INTERNATIONAL JOURNAL OF PLANT, ANIMAL AND ENVIRONMENTAL SCIENCES

IJPAES

Volume-7, Issue-2 April-June-2017Coden:IJPAJX-CAS-USA, Copyrights@2015ISSN-2231-4490

Received: 4th Mar-2017 ***Revised: 24th April-2017*** ***Accepted: 27th April-2017***

DOI: 10.21276/Ijpaes ***http://dx.doi.org/10.21276/ijpaes*** ***Research Article***

TOXICITY EVALUATION OF WASTEWATER GENERATED FROM AUTOMOBILE SERVICE STATIONS ON JUVENILE *CLARIAS GARIEPINUS*

Pravin C. Singru[1], S.B. Zade[2], Shanta Satyanarayan[3], S.R. Sitre[4]

[1]Research Scholar, PGTD of Zoology, RTM Nagpur University, Nagpur, India
[2]Professor, PGTD of Zoology, RTM Nagpur University, Nagpur, India
[3]Ex-Deputy Director, National Environmental Engineering Research Institute (CSIR-NEERI), Nagpur, India
[4]Asst. Professor, N.S. Science & Arts College, Bhadrawati, Dist. Chandrapur, M.S., India

ABSTRACT: The effect of spent engine oil laden wastewater from automobile service stations on fish *Clarias gariepinus* was evaluated in detail under laboratory conditions. LC_{50}values for 24, 48, 72 and 96 hours were arrived at and were 84, 76, 70 and 64 mg/l respectively. *Clarias gariepinus* being a delicacy it was envisaged to study the toxicity of this wastewater and protect this edible fish. The exposed fish displayed erratic swimming and became lethargic at higher concentrations. Colour of the skin became lighter and in few cases skin became transparent. In some fishes discoloured patches were observed. Tail fin showed curling. Results were subjected to statistical evaluation. Regression (R^2) values varied between 0.983 to 0.991 It is very clear from the studies that this wastewater needs proper and complete treatment prior its discharge. *Clarias gariepinus* being a very sturdy fish also indicated toxicity which clearly shows that other sensitive fishes may not survive in this wastewater.

Key words: Acute toxicity, gariepinus, Spent engine oil wastewater, Edible fish

*Corresponding author: Shanta Satyanarayan, Ex-Deputy Director, National Environmental Engineering Research Institute (CSIR-NEERI), Nagpur, India

INTRODUCTION

Increasing use of automobiles due to unending population growth is the main cause of increase in pollution levels in the environment. Aquatic environment is affected more than the other environments, as aquatic environment is very sensitive and fragile. Our rich faunal population is dwindling gradually and the day is not very far when our precious aquatic species may get extinct, unless proper steps are not taken urgently to control the pollution problems.

Automobile service station's wastewater contains high oil content of 989mg/l and different poly aromatic hydrocarbons (PAH) which exerts high toxicity on aquatic flora and fauna. Apart from the above two major pollutants, this wastewater also contains detergents, grease, phosphate and trace heavy metals. It is reported in literature that this wastewater effluent might contain many other toxicants that are harmful to living organisms because of their detrimental effect on aquatic organisms mainly fishes [1].

Light crude oil is toxic to *Clarias gariepinus* even at low concentrations. The continuous discharge of crude oil from tanker accidents or oil spills leads to severe environmental pollution [2].

Realizing the tremendous adverse effects associated with indiscriminate discharge of crude oil laden wastewater from automobile service stations, it is needed to study its toxic effects on aquatic fauna and thereby control the pollution level to certain desirable levels.

The degree of hazards due to spent crude oil has not been taken very seriously till date, but time now has come to take precautions to contain it. Contamination of water bodies by hydrocarbons present in the spent crude oil/ crude spent diesel oil has been shown to produce subtle changes in fish that are both chronically or briefly exposed [3]. The toxic effects on fish population depends on the amount of oil discharged/ spilled and its characteristics [4]. The fish species and the exposure time period of the spent crude oil also is very important.

Effects of industrial wastewaters on fish and large oil spills in sea affecting marine fauna are reported in literature. When major oil (crude oil) spills in sea spreads very fast in to a slick and reaches the coastal areas and estuaries which create chronic effects. It equally holds good for service station wastewaters also. Sometimes mass fish kills are observed. Fishes have been considered as a useful index for the purity of water and no river or water body should be considered in a satisfactory condition unless fishes thrive well in it [5].

