



E-ISSN: 2320-7078

P-ISSN: 2349-6800

JEZS 2017; 5(1): 452-458

© 2017 JEZS

Received: 05-11-2016

Accepted: 06-12-2016

Isaac Erhomosele Ehikhamele

Department of Zoology, Faculty of Science, Obafemi Awolowo University, Ile-Ife, Osun State

Sylvester Sunday Ogbogu

Department of Zoology, Faculty of Science, Obafemi Awolowo University, Ile-Ife, Osun State

Assessment of the concentrations of some heavy metals and their effects on the macroinvertebrate composition in Igun southwestern Nigeria, using reference site approach

Isaac Erhomosele Ehikhamele and Sylvester Sunday Ogbogu

Abstract

A survey was conducted from September 2014 to February 2015 to determine the resident aquatic macroinvertebrates and selected heavy metals of Igun and Osu Reservoirs, in Atakumosa West Local Government Area of Osun State, southwestern Nigeria. This was with a view to detecting the effects of gold mining activities on Igun Reservoir using Osu Reservoir as a reference site. Sampling for heavy metals and macroinvertebrate fauna was carried out fortnightly for six months. The collected aquatic macroinvertebrates were preserved in 70% ethanol and identified using appropriate identification keys. Water sample were collected for the determination of heavy metals which include, Cadmium (Cd), Gold (Au), Lead (Pb), Zinc (Zn) and Manganese (Mn), using (Atomic Absorption Spectroscopy) A total of 136 macroinvertebrate specimens representing 17 species in 12 families were collected from Igun Reservoir. Class Gastropoda had the highest number of specimens 61(44.85%), representing five species in three families (Planorbidae, Thiariidae and Physidae). A total of 408 macroinvertebrate specimens representing 32 species in 16 families were collected from Osu Reservoir. Ephemeroptera, represented by six species from two families (Caenidae and Baetidae) had the highest number of specimens, 136 (33.33%). Simpson Diversity Index (1-D) for Igun and Osu Reservoirs were 0.84 and 0.89 respectively. Cd correlated with Mn ($r = 0.82$) while Pb correlated with Mn ($r = 0.62$). Pb also negatively correlated with the abundance of Odonata ($r = -0.77$) in Igun Reservoir while Cd correlated negatively with the abundance of Coleoptera ($r = -0.58$) in Osu Reservoir. The concentration of zinc was within the WHO permissible limits for freshwater while other heavy metals were above the limits in both reservoirs.

Keywords: Heavy metals, macroinvertebrate composition, aquatic macroinvertebrates

1. Introduction

Aquatic insects are very good indicators of water quality since they have various environmental disturbance tolerance levels [1]. There are some that are very vulnerable and sensitive to contamination, while some can survive and boom in contaminated and unhealthy waters [2]. Pollutants in the surrounding watershed affect aquatic invertebrate existence depending on their toxicity, bioavailability and rate of uptake, and metabolic regulation by specific organisms [3]. Studies of the effects of metal contamination on macrobenthic assemblages have identified some common themes [4]. In any type of environmental pollution, taxonomic sensitivity distribution controlled assemblage dynamic. Increase in the level of the pollutants result in the elimination of the taxa that is most sensitive in the population and are replaced by the ones that can tolerate the disturbance, thereby changing the composition structure. As the contaminants increase, even taxa with the most resistivity will be reduced in abundance, even to zero [4]. Lotic organisms are generally known to be affected by heavy metal contaminations [5]. Different researchers [4] and [6] advocated the use of species sensitivity to contaminants of benthic macroinvertebrate assemblages to assess ecological effects of metal pollution in streams and the impacts of heavy metal contamination on leaf-litter breakdown [7] and secondary production of macroinvertebrates [8].

The aim of this study is to determine the faunal composition of the resident aquatic macroinvertebrates and heavy metal concentrations in Igun and Osu Reservoirs in Atakumosa West Local Government Area of Osun State. It is also to correlate the heavy metal concentrations with macroinvertebrate diversity, and compare the findings in both waterbodies. This was with a view to determine the effects of environmental perturbations in the reservoirs.

