).

[19] Adekunle, I. M., Arowolo, T. A., Omoniyi, I. T., & Olubambi, O. T. (2007). Risk assessment in Nile tilapia (Oreochromis niloticus) and African mud catfish (Clarias gariepinus) exposed to cassava effluent. Chemistry and Ecology, 23 (5), 383-392.

[20] ADAMU, H. (2021). COMPARATIVE EFFECT OF COMMERCIAL FEED AND FORMULATED DIETS ON THE GROWTH AND BIOCHEMICAL PARAMETERS OF HETEROBRANCHUS BIDORSALIS (CATFISH) FINGERLINGS (Doctoral dissertation).

[21] Adeyemo, O. K. (2005). Haematological and histopathological effects of cassava mill effluent in Clarias gariepinus. African Journal of Biomedical Research, 8 (3), 179-183.

[22] Oseni, K. (2015). Acute and sub lethal effect of potassium cyanide on the behaviour and ATPase enzyme activity in the freshwater fish, Clarias gariepinus (Catfish). International Letters of Natural Sciences, 49.

[23] Davis, S., Murray, J., & Katsiadaki, I. (2017). Cyanide in the aquatic environment and its metabolism by fish. A Report to Ornamental Aquatic Trade Association, UK.

[24] Vieira, A. M. S., Vieira, M. F., Silva, G. F., Araújo, Á. A., Fagundes-Klen, M. R., Veit, M. T., & Bergamasco, R. (2010). Use of *Moringa oleifera* seed as a natural adsorbent for wastewater treatment. Water, air, and soil pollution, 206 (1), 273-281.

[25] Nand, V., Maata, M., Koshy, K., & Sotheeswaran, S. (2012). Water purification using *moringa oleifera* and other locally available seeds in Fiji for heavy metal removal. International Journal of Applied, 2 (5), 125-129.

[26] Villaseñor-Basulto, D. L., Astudillo-Sánchez, P. D., del Real-Olvera, J., & Bandala, E. R. (2018). Wastewater treatment using *Moringa oleifera* Lam seeds: A review. Journal of Water Process Engineering, 23, 151-164.

[27] Kumar, V., Othman, N., & Asharuddin, S. (2017). Applications of natural coagulants to treat wastewater− a review. In MATEC Web of Conferences (Vol. 103, p. 06016). EDP Sciences.

[28] Njewa, J., Vunain, E., & Biswick, T. (2021). Wastewater clarification and microbial load reduction using agro-forestry and agricultural wastes. Tanzania Journal of Science, 47 (1), 19-33.

[29] Matouq, M., Jildeh, N., Qtaishat, M., Hindiyeh, M., & Al Syouf, M. Q. (2015). The adsorption kinetics and modeling for heavy metals removal from wastewater by Moringa pods. Journal of Environmental Chemical Engineering, 3 (2), 775-784.

[30] Kowanga, K. D., Gatebe, E., Mauti, G. O., & Mauti, E. M. (2016). Kinetic, sorption isotherms, pseudo-first-order model and pseudo-second-order model studies of Cu (II) and Pb (II) using defatted *Moringa oleifera* seed powder. The journal of phytopharmacology, 5 (2), 71-78.