Automobile service station's wastewater pollution has become very common worldwide and particularly in more advanced and affluent countries, where vehicle use is very high particularly in urban regions. Even as low concentration as 0.1 mg/l of crude oil, spent diesel oil can seriously affect fish, crustaceans and zooplankton population on their discharge without treatment. Studies are reported on the crude oil pollution to fish *Clarias gariepinus* and *Clarias anguillaris*. It is further reported that *C. gariepinus* is more tolerant to high levels of crude oil compared to *C. anguillaris* [6,7]

Effect of crude oil on the heamatological changes in Tilapia *Oreochromis niloticus* of Nile river has been reported [8]. Many studies have been carried out on the acute toxicological, histopathological changes in different fishes like Sarotherodon (Black jaw Tilapia) and African Catfish using engine oil, diesel and kerosene [9,10].

Acute toxicity due to spent engine oil on *Clarias gariepinus* has been studied and reported to be exerting genotoxicity [11]. Acute toxicity test using diesel oil on milk fish (*Chanos chanos*) has been studied and its effect on gill pathology has been reported [12]. Acute toxic effects of crude oil on the gills of *Clarias gariepinus* is very common in Niger Delta region, where pollution due to crude oil and refined oil is very common as most refineries in Nigeria are located in the Niger Delta region. Acute toxicity studies on *Clarias gariepinus* has been reported with a 96 hour LC_{50} value of 30.12 mg/l [13].

As the population is growing, wastewater generation from automobile service station is also increasing. Many car washing stations/ garages and service stations are generating huge volumes of wastewater which is disposed into an open channel without treatment. In the past few decades engine oil toxicity has become much more magnified in the field of ecotoxicology. In recent times regional pollution boards are making it mandatory to the large car washing/ automobile service stations to treat the generated wastewater. But small randomly scattered service stations do not treat their wastewater prior its disposal and hence leads to environmental pollution. Engine oil/ spent diesel oil laden wastewater apart from being a health hazard to public has also been found to cause serious damage to the surrounding environment.

Detail studies of coolant wastewater laden with spent engine oil on fish *Lebistes reticulatus* is reported [14]. Vehicle service station wastewater contains waste spent oil which is used for lubrication of various internal combustion engine parts. This oil also serves as a corrosion inhibitor, prevents scaling and prevents corrosion of engine parts [15].

Aquatic toxicity due to water soluble fraction of fuel oil on *O. niloticus* fingerlings has been studied and the report says that water soluble fraction of fuel oil seems to be slightly toxic to Tilapia fingerlings [16]. Recently, impact of automobile service station on common fresh water edible fish Koi carp has been reported [17].

Hence it is very urgently needed to determine their toxicity on the fish and also find out the limit up to which they can be rendered harmless by treatment or by simple dilution. In the context of prevailing conditions in the modern days of fast mechanical life style rapid and reliable methods are required to determine the impact of water pollution levels.

This article discusses in detail the toxicity exerted by the local automobile service station wastewater on edible fish *Clarias gariepinus*.

MATERIALS AND METHODS

For the experimental work juvenile *Clarias gariepinus* fishes were procured from the local fish supplier. Procured fishes were first washed with 0.1% $KMnO_4$ solution to remove dermal infections if any and then acclimatized for fifteen days in a rectangular glass aquarium to the laboratory environment. Dilution water needed for experimental works was prepared using tap water after dechlorination. This dilution water was analyzed as per the standard methods [18] and the results are indicated in Table 1. Experiments were initiated once the fishes were acclimatized as was observed from the swift and healthy movements. Acute toxicity assay was carried out as per the method quoted in literature [19].

Wastewater needed for the experiments was collected from a large automobile service centre on hourly basis for eight hours and a combined well homogenized wastewater was prepared. This homogenized wastewater was subjected to routine physico-chemical and metal analysis and also specialized test for poly-cyclic aromatic hydrocarbons (PAH). Results are indicated in Table 2 and 3.

Acute toxicity tests were initiated once the fishes were well acclimatized to the laboratory environment. A total of ten concentration of the wastewater were used and studied. During the experimental work the aquarium water was renewed every 24 hours with the same concentration to 96 hours. Automobile service station wastewater contains oil in the range of 980-1100 mg/l hence acetone was used to get a homogenized wastewater sample and the same amount of acetone was used in control for comparison. Acetone used for homogenization is around 5.0 ml/litre does not exert any toxicity and normally used for dissolving pesticides and oily samples [20].