Correspondence

Isaac Erhomosele Ehikhamele

Department of Zoology, Faculty of Science, Obafemi Awolowo University, Ile-Ife, Osun State

2. Materials and Methods

Igun community lies between Latitudes 07°30' and 07°35' N and between Longitudes 004°38' and 00 4°42' E in Atakumosa West Local Government Council of Osun State, southwestern Nigeria (Fig. 1). The study area is a rural community of about 2,600 people that engage predominantly in subsistent farming of cocoa [9]. The city is accessible through an untarred but motorable road, and a gold mining area is within one of the six (6) classes of the basement complex rock that is from slightly migmatized to non-migmatized, meta-sedimentary and meta-igneous rock, simply called the Schist belt [10]. The geological attributes of Igun Community can be found in [11]. The eastern part has

quartzite, quartz schist and amphibole schist. The gold deposit occurs in the eastern area. Igun Reservoir is located within Igun Community. It is formed as a result of the activities of miners. There are about nine reservoirs in Igun community which the artisanal miners use for their mining activities (After excavating the ground, they take the mines tailings which is always mud-like to the reservoir to wash and finally extract the material they are looking for) but most of them have been taken over by aquatic vegetation. Two of these reservoirs still retain sufficient water, open and accessible but one of them was used for this study because mining activities still occur very close to the reservoir.

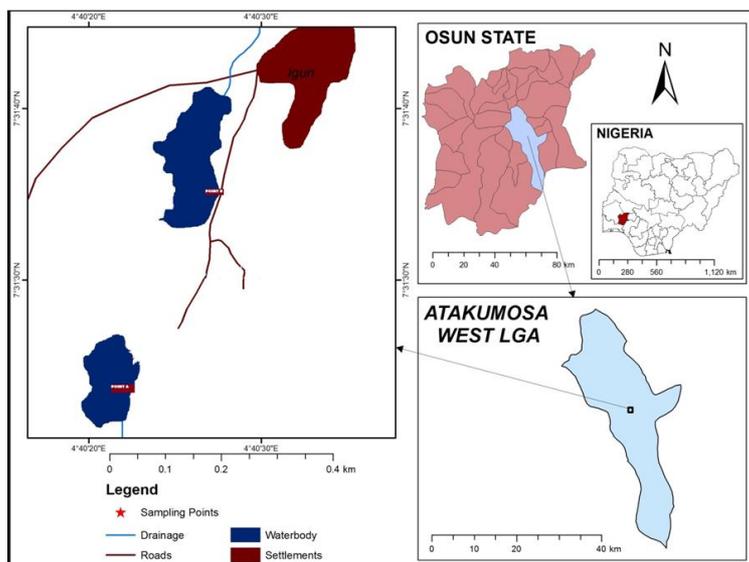


Fig 1: Study sites in the Igun Reservoirs, Atakumosa West Local Government Area of Osun State, Nigeria. A – Map of Osun State showing Atakumosa West Local Government Area (Inset is map of Nigeria) B; Igun Community and C; Igun Reservoir.

Osu Reservoir is also located in Atakumosa West Local Government Area. The reservoir was formed by the impoundment of River Sasa with an embankment type of dam in the year 2005 (Fig. 2). The surface area of the reservoir is about 0.118 km² while the maximum capacity is about 102.4m³ [12]. The substratum of the reservoir is mainly mud

and sand. Shoreline vegetation is dense with submerged aquatic macrophytes, some of which eventually decompose during the wet season. The reservoir was chosen as reference site because it is used mainly for domestic for domestic water supply to rural dwellers

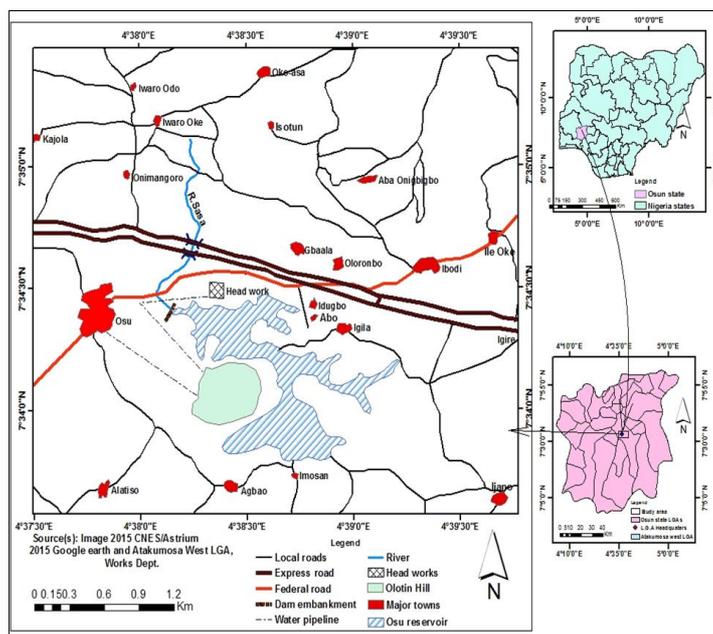


Fig 2: Osu community showing the study site in Osu Reservoir, Atakumosa West Local Government Area of Osun State, Nigeria. A- Sampling site in the reservoir

