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Toxicity of Leachates from the Aba-Eku Landfill Leachate Lagoon, Ibadan, South-Western Nigeria

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ABSTRACT

Landfill monitoring is necessary to avoid adverse environmental impacts. Toxicity tests are now incorporated into landfill hazard assessment. Toxicity of leachate in a lagoon draining the Aba-Eku landfill, Ibadan, Nigeria was investigated using bioassays. 96hr bioassays were carried out using arthropods (Insecta - larvae of Chironomus sp.; Culex pipiens); amphibians (tadpoles of Bufo regularis); and fish (seven and fourteen weeks old fingerlings of Clarias gariepinus). Physico-chemical parameters were analysed using American Public Health Association standard methods. Toxicity results showed reduced mortality in Chironomus sp.(10%); Culex pipiens however showed a reduced toxicity with increasing leachate concentration. 96 hr LC_{50} in Culex pipiens was 20% leachate. No mortality of leachate was observed in tadpoles of Bufo regularis and fourteen week old fingerlings of Clarias gariepinus. Percentage mortality in seven week old fingerlings of Clarias gariepinus was 37.5%. Filtering of leachate reduced mortality in C. gariepinus from 37.5% to 12.5%. Due to the low mortality, LC_{50} for all organisms except Culex pipiens were indeterminate. The observed mortalities were not concentration dependent. Toxicity test results were supported by leachate analysis which showed considerable reduction in contaminant levels except for suspended solids (144.94 ± 117.99 mg/l and chloride (597.98 ± 209.73 mg/l) which exceeded and approached regulatory limits respectively. Leachate strength and toxicity was reduced by natural attenuation. Absence of a dose dependent response may be due to the leachate's unstable nature. Sediment presence may have reduced toxicity via contaminant sorption. Low-cost attenuation processes are useful in reducing contaminant levels in leachate.

Keywords: *Chironomus sp., Clarias gariepinus, Culex pipiens,* Landfill Leachate, Toxicity,

INTRODUCTION

Leachate is described as the water based solution of compounds from solid wastes dumped in a landfill [1]. It is known to contain a variety of naturally forming and anthropogenic chemicals which may be toxic to a wide variety of organisms. The leachate takes up organic and inorganic constituents and thus contains a high concentration of organic and inorganic ions, including heavy metals [2]. In many developing countries, wastes are dumped in unlined landfills and the leachates discharged without treatment into the environment. This has implications for the environment, particularly for organisms in surface water receptors into which the leachates are discharged. The quality of leachate is often assessed through physico-chemical methods and the results compared to thresholds stipulated by the regulatory authorities. However, increasing attention has focused on the incorporation of toxicity tests in hazard evaluations of waste dumps. The complex nature of landfill effluents requires that a battery of bioassays be used to assess toxicity [3]. Some criteria that have been used in the selection of organisms in a test battery include different groups, different habitats and different trophic levels [4].

The Aba-eku landfill site is the major repository of municipal solid wastes in Ibadan. It receives domestic, industrial and institutional waste by public and private waste management operators [5]. It has been an open dump in use since 1994. Residents have reported the death of some domestic animals in 1996, which they attributed to the impacts of waste disposal in the landfill. Following these reports, efforts were undertaken to improve and upgrade the landfill site. This resulted in its closure for upgrading efforts and subsequent re-opening for use in 1999. As part of the upgrading efforts, a leachate collection and removal system (lagoon) was designed and is located 250m down-gradient of the active fill area. The Ibadan Solid Waste Management Authority (ISWMA) claims that the leachates are pre-treated before discharge into the Omi stream, which flows from the landfill towards Aba-eku community, located 600m away from the site.

The discharge of the leachate from the lagoon into this stream has implications for the organisms present. Literature search showed that leachates draining into the leachate lagoon at this site have not been previously studied. Previous studies [5][7] have focused only on raw leachate obtained directly from the landfill area. The objective of this study was therefore to assess the toxicity of the leachates in the lagoon using a variety of organisms cutting across different groups: Aquatic insects which are important in the food chain since they are the principal diet for many species of fish [4]; Amphibians, also important as they have been used in leachate assessment studies [6] and the mudcatfish due to its economic importance. The specific objectives of the study were to determine the physico-chemical parameters of the leachate and the acute toxicity of the leachate to aquatic insects (represented by *Culex pipiens*, and *Chironomus sp.*), an amphibian (*Bufo regularis*) and fingerlings (7-week old) and post fingerlings (14-week old) of *Clarias gariepinus*. The toxicity assessment endpoints in all cases were lethal effects (toxicity) as well as sub-lethal effects (pupal emergence) in *Culex pipiens*. were also determined.