It is very urgent to study the toxicity levels of this spent oil laden wastewater from automobile service stations to take necessary steps to treat the wastewater before its discharge in to the water bodies. Poly-cyclic aromatic hydrocarbons, and its metabolites present in this wastewater are considered to be potential carcinogens [21,22]. Considering the high toxic nature of this wastewater, it was envisaged to study its toxicity levels to arrive at a safe disposal concentration by required dilution or by treatment.

RESULTS AND DISCUSSION

The fish *Clarias gariepinus* is found in marshy environment and are air breathers. They are edible and are a delicacy in many communities. Initial stages of acute toxicity test, this fish did not exhibit any uneasy reactions. They swam normally at lower concentrations of the wastewater. At higher concentrations initially they exhibited uneasiness but later they settled at the bottom and exhibited normal movement. But once in a while they came to the surface to breathe normally with quick opercular movements. This behavior of the fish may be due to the presence of toxic materials in the wastewater. Fishes exhibited sheen on their skin and colour of the fishes also became much lighter than their original colour. In lower concentrations patches of discolorization were also observed with curling of tail fin.

From the results it can be inferred that the spent engine oil laden automobile service station wastewater is toxic to test fish. LC_{50} values of *Clarias gariepinus* is much higher than what has been observed in case of Koi carp with same wastewater i.e. LC_{50} values observed for 24, 48, 72 and 96 hours were between 0.8-1.95 mg/l in case of Koi carp while it varied between 64-84 mg/l in case of *Clarias gariepinus*. It hence clearly shows that *Clarias gariepinus* is much sturdier and not very sensitive to this wastewater.

LC_{50} values for 24, 48, 72 and 96 hours were 84, 76, 70 and 64 mg/l respectively as shown in the Table 4. Toxicity nature of this spent oil laden wastewater from an automobile service station to *Clarias gariepinus* is a function of this wastewater concentration and exposure period. Results obtained during acute toxicity test were subjected to statistical evaluation to see the authenticity of the results. Statistical evaluation indicates that the regression (R^2) values ranged between 0.983 to 0.991 indicating good correlation between the fish mortality and wastewater concentration. 95% confidence interval were also calculated and reported [23].

Apart from statistical evaluation safe concentration and safe application rates were also calculated based on the 96 hour LC_{50} values and are of important relevance thereby protecting the aquatic environment and also fish community [24]. Moreover susceptibility of an organism to toxic effluents cannot be judged only by comparing lethal concentrations but should also be considered while assessing the susceptibility of organism/ fish to any toxic effluent.

Table 1: Characteristics of dilution water

Parameters*	Values range
pH	7.3-7.6
Temperature ^{o}C	24.8-24.6
Alkalinity as $CaCO_3$	162-178
Dissolved oxygen	7.0-7.6
Chlorides as Cl	154-162
Total Hardness as $CaCO_3$	168-176
Calcium Hardness as $CaCO_3$	70-74
Magnesium Hardness as $CaCO_3$	98-102
Sodium as Na	40-46
Potassium as K	7-9

* All the values are expressed as mg/l except pH and temperature.

Table 2: Physico-chemical characteristics and metal analysis of wastewater of the vehicle servicing stations

Parameters*	Values (mg/l)
pH	7.45
Alkalinity as $CaCO_3$	375
Chloride as Cl^-	178
Phosphate as PO_4	0.82
Sulphide as S^-	<0.02
Sulphate as SO_4^-	35.0
Sodium as Na	173
Chemical Oxygen Demand (COD)	78
Oil and Grease	989
Magnesium as Mg	17.36
Alkyl benzene sulphonate	2.0
Heavy Metal Range	
Copper	0.024 - 0.279
Cobalt	0.0060 - 0.069
Iron	6.99 - 26.047
Nickle	0.002 - 0.043
Lead	ND
Cadmium	0.001 - 0.021
Chromium	0 - 0.025
Manganese	0.211 - 1.344
Zinc	0.117 - 0.530

* All the values are expressed as mg/l except pH

Table 3: Presence of Poly aromatic hydrocarbons in the wastewater discharged from vehicle service station