Macroinvertebrates were sampled every two weeks between September 2014 to February 2015 from Igun and Osu Reservoirs. A standard dipnet of 500µm mesh size was used for macroinvertebrates collection, while organisms attached to small pieces of rock from the substratum were handpicked. Sampling was done between the hours of 08:00 am and 09:00 am. Sorting of collected samples was done immediately on the site, and in situations where sorting could not be done, samples were washed into a bucket and taken to the laboratory to be sorted. Sorted samples were then stored in vials with 70% ethanol for identification subsequently. Identification of specimens was carried out down to the lowest taxonomic level where possible with the aid of a high power microscope (Carl Zeiss Microscopy GmbH 37081 Göttingen, Germany) using the Guides to the Freshwater Invertebrates of Southern Africa ^{[13] [14] [15] [16]}. Water samples were collected fortnightly from Igun and Osu Reservoirs for a period of 6 months for heavy metal analysis. To determine the concentrations of heavy metals; Cadmium (Cd), Gold (Au), Lead (Pb), Zinc (Zn) and Manganese (Mn), water samples were determined with nitric perchloric hydrofluoric acid digestion method ^[17]. The digested samples were analyzed for the heavy metals using atomic absorption spectroscopic (AAS) method at the appropriate wavelengths.

Pearson Correlation was used to examine the interrelationships among the heavy metals. Analysis of variance (ANOVA) was used to determine the interactions between the heavy metals and abundance of macroinvertebrates, while Student's t-test was used to compare the means of the heavy metals across the study sites

3. Results

The mean concentration of cadmium recorded at Igun Reservoir was (0.02 ± 0.19mg/L) and the mean concentration of cadmium recorded at Osu Reservoir was (0.01 ± 0.00mg/L). There was a significant difference ($p < 0.001$) in the mean concentration of cadmium in both reservoirs. The recorded mean concentration of Gold during the period of study ranged from 0.001 to 0.832 as presented in Table 1. The mean concentration of Gold recorded at Igun was (0.49 ± 0.25mg/L) while the mean concentration of Gold recorded at Osu Reservoir was (0.00 ± 0.00mg/L). Significant difference ($p < 0.001$) was observed in the mean concentration of gold in

Igun and Osu reservoirs. The concentration of lead during the study ranged from 0.002 to 0.076. The mean concentration of lead recorded at Igun reservoir was (0.04 ± 0.03mg/L) and the mean concentration of lead recorded at Osu reservoir was (0.01 ± 0.01mg/L). There was a significant difference ($p < 0.001$) in the mean concentration of lead in Igun and Osu Reservoirs. The mean concentration of zinc recorded at Igun Reservoir was (0.15 ± 0.06mg/L) and the mean concentration of zinc recorded at Osu Reservoir was (0.06 ± 0.04mg/L) was recorded at Osu Reservoir, hence, there was a significant difference ($p < 0.001$) in the value of Zinc in both reservoirs. The concentration of Manganese during the study ranged from 0.009 to 0.816. The mean concentration of manganese recorded at Igun Reservoir was (0.30 ± 0.27mg/L) while mean concentration of manganese recorded at Osu Reservoir was (0.05 ± 0.03mg/L) was recorded at Osu Reservoir. There was a significant difference ($p < 0.01$) in the mean concentration of Zinc in both reservoirs (Table 1).

The mean values of all the selected heavy metals are significantly different ($p < 0.001$) in both Igun and Osu Reservoirs (Table 1).