MATERIALS AND METHODS

2.1.1 STUDY AREA

The study area is the Akanran / Aba-Eku refuse landfill site located at Km 13, along Akanran – Ijebu Igbo road in Ona-Ara Local Government Area (LGA) of Oyo state, South-Western Nigeria. It is located approximately on longitudes $3^{o}5'$ E and latitudes $7^{o}23'$ N respectively [7]. Figure 1 is the map of Ona-Ara LGA showing the location of the landfill site at Aba-Eku. Leachates from the landfill are drained through a system of pipes into a central leachate lagoon. This is situated approximately 250m down gradient of the active fill area, where the domestic and industrial solid wastes are dumped. A GPS 76 garnier model was used to determine the distance from the landfill. Leachates from the leachate lagoon are discharged into the nearby Omi stream which flows past the Aba-Eku settlement.

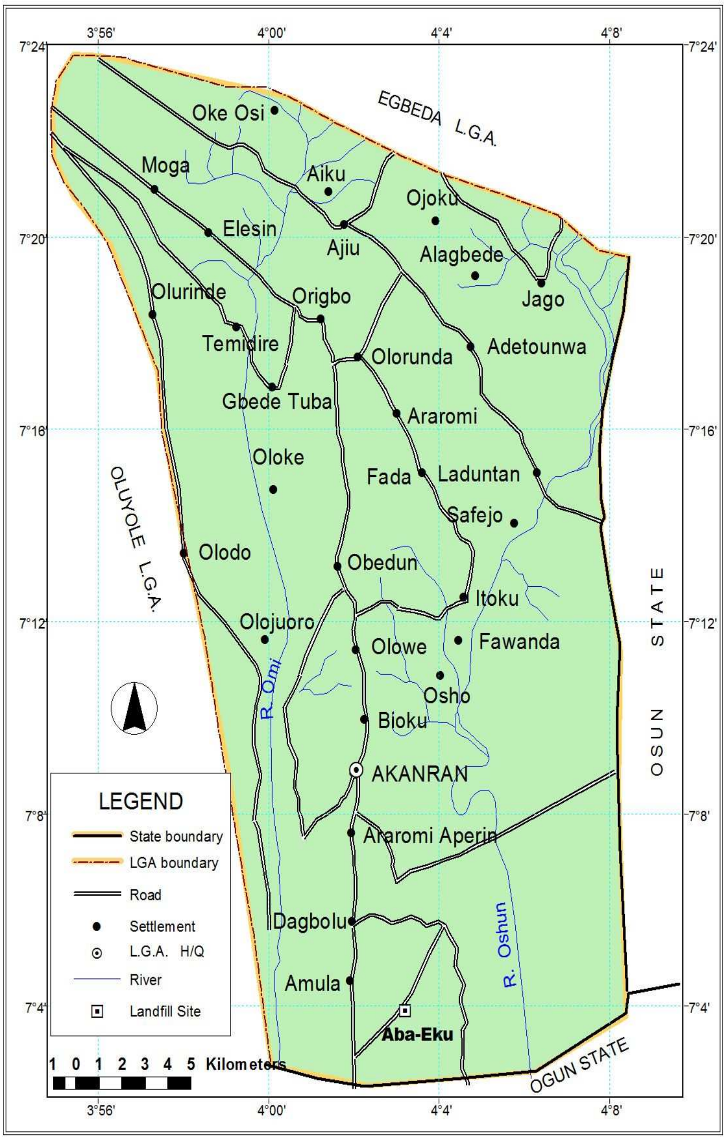

Fig. 1: Map of Ona-Ara LGA showing the Akanran / Aba-Eku landfill site.

