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Research article

Assessing the effects of tropical wood leachate to *Desmodesmus subspicatus, Lemna minor* and *Daphnia magna*

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ABSTRACT

Ghana has a long history as a major supplier of high-value hardwood timber and wood products to many countries. The research seeks to assess the effects of tropical wood leachates to aquatic organisms. Hence, five wood samples were selected; Mahogany (*Khaya ivorensis*), *Cedrela (Cedrela odorata)*, Emire *(Terminalia ivorensis)*, Wawa (*Triplochiton scleroxylon*) and Ceiba (*Ceiba pendandra*) from Oboyow forest reserve in Eastern Region-Ghana to assess their toxicity to aquatic organisms. Toxicity tests: Algal (*Desmodesmus subspicatus*) Duckweed (*Lemna minor*) and crustacean (*Daphnia magna*) were carried out using exposures to concentrations of 20, 30, 45, 67 and 100% v/v wood leachate in control media. The high levels of phenols measured in the various wood leachates was the main cause of toxicity. The percentage median Inhibition Concentration (%IC_{50}) of the various wood leachate, ranged from 21.5 - 55.6% with mahogany exhibiting the highest toxicity and wawa the lowest. All the wood leachates were toxic to the aquatic organisms. The %IC_{50} showed both confirmed and potential toxicity among the various wood leachates and established that there was significant difference between various wood leachate toxicity.

1. Introduction

Wood has been useful to human societies for thousands of years and used across a wide range of human activities. Wood is the hard fibrous material that forms the main substance of the trunk or branches of a tree or shrub. Tropical woods are wood obtained from trees that grow in tropical forests and in total, the tropical forests cover about 1700 million hectares of the world land size, an area roughly that of South America Food (FAO-UN, 1993). Ghana has a land surface of 22.8 million hectares and forested land around 9.3 million hectares and this constitutes 41.0% of the total land area (FAO, 2015). According to Ministry of Lands and Natural Resources (MLNR, 2012), Ghana forest is divided into two vegetation: the High Forest Zone in the south covering 34% and the Savannah Zone in the north covering 66% of the land area. The 2.6 million hectares of forest reserve land, 1.6 million hectares fall within High Forest Zone and of these reserves, 715,000 ha dedicated to natural timber production (Ghana – EU, 2012). International Tropical Timber Organization (ITTO, 2015) reported that Ghana produced in 2015 about 2.6 million m^3 of round wood and the exports of primary timber products accounted for a total export value of 230 million US dollars.

In view of adding value to the timber, the country aims to make more efficient use of the wood and to produce more high-end products such as shaped and machined mouldings, flooring, and furniture components. This has resulted in a spring up of a large number of woodpiles, log yards and wood storage areas within the forest catchment areas. Most of the forest area is in the southwestern part of the country, which has high rainfall patterns annually of about 2,100 mm (Logah et al., 2013). This is where most of these sawmills located. During the process of value addition, the woods must be stored and processed. When the process is not managed well and the wood or wood waste gets into contact with water leachate is generated. Leachate is produced when liquid percolates through solid material (Cheremisinoff, 1997; Jayawardhana et al., 2016). During this process, compounds leached from the solid material and when toxic affect aquatic organisms when released into water bodies. However, wood contains organic compounds that are toxic and leached in contact with water. In particular, a negative impact shown on water bodies that runoff from industries that process wood-based materials (Svensson et al., 2013). The compounds leached out during this process are the extractives and these are compounds extracted from wood using water, solvents or other extraction methods. They include waxes, fatty

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acids, resin acids, and terpenes and classified as phenolic, aliphatic, alicyclic or other lesser compounds (Sjöström and Alén, 1998). The overall composition of the extracts varies from tree species to tree species. Various studies have shown that wood leachate can influence quality of the receiving water and be toxic to the aquatic life within it. Taylor and Carmichael (2003) showed that untreated wood leachate from fresh aspen was toxic to rainbow trout, daphnia, and luminescent bacteria. Libralato et al. (2007) assessed toxicity of untreated wood leachates towards two salt water organisms (*Crassostrea gigas and Artemia franciscana*), showing that the leachate from all types was toxic to the aquatic organisms tested. Consequently, the potential toxicity of wood leachate has become an issue of concern.