3.1 Abundance and Species richness of macroinvertebrates

A total of 136 individuals representing 17 species in 12 families were collected during this study from Igun Reservoir. Gastropoda had the highest number of individuals 61(44.85%) (Table 2) representing five species in three families (Planorbidae, Thiariidae and Physidae) and the least was from the dipteran order, having 6 (4.41%) individuals representing one species from a single family Chironomidae. A total of 408 individuals representing 32 species in 16 families were collected during this study from Osu Reservoir. Ephemeroptera had the highest number of individuals, 136 (33.33%) (Table 2) representing six species from two families (Caenidae and Baetidae). The least was from coleopteran order, having 19 (4.66%) individuals representing one species from a single family Gyrinidae. The overall species richness and diversity of macroinvertebrates collected in the sampled sites are indicated in Table 2. Osu reservoir had the highest species richness with Simpson diversity index value of 0.89 compared to Igun with 0.84.

Table 1: Analysis of variance (ANOVA) showing relationships among the selected heavy metals in Igun and Osu Reservoirs

Conc. Of heavy metals (mg/L)	Igun		Osu		Anova		Student t - test	
	Min - Max	Mean ± S.D	Min - Max	Mean ± S.D	F	p	T	p
Cadmium	0.011 - 0.074	0.02 ± 0.19	0.002 - 0.011	0.01 ± 0.00	14.87	0.0008571 ***	3.11	0.00 ***
Gold	0.101 - 0.832	0.49 ± 0.25	0.001 - 0.008	0.00 ± 0.00	40.36	0.000002156 ***	7.94	0.00 ***
Lead	0.01 - 0.076	0.04 ± 0.03	0.002 - 0.021	0.01 ± 0.01	17.09	0.0004348 ***	3.89	0.00 ***
Zinc	0.092 - 0.321	0.15 ± 0.06	0.005 - 0.093	0.06 ± 0.04	20.19	0.0001807 ***	4.50	0.00 ***
Manganese	0.11 - 0.816	0.30 ± 0.27	0.009 - 0.093	0.05 ± 0.03	9.578	0.005289 **	3.19	0.00 ***

*** Very highly significant ($p < 0.001$)

** Highly significant ($p < 0.01$)

* Significant ($p < 0.05$)

Table 2: Composition, distribution and abundance of macroinvertebrates collected in Igun and Osu Reservoirs.

Phylum	Class	Order	Family	Species	Igun	Osu	
Arthropoda	Insecta	Ephemeroptera	Caenidae	<i>Afrocaenis major</i>	0	41	
				<i>Caenis bermeri</i>	0	27	
				<i>Caenis carpensis</i>	0	6	
				<i>Caenospella major</i>	0	22	
				Baetidae	<i>Acanthiop sp</i>	0	32
					<i>Baetis sp</i>	0	8
						0	136(33.33%)
			Odonata	Synlestidae	<i>Chlorolestes conspicuus</i>	0	12

				<i>Chlorolestes elegans</i>	8	20
				<i>Chlorolestes apricans</i>	0	2
				<i>Chlorolestes sp.</i>	8	6
			Lestidae	<i>Lestes plagatus</i>	1	0
				<i>Lestes ictericus</i>	0	12
			Gomphidae	<i>Ophiogomphus sp.</i>	0	6
				<i>Actinogomphus ferox</i>	0	3
				<i>Ceratogomphus triceratus</i>	6	13
				<i>Actinogomphus sp.</i>	0	3
			Libellulidae	<i>Trithemis dorsalis</i>	3	0
				<i>Trithemis sp.</i>	0	7
				<i>Diplacodes sp</i>	5	0
			Coenagrionidae	<i>Pseudagrion inopinatum</i>	0	4
				<i>Ceragrion bakeri</i>	0	5
			Platyncnemididae	<i>Allocnemis leucostita</i>	2	0
				<i>Metacnemis leucostita</i>	0	6
					33(24.26%)	99(24.26%)
		Diptera	Chironomidae	<i>Chironomous sp.</i>	6	0
					6(4.41%)	0
		Coleoptera	Gyrinidae	<i>Orectogyrus sp.</i>	22	19
					22(16.18%)	19(4.66%)
		Hemiptera	Notonectidae	<i>Notonecta lunata</i>	6	0
				<i>Notonecta sp.</i>	0	105
			Naucoridae	<i>Illycoris sp.</i>	0	6
				<i>Pelecoris sp.</i>	5	0
			Belastomatidae	<i>Belastoma sp.</i>	0	1
				<i>Abedus sp.</i>	0	1
			Hydrometridae	<i>Hydrometra sp.</i>	3	1
			Gerridae	<i>Gerris sp.</i>	0	4
					14(10.29%)	118(28.29%)
Mollusca	Gastropoda	Heterobranchia	Planorbidae	<i>Bulinus sp.</i>	0	2
				<i>Bulinus africanus</i>	0	6
				<i>Planorbis sp.</i>	8	5
				<i>Helisoma sp.</i>	4	1
				<i>Lentobis sp.</i>	2	0
			Physidae	<i>Physella sp.</i>	2	0
		Caenogastropoda	Thiaridae	<i>Melanoides tuberculata</i>	45	22
					61(44.85%)	36(8.82%)
Total					136	408
Simpson diversity index					0.84	0.89