2.1.2 Sampling, preservation and analytical methods for leachates.

Leachate samples were collected monthly from January 2003 to September 2004 in 1.5 litre plastic containers from the leachate lagoon (LL) shown in Figure 2. They were collected in pre-washed polyethylene bottles and taken to the laboratory, where they were stored at approximately 4^{o}C until when analysed.

Analytical parameters determined were:

- pH using a pH meter model PHS-3B;
- Total Dissolved Solids (TDS) and Electrical Conductivity (EC) using a WTW conductivity meter LF 95 model;
- The following were determined using the method stipulated by [8]:
  Total Suspended Solids (TSS); Dissolved organic matter expressed as
  Chemical Oxygen Demand (COD);
- The metals and cations were preserved as follows: 100 ml of sample was acidified with 1 ml concentrated nitric acid (HNO_3) for preservation prior to digestion and subsequent analysis [8].

Metals and cations determined include: Cadmium(Cd), Chromium (Cr), Lead (Pb), Nickel (Ni), Copper (Cu) and Zinc (Zn). Others include Calcium (Ca), Magnesium (Mg), Potassium (K), Iron (Fe) and Manganese (Mn).

- The above metals and cations were analysed after nitric – perchloric acid digestion [8] using an Inductively Coupled Plasma - Optical Emission Spectroscopy - ICP-OES Perkin Elmer Optima 3000.
- The following anions and ammonium - NH_4^+ were determined using an Ion Chromatograph IC 1010 model with a detection limit of < 0.005. They include: Sulphates (SO_4^{2-}), Chloride (Cl^-), Nitrates (NO_3^-) and Phosphates – (PO_4^{3-}).
- Biological Oxygen Demand (BOD) of the samples was not determined. This is because of the inhibition of the activity of micro-organisms in a leachate sample, by heavy metals and organics present in the sample. These make the BOD give suppressed results [9]. Hence the Chemical Oxygen Demand (COD) was thus used as a measure of the organics present in the leachate sample. All analytical procedures for leachate physico-chemical parameters were carried out at the Shenyang Institute of Applied Ecology, Shenyang, China. Results are presented as Means ± Standard Deviation (SD).

Figure 2: Slide showing part of the Leachate Lagoon.

2.1.3 Toxicity assessments: Organism collection and laboratory maintenance

Fish - *Clarias gariepinus* (Burchel, 1822)

Clarias gariepinus were collected from the Oyo State Ministry of Agriculture & Natural Resources fish farm at Agodi, Ibadan, in 25 litre containers. In the laboratory, the fish were transferred to large water holding tanks of dimension (90 x 50 x 60) cm at a stocking density of 120 and 60 per tank for fingerlings and post fingerlings respectively. The average weight of each fish was about 0.5 g for the 7 week old fingerlings and 2.5 g for the 14 week old post-fingerlings. Aeration was provided by a TOTA-TA 310 air pump. The fish were fed twice daily using commercial fish feed. They were fed at 4% of body weight. The water was changed every 48 hours to prevent accumulation of waste products. Tap water, left to stand for 48 hours for dechlorination, was used to change the water. Fish were allowed to acclimatise in the laboratory for about one week before the bioassays commenced.

2.1.4 Aquatic dipteran larvae: *Chironomus sp.* and *Culex pipiens*

Chironomids.

Chironomids were collected using an Ekman grab from bottom sediments of the sewage contaminated Awba stream in the Zoological garden, University of Ibadan. The sediments along with contaminated water from the stream were collected in plastic containers for laboratory usage. A white muslin cloth was used to sieve out the organisms from the sediments between washings, after which they were transferred into plastic containers containing dechlorinated tap water. A thin layer of silt was placed at the bottom of the containers according to the suggestion of Reish and Oshida [4]. A few drops of evaporated milk were added to the dechlorinated tap water containing the chironomids [10]. Chironomids were acclimatised for 12 hours before the commencement of the toxicity tests. They were covered with a wire mesh to prevent the escape of emerging adults.

Mosquito larva (*Culex pipiens)*

Culex pipiens was collected from pools in abandoned tyres in a mechanic workshop at Ojoo, Ibadan. They were collected in plastic containers and taken to the laboratory where they were then transferred into plastic containers and acclimatised in water from their natural environment for 12 hours before the start of the toxicity tests.