In this experiment, algae and duckweed will be used as plant bioindicators because they have been used for treatment of wastewater and genetic or biochemical research and has proven to be reliable (Jaafari and Yaghmaeian, 2019; Cantó-Pastor et al., 2015). *Daphnia* a crustacean has also been used for ecotoxicological evaluation of conventional and emerging contaminants and also genetical or biochemical research works.

Although research has established the negative impact of wood leachate on the aquatic environment, most of such research conducted the leachate obtained from wood waste piles, wood storage areas or wood disposal sites from America and Europe. Not much research carried out on tropical wood. Libralato et al. (2007) conducted research on the effect of toxicity of tropical wood to two saltwater organisms, which proved toxic, but no typical research is done on tropical wood from African. Hence, this research seeks to assess the of toxicity of tropical wood leachates to *Desmodesmus subspicatus, Lemna minor and Daphnia magna.*

2. Material and methods

2.1. Sample selection

Five fresh species of wood logs were obtained from a forest reserve in Eastern Region in Ghana: Mahogany (*Khaya ivorensis*), *Cedrela (Cedrela odorata)*, Emire *(Terminalia ivorensis),* Wawa (*Triplochiton scleroxylon*) and Ceiba (*Ceiba pendandra*). The logs were selected randomly in a the section of the forest designated for research work of the forest commission with assistance from a research officer. These woods selected for the research are the commonly harvested wood that is used in diverse human activities worldwide.

2.2. Sample preparation

The wood logs were cut into pieces with volume ranging from 2 - 3 cm^3. The dry mass of 100 g of wood samples were determined at 105 ± 5 °C according to International Organisation for Standardization ISO 11465 (ISO, 1993) using Memmert hot air oven, model UNE 300 (Germany). The air dried wood was transferred into a glass bottle containing 1000 mL of distilled water and capped. The content was placed in an agitation device at 5–10 rpm for 24 ± 0.5 h at a temperature within the range of 15–20 °C. The suspended solid was made to settle for 5–15 min and the mixture centrifuged at 4500 rpm for 10 min to get the solid portion of the mixture settled for onwards filtration using 5 μm filter paper. The filter paper was folded to form a conical shape and then fitted into a funnel for the filtration with a receptacle to collect the filtrate. The liquid obtained after filtration was the wood leachate in which the toxicity test to the aquatic organisms were conducted. The pH, conductivity and dissolved oxygen of the leachates were determined using the WTW Multiline P4, 2.2. The Biochemical Oxygen Demand (BOD) was determined using the suppression of nitrification method (Standard Method, 2017). The Chemical Oxygen Demand (COD) was determined using potassium dichromate, modified semi-automated colorimetry method (EPA, 1993). Total Organic Carbon (TOC) was determined using the high-temperature combustion method (Standard Method, 2017), because it is suitable for samples with high levels of TOC that will require dilution. The total phenol was determined quantitatively using Folin Ciocalteu's phenol reagent with Gallic acid (Singleton and Rossi, 1965).

2.3. Toxicity test

2.3.1. Algal toxicity test

Algal medium, Bold's Basal Medium (BBM) for algae cultivation was constituted from distilled water by dissolved appropriate concentration of given salts to have pH 6.6 ± 0.2 according to Bold (1949) and International Organisation for Standardization (2012). The BBM was sterilized by using pressure cooker for 20 min at 121 °C.

A stock culture of the algae was prepared from strain number BRINKMANN 1953/SAG 86.81 from a Culture Collection of Autotrophic Organisms (CCALA), Institute of Botany of the AS CR, Třeboň, Czech Republic. The algae stock culture was prepared in a flow chamber by adding 4 mL of stock algae concentrate to 150 mL of cultivation medium in flat bottom glass flasks. Incoming air was first cleaned from algae and bacteria free flying in round atmosphere using bacterial filter and immediately afterwards fed into algal suspension. The stock culture was transferred into a thermostat with light cycle (16/8 h) under stable florescent light and temperature (22 ± 2 °C; 6000–8000 lux) for 72 h under continual aeration.

The test was carried out in 25 mL Erlenmeyer flasks. Three replicates of 15 mL control medium and samples were prepared to a percentage leachate concentrations (20, 30, 45, 67 and 100% v/v). The experiment was carried under sterile conditions. Initial algal concentration 80,000 cells/mL was estimated by cell counting in the Bürker chamber. The control medium and samples were incubated for 72 h under stable light 6000–8000 lux and temperature 22 ± 2 °C. After the exposure period, the algae cells were counted in the Bürker chamber under a microscope to calculate the algae growth rate.