Name of the poly aromatic hydrocarbon	Values (Range)
Naphthalene	0 - 32
Acchephthylene	0 - 0.27
Acenophthene	0.33 - 0.84
Fluorene	1.40 - 3.29
Phenanthrene	12.06 - 20.09
Anthracene	1.66 - 10.26
Fluoranthene	0.00 - 2.33
Pyrene	1.54 - 21.65
Benz(a) anthracene	0.43 - 1.69
Chrysene	0.51 - 0.53
Benzo(a)pyrene	0.03 - 2.05
Indenol(1,2,3,-cd)pyrene	0.17 - 0.81
Benzo(g,h,i)pyrelene	0.04 - 0.14

All the values are expressed in μg/l

Table 4: LC_{50} , 95% confidence limit, slope function and regression values

Exp. period	Parameters	Values
24 hrs	NOEC, mg/l	55
	LC_{50}, mg/l	84
	95% confidence interval	78.1 - 90.3
	Slope function	y=8.406x-49.34
	Regression values (R^2)	R^2 = 0.991
48 hrs	NOEC, mg/l	50
	LC_{50}, mg/l	76
	95% confidence interval	69.7 - 82.84
	Slope function	y=9.055x-42.58
	Regression values (R^2)	R^2 = 0.991
72 hrs	NOEC, mg/l	45
	LC_{50}, mg/l	70
	95% confidence interval	63.6 - 77.0
	Slope function	y=9.055x-33.53
	Regression values (R^2)	R^2 = 0.991
96 hrs	NOEC, mg/l	35
	LC_{50}, mg/l	64
	95% confidence interval	57.1 - 71.68
	Slope function	y=8.846x-18.84
	Regression values (R^2)	R^2 = 0.983

SAFE mg/l = 0.388

SAR mg/l = 24.832

Fish toxicity is widely used to study the toxicity of xenobiotics/ carcinogens. Along with fish toxicity now a days toxicity evaluations using zooplanktons are also carried out. Results obtained provide baseline details in formulating strategy for controlled discharge of the treated wastewater in to the receiving water bodies. It also helps in proper/significant dilution required to reduce the toxicity to non toxic level.

For application of toxicity data in regulation of wastewater discharge and prediction of environmental impacts, both acute and chronic toxic levels need to be determined to conserve our precious environment along with zooplankton which is a food for fish and fish community as a whole. Majority of our rural population living by the side of river banks depend on the fish as a source of their protein requirement and hence needs to be protected.

It can be confirmed from the studies that spent engine oil is very toxic to aquatic organisms and needs proper treatment prior its discharge. Toxicity evaluation studies would help in protecting our aquatic as well as land environment. It should be made mandatory to the service stations irrespective of their capacity/ size to treat the water and reduce the toxicity levels.

CONCLUSIONS

It can be inferred from the studies that automobile service station wastewater is quite toxic to the *Clarias gariepinus* in spite of this species being very sturdy. This wastewater must be treated completely to reduce its toxicity prior its discharge. Urgent steps are needed to control its pollution level as this wastewater generation is increasing day by day due to uncontrolled population growth and industrialization.

REFERENCES

[1] Adewoye, S.O., Fawole, O.O., Owolabi, O.D. and Omotosho, J.S., 2005. Toxicity of cassava wastewater effluents to African catfish : *Clarias gariepinus* (Burchell 1822), Ethiop. J. of Sci.,28(2): 189-194.

[2] George Ubang U., Urom Sunday E. and Etanketuk Nseabasi, 2014. Acute toxic effect of Qua-iboe Light crude oil on the gills of *Clarias gariepinus* juveniles. International J. of Environment and Pollution Research, 2(2): 16-30.

[3] Sabo, D.J. and Stegeman, J.J., 1977. Some metabolic effects of petroleum hydrocarbons in marine fish. In: Physiological response of marine biota to pollutants. Verberg F.J. :Calabrese A., Thurbury F.P., Boernberg W. (ed.) Academic Press, New York, pp. 279-287.

[4] Kiihnhold, W.W., 1980. Some aspects of the impact of aquatic oil pollution on fishery resources, FAO/UNDP. South China sea fisheries development and co-ordinating programme, Manilla, Philiphines, pp. 1-26.

[5] Klein, L., 1969. River pollution its causes and effects, Butterworth Publishers, London, 456.