2.1.5 Amphibians: Tadpoles of *Bufo regularis*

Tadpoles of *Bufo regularis* were collected from Awba stream along the Appleton Road, University of Ibadan. They were collected in a plastic container and taken to the laboratory, where they were maintained in holding tanks and acclimated for two days. They were fed with commercial fish feed.

2.1.6 Bioassays

Range finding and acute toxicity definitive tests: Freshly collected leachates from the leachate lagoon at the "Aba-eku" landfill site, Ibadan, were used for the bioassays. 10, 25, 50, 75 and 100% dilutions of the leachates were prepared for the concentration of leachates for acute toxicity tests. A control with only dechlorinated tap water (0 % dilution) was also included.

Fish: Eight fishes were used per tank containing 300ml of test solution (for fingerlings) while 500ml was used for post fingerlings. Mortality was assessed at 24, 48, 72 and 96 hour intervals after the commencement of the test. The fish were taken as dead, if they did not show any signs of movement on examination and gentle prodding. Each concentration was replicated twice, with eight fishes in each replicate. Feeding was discontinued 24 hours before the start of both range finding and the acute toxicity tests [4]. Fish were not fed throughout the duration of the test, which lasted 96 hours, with mortality assessed at time intervals of 24, 48, 72 and 96 hours after the commencement of the tests. The pH, temperature and dissolved oxygen content of the test concentrations were determined before and after the tests as above. Mortality was assessed at 24, 48, 72 and 96 hours after the start of the tests.

Aquatic insect larva and amphibians: Each concentration was replicated twice and twenty organisms were used in each replicate consisting of 300ml of test sample [4]. Care was taken to select organisms in the same inter-molt stage. This was achieved by using larvae of about similar size in both range-finding and definitive tests. Larvae were considered dead if they showed no movement on gentle prodding.

Statistical analysis of the obtained results
Probit analysis was used to determine the Lethal Concentration – LC_{50} for the leachate.

RESULTS AND DISCUSSION

Physico-chemical parameters:
Leachate from the leachate lagoon was slightly straw colored. The physico-chemical parameters of the leachates from the lagoon are as shown in Tables 1 and 2; results shown represent the mean of eighteen monthly samples. The results showed that all parameters except for suspended solids and chloride (which was approximately at the limit) were within range of the Nigerian Federal Ministry of Environment Standards [11][12]. The reduced level of several parameters may be indicative of some pre-treatment of the leachate. On the other hand, it may also indicate the use of low cost natural attenuation processes to reduce the concentration and amount of contaminants / pollutants at the site. Such processes may be physical (e.g dilution, sorption, precipitation) or biological processes (bio-degradation) resulting in the reduction in toxicity, mass and/or mobility of a contaminant [1][13].

In order to confirm the action of attenuation processes, raw leachate samples were obtained for comparison (Tables 1 & 2). The values obtained were much higher and similar to those obtained by Aluko *et al,* [5] in their studies on the characterization of raw leachates of the landfill site. The considerable reduction in contaminant levels demonstrates the action of the various attenuation processes in reducing contaminant mass and mobility in leachates draining into the leachate lagoon down-gradient of the active fill area. According to Christensen *et al*, [1], natural attenuation can reduce the concentration of pollutants down-gradient. It is also among leachate treatment methods, one of the possible strategies for pollutant reduction in leachate plumes [1][14][15]. However the effectiveness of natural attenuation depends on site and leachate characteristics, which require investigation before this can be implemented [16]. The above results show the impacts of natural attenuation in reducing contaminant levels.

Bioassay: pH, temperature and dissolved oxygen parameters for the leachate before and after the 96 hr bioassay for *C. gariepinus* are also presented. The results showed that temperatures fell and remained relatively constant after the 96hr exposure of *C. gariepinus* to the leachate, while pH increased and dissolved oxygen levels fluctuated widely in the treatments after the 96 hour exposure (Figs. 3-5).