3.2 Relationships among heavy metals and macroinvertebrates in Igun and Osu Reservoirs

Table 3 and 4 show the relationships among the heavy metals and aquatic macroinvertebrates. In Igun Reservoir, Cd significantly correlated with Mn (r = 0.82), Pb significantly

correlated with Mn (r = 0.62). Also there is a significant negative correlation between Pb and the abundance of Odonata (r = -0.77) while Cd correlated negatively with the abundance of coleoptera (r = -0.58) in Osu reservoir (Table 4)

Table 3: Correlation (r) values showing the interaction between selected heavy metals as well as macroinvertebrates fauna in Igun reservoir.

	Cd	Au	Pb	Zn	Mn	Mollusca	Hemiptera	Diptera	Odonata	Coleptera
Cd	0									
Au	0.37	0								
Pb	0.38	0.17	0							
Zn	0.45	0.16	0.52	0						
Mn	0.82*	0.26	0.62*	0.8	0					
Mollusca	-0.19	-0.39	-0.42	-0.03	-0.22	0				
Hemiptera	-0.17	-0.08	-0.55	-0.26	-0.3	0.55	0			
Diptera	-0.14	-0.46	-0.31	0.03	-0.14	0.98*	0.55	0		
Odonata	-0.43	-0.19	-0.77*	-0.42	-0.57	0.46	0.08	0.32	0	
Coleptera	-0.14	-0.46	-0.31	0.03	-0.14	0.98*	0.55	1	0.32	0

*Significant correlation, p<0.05.

Table 4: Correlation (r) values showing the interaction between selected heavy metals as well as macroinvertebrates fauna in Osu reservoir

	Cd	Au	Pb	Zn	Mn	Mollusca	Hemiptera	Odonata	Coleptera	Ephemeroptera
Cd	0									
Au	0.4	0								
Pb	0.26	-0.43	0							
Zn	0.01	0.01	0.15	0						
Mn	0.09	0.01	-0.11	-0.14	0					

Mollusca	-0.53	-0.03	-0.23	0.11	-0.19	0				
A	-0.1	-0.18	-0.28	0.25	-0.23	0.58*	0			
Odonata	-0.49	-0.05	-0.31	0.23	-0.23	0.88*	0.85*	0		
Coleptera	-0.58*	0.1	-0.32	0.24	-0.19	0.81*	0.64*	0.92*	0	
Ephemeroptera	0.03	0.48	-0.34	0.01	-0.04	0.62*	0.41	0.57	0.51	0

* Values that are significant ($p < 0.05$)

4. Discussion

The condition of the reservoir at Igun Community presents the status of a waterbody that is greatly impaired by gold mining activities. This is seen in the type of macroinvertebrates collected at the reservoir. The reservoir has *Chironomus* sp. which is characterized by its tolerance to pollution and high organic matter concentrations as reported for some impacted ecosystems [18] [19]. Low abundance of Planorbidae present in Igun Reservoir showed that mining activities has caused a deterioration of the health of the water body. Fewer species of Planorbidae were collected in Igun than Osu Reservoir, corroborating the fact that they are seldom found in severely polluted waters [20]. In addition, earlier findings had shown that heavy metals reduce the diversity of planorbid snails in a waterbody [21]. Results showed that there were low diversity and abundance of semi-pollution-tolerant macroinvertebrates at Igun such as *Orectogyrus* sp. Previous studies demonstrated that discharge from mined areas decrease species diversity and altered species composition [22] [23] [24] [25] [26]. Gold-mining activities also lead to the loss of physical habitats such as macrophytes and mineral substrates. Plants such as aquatic macrophytes provide direct refugia or indirect (support for the development of algae and biofilm that constitute food) resource supplies [27]. Macroinvertebrates sampled at Osu Reservoir however showed that a pollution sensitive order, Ephemeroptera had the highest abundance, accompanied by Libellulidae (Odonata). These taxa usually indicate clean, unpolluted waters [21].