[6] Awoyinka, O.A., Atulomah, E. and Atulomah, N.O.S., 2011. Comparative effects of crude oil on juveniles *Clarias gariepinus* and *Clarias anguillaris*, International Journal of Fisheries & Aquaculture, 3(13): 239-243.

[7] Sunmonu, T.O. and Oloyede, O.B., 2006. Changes in liver enzyme activities in African catfish (*Clarias gariepinus*) exposed to crude oil, Asian Fish Sci., 19: 104-109.

[8] Omuregie, E., 1998. Changes in the hematology of the Nile Tilapia (Oreochromis niloticus) under the effects of crude oil, Acta. Hydrobiol., 84: 287-292.

[9] Ayoola, S.O. and Alajabo, O.T., 2012. Acute toxicity and histopathological effects of engine oil on *Sarotherodon melanotheron* (Black Jaw Tilapia), American Eurasian Journal of Toxicological Sciences, 4(1): 48-55.

[10] Doherty, V.F., Kanife, U.C. and Okeleye, B.T., 2013. Toxicological effects and histopathology of African catfish (*Clarias gariepinus*) exposed to water soluble fractions of diesel and kerosene. Current Advances in Environmental Sciences, 1(2): 16-21.

[11] Ayoola, Simeon Oluwatoyin and Akaeze, C.O., 2012. Genotoxic evaluation and toxicity of spent engine oil on *Clarias gariepinus*, Research J. of Environmental Toxicology, 6: 133-141.

[12] Majid Askari Hesni, Ahmad Savari, Ali Dadolahi Sohrab and Mohammad Sediq Mortazavi, 2011. Gill histapathological changes in milk fish (*Chanos chanos*) exposed to acute toxicity of diesel oil. World Applied Science Journal, 14(10): 1487-1492.

[13] George, Ubong U., Urom, Sunday E. and Etanketuk, Nseabasi, 2014. Acute toxicity of Qua-iboe Light crude oil on the gills of *Clarias gariepinus* juveniles, Int. J. of Environment and Pollution Research, 2(2): 16-30.

[14] Shanta Satyanarayan, Ahana Satynarayan and Sanyogita Verma, 2013. Impact of raw coolant wastewater and effluent from different stages of treatment on fish *Lebistes reticulatus* (peter), Res. J. of Chem. Environ., 17(7).

[15] Koyama, J. and Kakuno, A., 2004. Toxicity of heavy fuel oil, dispersant oil dispersant mixtures to a marine fish, Pagrus Major, 70(4): 587-594.

[16] Dede, E.B. and Kaglo, H.D., 2001. Aqua toxicological effects of watersoluble fractions (WSF) of diesel fuel oil on *O. niloticus* fingerlings, J. Applied Sci. Environ. Mgt., 5(1): 93-96.

[17] Pravin C. Singru, S.B. Zade, Shanta Satyanarayan and S.R. Sitre, 2016. Impact of spent engine oil laden wastewater from automobile service stations on Koi carp (*Cyprinus carpio*), Indian Streams Research Journal, 6(4): 6-12.

[18] Standard Methods for the Examination of Water and Wastewater, 2012. APHA, AWWA, WPCF, 20th edition, Washington D.C.

[19] Sprague J.B., 1969. Measurement of pollutant toxicity to fish. Bioassay Methods for Acute Toxicity, Water Research, 3: 793.

[20] Hattulla, M.L., 1974. Some aspects of the recovery of chlorinated residues (DDT- type compounds and PCB) from fish tissues by using different extraction methods, Bull. Environ. Contam. Toxicol., 12(1): 301.

[21] Catoggio, J.A., 1991. Other organic toxic substance. In: Guidelines of lake management, toxic substances management in lakes and reservoirs, 4: 113-126.

[22] Varanasi, U. and Stein, J.E., 1991. Deposition of xenobiotic chemicals and metabolites in marine organisms, Environ. Health Perspect., 90: 93-100.

[23] Litchfield, J.T. and Wilcoxon, F.A., 1949. A simplified method of evaluating dose effect experiments, J. Pharmacol Exper. Thera., 96: 99-113.
[24] Basak, P.K. and Konar, S.K., 1977. A new method for the determination of safe concentration of insecticides to protect fishes, Ind. J. of Environmental Health, 19(4): 283-292.

INTERNATIONAL JOURNAL OF PLANT, ANIMAL AND ENVIRONMENTAL SCIENCES

ISSN 2231-4490

International Journal of Plant, Animal and Environmental Sciences