Toxicity bioassays: Mortality was observed in seven week old fingerlings of *C. gariepinus* exposed to the leachate (Tables 3 and 4), while no mortality was observed in 14 week old fingerlings of *C. gariepinus* exposed to the leachate. In the seven week old fingerlings, 12.5 and 6.25% mortality were observed at only 10 and 50% leachate dilution respectively. The low toxicity observed prompted a repeat of the bioassays with the leachates filtered to remove fine suspended particles. A higher mortality was observed in the seven week old fingerlings ranged from 18.75% to 37.5%. Mortality was also observed in the control; however this was below 10%. The experiments with the filtered and unfiltered leachates show the role of suspended matter and sediments in reducing the toxicity of the leachate. This is probably due to the fact that metal sorption by the sediments present is expected to decrease the bio-available fraction, which probably explains the observed effects [17][18].

Table 1: Cations and metals in the leachate over the study period

	Ca	Mg	K	Fe	Mn	Cu	Zn	Pb	Cd	Ni	Cr
MEAN LCHT	86.40	83.85	213.21	5.27	2.31	0.014	0.129	0.035	0.157	0.021	0.013
SD	41.81	41.09	107.00	8.87	1.60	0.028	0.094	0.019	0.128	0.022	0.009
FMEnv STD	200	200	-	20	5	< 1	< 1	< 1	< 1	< 1	< 1
ABA-EKU RL	117	207	3080	31.29	1.91	1.06	1.33	0.35	0.38	0.11	0.11

Unit: mg/l; LCHT (Leachate Lagoon); SD (Standard Deviation); RL (Raw Leachate)

Selected parameters in leachates before and after 96hr exposure of *Clarias gariepinus*

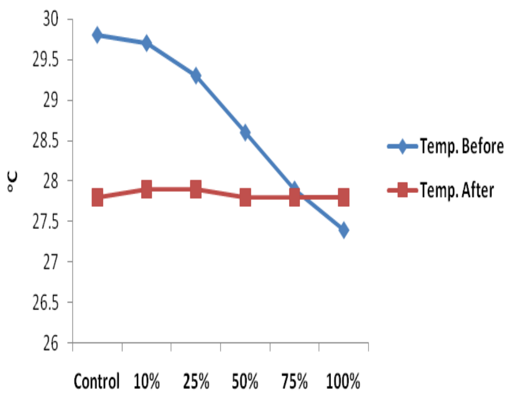

Fig. 3: Variation in Temperature before and after 96hr exposure

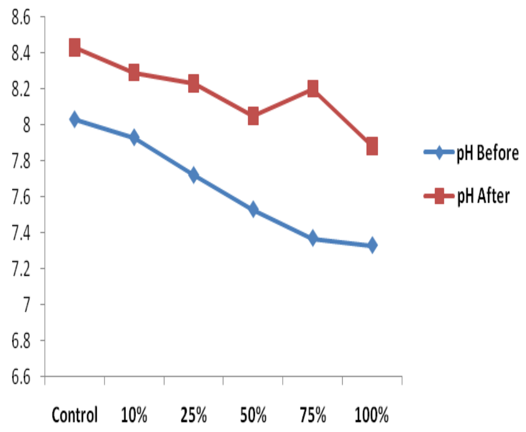

Fig. 4: Variation in pH before and after 96hr exposure

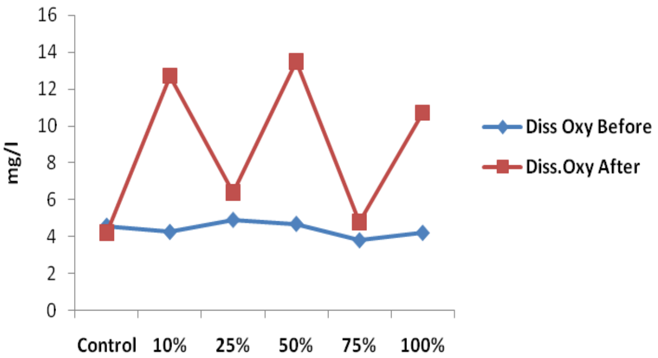

Fig. 5: Variation in dissolved oxygen before and after 96hr exposure

Table 2: General parameters, anions and ammonium in the leachate over the study period