The range of concentrations of Mn, Cd, Au, Pb in Igun Reservoir were above the [28] [29] permissible limits for drinking water. However, the concentration range of zinc in Igun Reservoir (0.09 – 0.321mg/L) were within the [28] permissible limits for drinking water. Zinc is one of the important trace elements that play a vital role in the physiological and metabolic processes of many organisms. Nevertheless, higher concentrations of zinc can be toxic to the organism [30]. The values of all the heavy metals at Igun were significantly higher than those of Osu Reservoir. The results clearly show that heavy metal pollution adversely affected macroinvertebrate populations and alters their community in Igun Reservoir. In particular, total abundance and taxa richness of Ephemeroptera were dramatically decreased even at the polluted sites. The sensitivity of Ephemeroptera to heavy metals is well documented from previous studies [31] [4]. Heavy metals may affect benthic invertebrates indirectly through alteration of habitat conditions [32] or trophic relationships [33] [34] and directly through water contamination. Mining activity often cause sedimentation and increases substratum embeddedness [35]. According to [36] [37] [38], the tolerance of benthic invertebrates to heavy metals decreases from Trichoptera to Plecoptera and Ephemeroptera. [39] [40] [41] reported that the reduction of potential habitats, the low water discharge, slight organic pollution and the accumulation of heavy metals in water and sediments led to the deterioration of water quality. The deleterious effects of the contaminating heavy metals on macroinvertebrate communities at Igun Reservoir resulted in reduced species richness and changes in the community compositions.

Lead showed a negative correlation with abundance of odonates in Igun Reservoir. Lead is a cumulative toxic metal and it poses a threat to the aquatic environment, the most severe effect being brain damage caused by lead poisoning. [42] reported that lead has been found to be carcinogenic and a potent enzyme inhibitor as it inhibits utilization of iron in the body, interferes with fertility and causes renal damage. Given these, it is possible that odonates were greatly affected by the concentration of lead in the reservoir leading to low abundance. The toxicity of lead to aquatic organisms varies considerably depending on availability, uptake, and species sensitivity. Generally, the earlier life stages are more vulnerable [43]. According [44] the tolerance limits of lead for aquatic life are between 0.0013-0.077 mg/L. The values of lead in Igun was above the USEPA limit. In communities of aquatic invertebrates, some populations are more sensitive than others and community structure may be adversely affected by lead contamination. However, it has been reported that populations of invertebrates from polluted areas can show more tolerance to lead than those from non-polluted areas [43]. High concentration of heavy metals in Igun Reservoir may be responsible for low diversity of macroinvertebrates and this is corroborating by [45] report that active mining sites cause lower densities of macroinvertebrates. Low concentration of heavy metals in Osu Reservoir possibly explains the high diversity and abundance of macroinvertebrates. Also, the presence of pollution-sensitive species such as EPT taxa indicates that the water is somewhat in pristine condition. The EPT taxa have been treated as indicators for good water quality for a long time [46]. Recent findings show that EPT abundance was higher in semi-natural compared to heavily disturbed active mining sites [45].

5. Conclusion

To conclude, this study assessed the effect of heavy metal concentrations on macroinvertebrate fauna of a reservoir in a gold mining area and compared it with that of a reference site, Osu Reservoir. The concentrations of heavy metals analyzed in Igun reservoir were above the recommended values by WHO, except zinc, which was within the permissible limits. Low abundance of macroinvertebrates indicates the biological status of the reservoir due to the mining activities. It is very expedient to protect the quality of the water and biota from further degradation.

6. References

1. Arimoro FO, Ikomi RB. Ecological Integrity of upper Warri River, Niger-Delta using aquatic insects as bioindicators. Ecological Indicators. 2008; 395:17.
2. Merritt RW, Cummin KW. An introduction to the aquatic insects of North America. 3rd ed. Kendall/Hunt Publishing Company, 1996.
3. Rainbow PS. Trace metal concentrations in aquatic invertebrates: Why and so what? Environmental Pollution. 2002; 120:497-507.
4. Clements WH, Carlisle DM, Lazorchak JM, Johnson PC. Heavy metals structure benthic communities in Colorado mountain streams. Ecological Application. 2000; 10:626-638.