	pH	TDS	TSS	TS	EC (µs/cm)	COD	Cl^-	NO_3^-	NH_4^+	SO_4^{2-}	PO_4^{3-}
MEAN LCHT	8.21	1233.13	144.94*	1377.67	2466.00	57.63	597.98**	12.37	3.55	118.86	ND
SD	0.11	336.56	117.99	396.85	673.02	15.11	209.73	24.79	1.64	77.65	ND
FMEnv STD	6-9	2000	**30**	-	-	-	**600**	20	-	500	5
ABA-EKU RL	8.65	4150	1890.74	6040.74	8300	1128.4	1250.27	1.52	22.78	486.32	ND

*Unit: mg/l except for pH and where otherwise stated; LCHT (Leachate Lagoon); SD (Standard Deviation); RL (Raw leachate) ND: Below detection limit (0.005<); *: Exceeds FMEnv Limit; **: Approaching FMEnv Limit*

No mortality was observed in *Bufo regularis* after the 96hr exposure to the leachates. Results of the 96 hour bioassays using the larvae of *Chironomus sp.* & *Culex pipiens* are given in Tables 5, 6 and 7. The results of bioassays using *Chironomus* larvae showed mortalities of 5, 10 and 5% for 25%, 50% and 75% leachate dilutions respectively. No mortality was recorded in the control and 100% leachate (Table 5). Results of the bioassays on fish (*Clarias gariepinus)*, the amphibian *(Bufo regularis)* and insects *(Chironomus sp.*) showed that the toxicity of the leachate had been reduced by the various attenuation processes acting on the leachate. Consequently, % concentration lethal to 50% of the organisms (LC_{50}) could not be computed as the leachates induced 0% (*Bufo regularis*; 14 wk old fingerlings of *Clarias gariepinus*) to less than 50% mortality in *Chironomus sp*. and 7 wk old fingerlings of *C. gariepinus*. Ammonia, a parameter that contributes to toxicity [19] was low in the leachate (Table 2) and this may have had a contributory effect to the low mortalities observed in the test organisms. The toxicity of leachate reliably depends on its ammonia content and leachate toxicity is considerably lower in landfills where the ammonia had been degraded [20]. Similar conclusions were made by Dave and Nilsson [21] after sub-lethal tests on the fish (Molnbyggen). The responses induced in *Chironomus sp.* and seven week old fingerlings of *C. gariepinus* by the leachate were not dose dependent. Although rare in toxicity studies, responses that are not dose dependent have been encountered in literature. In their experiments on the toxicity and dose dependent response of a vitamin D analogue to transgenic mice Dawson *et al*, [22] observed a low toxicity with no dose dependent curve evident.

Table 3: 96 hr acute toxicity bioassay using unfiltered leachate on 7 week old fingerlings of *Clarias gariepinus*

%	Mortality (%)				
Leachate	**24hrs**	**36hrs**	**48hrs**	**72hrs**	**96hrs**
0	0	0	0	0	0
10	0	0	0	0	12.50
25	0	0	0	0	0
50	0	0	0	0	6.25
75	0	0	0	0	0
100	0	0	0	0	0

Table 4: 96 hr acute toxicity bioassay using filtered leachate on 7 week old fingerlings of *Clarias gariepinus*

%	Mortality (%)				
Leachate	**24hrs**	**36hrs**	**48hrs**	**72hrs**	**96hrs**
0	0	0	0	6.25	6.25
10	6.25	6.25	6.25	6.25	31.25
25	0	0	0	0	37.50
50	0	0	0	6.25	18.75
75	0	0	0	6.25	31.25
100	0	0	0	6.25	18.75

Table 5: 96 hr acute toxicity bioassay using leachate on *Chironomus sp.*

%	Mortality (%)				
Leachate	**24hrs**	**36hrs**	**48hrs**	**72hrs**	**96hrs**
0	0	0	0	0	0
10	0	0	0	0	0
25	5	5	5	5	5
50	10	10	10	10	10
75	5	5	5	5	5
100	0	0	0	0	0

Table 6: 96 hr acute toxicity bioassay using leachate on larvae of the mosquito (*Culex pipiens)*

%	% Mortality			
Leachate	**24hrs**	**48hrs**	**72hrs**	**96hrs**
0	40	70	80	80
10	0	22	55	70
25	0	37.5	40	40
50	0	10	33	40
75	0	11	10	20
100	10	10	20	40

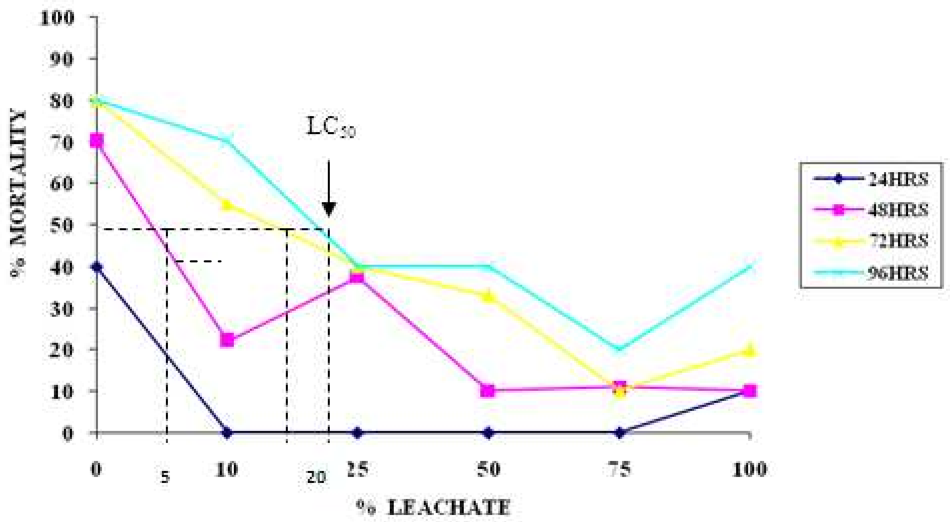

Fig 6: Probit graph for leachate exposure to larvae of *Culex pipiens*

Table 7: Effects of leachate exposure on pupal emergence in *Culex pipiens*

%			% Emergence		
Leachate	**24hrs**	**36hrs**	**48hrs**	**72hrs**	**96hrs**
0	100	100	100	100	100
10	90	100	100	100	100
25	100	100	100	100	100
50	100	100	100	100	100
75	100	100	100	100	100
100	80	100	100	100	100

The absence of a distinct dose-dependent curve and the induction of less than 50% mortality in almost all the above organisms may be suggestive of an unstable leachate with reduced toxicity probably due to the various attenuation processes acting on the leachate. In all cases, except in the bioassays involving *Culex pipiens*, mortality in controls were below 10%. 96hr LC_{50} in *Culex pipiens* was 20% leachate dilution (Figure 6) while LC_{50} values for all the other organisms were indeterminate. The results of bioassays using the larvae of *Culex pipiens* showed a preference of these organisms for the leachate. Lower percentage mortalities were observed at higher dilutions of the leachate, compared to the control (Table 6; Figure 6). This may not be unconnected with the preference of this particular species for a contaminated rather than an uncontaminated environment as toxicity reduced with increasing concentrations of the leachate. This was also supported by the results of pupal emergence which further indicate the preference of this species for a contaminated environment.

Larvae of *Culex pipiens* usually occurs in aquatic habitats directly or indirectly exposed to chemical contaminants [23]. Furthermore, in the studies by Tranchida *et al*, [24] on the biological control of *Culex pipiens* in natural habitats, all the immature stages of *C. pipiens* were found in drainage ditches with accumulated refuse, suggesting a preference by this organism for contaminated habitats, inclusive of those contaminated by landfill leachate. One hundred percentage emergence was observed in almost all dilutions and in the control over the duration of the experiment. The exceptions were the ninety and eighty percent reduction in emergence that was observed at 10% and 100% of the leachate after 24 hours respectively (Table 7).

CONCLUSION

Toxicity bioassays on leachates of the Aba-Eku leachate lagoon suggest that natural attenuation processes may have been used to achieve some measure of leachate treatment. This is supported by the results of the physico-chemical analysis. However suspended solids and chloride were still high in the leachate and highlight the need for further purification of the leachate effluent.

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