5. Newman MC, McIntosh AW. Metal Ecotoxicology: Concepts and Applications. Lewis, Boca Raton, FL, U.S.A, 1991.
6. Maret TR, Cain DJ, MacCoy DE, Short TM. Response of benthic invertebrate assemblages to metal exposure and bioaccumulation associated with hard- rock mining in northwestern streams, U.S.A. *Journal of the North American Benthological Society*. 2003; 22:598-620.
7. Schultheis AS, Sanchez M, Hendricks AC. Structural and functional responses of stream insects to copper pollution. *Hydrobiologia*. 1997; 346:85-93
8. Carlisle DM, Clements WH. Growth and secondary production of aquatic insects along a gradient of Zinc contamination in Rocky Mountain streams. *Journal of the North American Benthological*. 2003; 22:582-597.
9. Ayantobo OO, Awomeso JA, Oluwasanya GO, Bada BS, Taiwo AM. Gold mining in igun-ijesha, southwest Nigeria: Impacts and implications for water quality. *American Journal of Environmental Sciences*. 2014; 10(3):289-300.
10. Ademeso OA, Adekoya JA, Adetunji A. Further evidences of cataclasis in the Ife-Ilesa schist belt, southwestern Nigeria. *Journal of Natural Science Research*. 2013; 11:50-59.
11. Tropical Mines Limited. A Pre-Investment Study of the Primary Goldmine-Odo Ijesa (primary) Gold Deposit Report. 1996; 2:1-12.
12. Ogunribido AO. Some aspects of the biology of fish species inhabiting newly impounded Osu reservoir, Atakumosa local Government Osun State. M.Sc thesis submitted to the department of Zoology, Obafemi Awolowo University, Ile-Ife, Nigeria. 2015.
13. Day JA, de Moor IJ. Guides to the Freshwater of Southern Africa. Arachnida & Mollusca. 2002; 6:42-121.
14. de Moor IJ, Day JA, de Moor FC. Guides to Freshwater of Southern Africa, 2003; 7(1):6-212.
15. Day JA, Harrison AD, de Moor IJ. Guides to the Freshwater of Southern Africa, 2003; 9:11-174
16. Stals R, de Moor IJ. Guides to the Freshwater of Southern Africa, 2007, 10. Coleoptera.
17. Ademoroti CMA. Standards for water and effluent analysis foludex limited, Ibadan, 1996.
18. Marques MMGSM, Barbosa FAR, Callisto M. Distribution and abundance of Chironomidae (Diptera, Insecta) in an impacted watershed in south-east Brazil. *Revista Brasileira de Biologia*. 1999; 59:553-561.
19. Helson JE, Williams DD, Turner D. Larval Chironomid community organization in four tropical rivers: human impacts and longitudinal zonation. *Hydrobiologia*, 2006; 559:413-431
20. Pennak RW. Freshwater invertebrates of the United States: Protozoa to Mollusca. 3rd ed. John Wiley & Sons, Inc., New York. 1989.
21. Last LL, Whitman RL. Aquatic macroinvertebrates of the Grand Calumet River. *Proceedings of the Indiana Academy of Science* ISSN 0073-6767, 1999.
22. Beltman DJ, Clements WH, Lipton J, Cacula D. Benthic invertebrate, metals exposure, accumulation, and community level effects downstream from a hardrock mine site. *Environment Toxicology and Chemistry*. 1999; 18:299-307.
23. Malmqvist B, Hoffsten PO. Influence of drainage from old mine deposits on benthic macroinvertebrate communities in central Swedish streams. *Water Resources*. 1999; 33:2415-2423.
24. Soucek DJ, Cherry DS, Trent GC. Relative acute toxicity of acid mine drainage water column and sediments to *Daphnia magna* in the Puckett's Creek watershed, Virginia, USA. *Archive Environmental Contamination Toxicology*. 2000; 38:305-310.
25. Tarras-Wahlberg NH, Flachier A, Lane SN, Sangfors O. Environmental impacts and metal exposure of aquatic ecosystems in rivers contaminated by small- scale gold mining: the Puyango River basin, southern Ecuador. *Science of Total Environment*. 2001; 278:239-261.
26. Yule CM, Boyero L, Marchant R. Effects of sediment pollution on food webs in a tropical river (Borneo, Indonesia). *Marine of Freshwater Resources*. 2010; 61:204-213.
27. Allan JD, Castillo MM. *Stream Ecology, Structure and function of running waters*, seconded. Springer, Dordrecht, The Netherlands and Sons, Inc, New York, 2007.
28. World Health Organisation (WHO) Guidelines for drinking-water quality, second edition, 1996, 2.
29. United Nations Environment Programme. *Global Drinking Water Quality Index Development and Sensitivity Analysis Report*. 2007.
30. Rajkovic MB, Lacnjevac CM, Ralevic NR, Stojanovic MD, Toskovic DV, Pantelic GK *et al*. Identification of (heavy and radioactive) in drinking water by indirect analysis method based on scale tests. *Sensors*, 2008; 8:2188-2207.
31. Clements WH. Benthic invertebrate community responses to heavy metals in the upper Arkansas River basin, Colorado. *Journal of the North American Benthological Society*. 1994; 13:30-44.
32. Courtney LA, Clements WH. Assessing the influence of water and substratum quality on benthic macroinvertebrate communities in a metal-polluted stream: An experimental approach. *Freshwater Biology*. 2002; 47:1766-1778.
33. Farag AM, Woodward DF, Goldstein JN, Brumbaugh W, Meyer JS. Concentrations of metals associated with mining waste in sediments, biofilm, benthic macroinvertebrates, and fish from the Coeur d'Alene River basin, Idaho. *Archives of Environmental Contamination and Toxicology*. 1998; 34:119-127.
34. Fleeger JW, Carman KR, Nisbet RM. Indirect effects of contaminants in aquatic ecosystems. *Science of Total Environment*. 2003; 317:207-233.
35. Church SE, Kimball BA, Fey DL, Ferderer DA, Yager TJ, Vaughn RB. Source, transport, and partitioning of metals between water, colloids, and bed sediments of the Animas River, Colorado. U.S. *Geological Survey*, Survey Open-File Report. 1997; 135:97-151.
36. Chadwick JS, Canton RD. Recovery of benthic invertebrate communities in Silver Bow Creek, Montana, following improved metal mine wastewater treatment. *Water, Air and Soil Pollution*, 1986; 28:427-438.
37. Clements WD, Cherry J, Van Hassel. Assessment of the Impact of Heavy metals on benthic communities at the Clinch River (Virginia): Evaluation of an Index of Community Sensitivity – *Canadian Journal of Fisheries and Aquatic Science*, 1992; 49:168-1694.
38. Garsia-Criado F, Fernandez-Alaez M, Fernandez-Alaez C. Relationship between benthic assemblage structure and coal mining in the Boeza River Basin (Spain). *Archiv für Hydrobiologie*, 2002; 154(4):665-689
39. Stoyanova T, Traykov I, Yaneva I, Bogoev V. Ecological

- quality assessment of Luda River, Bulgaria. *Natura Montenegrina*, Podgorica, 2010; 9(3):341-348.
40. Stoyanova T, Traykov Yaneva, II, Bogoev V. Heavy metals and radionuclides in river impacted by uranium mining, Bulgaria. *Journal of Balkan Ecology*. 2011; 14 (1):83-91.
 41. Stoyanova T, Traykov I, Kenarova A, Bogoev V, Yaneva, I. The Impact of abandoned uranium mine Senokos on Luda River, Pirin Mountain, Bulgaria. *Balwois* 2012.
 42. Greenberg AE, Clesceri LS, Eaton AT. Standard Methods for the examination of water and wastewater, 18th ed. *American Public Health Association*. 1992, 490-596.
 43. Girgin S. Evaluation of the benthic macroinvertebrate distribution in a stream environment during summer using biotic index. *International Journal of Environmental Science and Technology*. 2010; 7:11-16.
 44. United States Environmental Protection Agency (USEPA) Current national recommended water quality criteria, 2005.
 45. Rutebuka Evariste, Patteson M Chula, Paul L. S Yáñez. The effect of illegal small-scale gold mining on stream macro-invertebrate assemblages in the East Usambara Mountains. *Tropical Biology Association*. 2013.
 46. Raburu PO, Masese FO, Mulanda CA. Macroinvertebrate Index of Biotic Integrity (M-IBI) for monitoring rivers in the upper catchment of Lake Victoria Basin, Kenya. *Aquatic Ecosystems and Health Management*. 2009; 12:1-9.