2.3.2. Duckweed aquatic plant toxicity test

Steinberg nutrient solution for duckweed cultivation was constituted from distilled water and dissolved appropriate concentrations of given salts with the constituted solution having pH 5.5 ± 0.2 according to ISO guideline 20079, ISO (2005). The nutrient medium was sterilized in pressure cooker for 20 min at temperature 121 °C.

Prior to the test, the duckweed were transferred from a solid agar into the Steinberg nutrient solution and cultivated under 24 ± 2 °C and light cycle (16 h/8 h; light/dark; 5000–6000 lux) for 168 h. The test was conducted in 150 mL beakers. A volume of 100 mL of samples and growth medium were prepared in three replicates of different concentrations of sample: 20, 35, 45, 67 and 100% v/v for the toxicity test. An initial frond number of ten was transferred into each replicate and the beakers were covered with transparent film. The samples were incubated for 168 h under the same temperature and light conditions as the culture. The plants were photographed at the beginning (0 day), middle (3 days) and end of the exposures (7 days) for frond number and area estimation. After the 7-day exposures, the chlorophyll content was extracted in 99.8% methanol (48 h; 4 °C, dark) and measured by spectrophotometry (Hach, DR/2400, Germany). The calculation of the total chlorophyll content was made according to Wellburn (1994). The frond number and area were calculated by image analysis according to NIS-Elements 4.2 (2004).

2.3.3. Daphnia toxicity test

The daphnia acute mobility inhibition assay was performed using juvenile individuals of *Daphnia magna* Straus aged up to 24 h, originating from ephippia (Microbiotests Inc., Mariakerke (Gent), Belgium). The test design was based on ISO guideline 6341 (ISO, 2012). Aerated ADaM medium (pH ~ 7.8 ± 0.2; $O_2 \geq 7.0$ mg/L) according to Klüttgen et al. (1994) was used as a control. To ensure sufficient amount of dissolved oxygen (minimum 90%) the ADaM medium was aerated for at least a day before used for toxicity test. Before the toxicity test, ephippia were

culture in ADaM medium for 3–4 days under 24 ± 2 °C and 16/8 h light cycle for hatching of ephippia.

The test was conducted in 25 mL beakers. A volume of 20 mL of samples and growth medium were prepared in three replicates of different concentrations of sample: 20, 35, 45, 67 and 100% v/v for the toxicity test. This was made in three replicates of both control and samples. 5 juveniles were transferred into the 25 mL beakers filled with various concentrations rate samples by Pasteur pipette, covered with transparent film and incubated at 24 ± 2 °C and a 16h/8h light/dark cycle (2000–3000 lux). The mobility (viability) of the test organisms was observed after the 48 h exposure.

2.4. Statistical analysis

Non-linear regression (logit model) was used to calculate the dose-response curve and the curves fitted by GraphPad prism ver. 5.01 (2009) to obtain the percentage median Inhibition Concentration (% IC_{50}). The difference in %IC_{50} among the various wood leachates samples toxicity was determined by using one-way analysis of variance (ANOVA, $P < 0.05$).

3. Results and discussion

3.1. Nature of wood leachates

The wood leachates had foams on the surface and showed different colours: Mahogany - dark brown, *Cedrela* – light pink, Wawa – Colourless, Emire – Yellowish and *Cedrela* – Brown. The different colours of the wood leachates could be due to the different chemical composition (Tao et al., 2005; Rex et al., 2016). The temperature generally was between 20 – 23 °C and all of them had a smell of lumber. The pH ranges between 4.7 and 11.2, the lowest was measured in emire and the highest in cedrela (Tables 1, 2, and 3). The leachate showed both acidity and alkalinity properties and generally all the leachates were in the weak acidity and alkalinity range which indicates that its impact on the aquatic organisms will not be impactful. Many research has reported of rather strong acid of wood leachates contributing to toxicity of aquatic organisms, but in this research work the results was different (Taylor et al., 1996; Tao et al., 2005). The hydrogen ion concentration reduces as the concentration ratio between the control medium to wood sample decreases except with cedrela which pH trend was inconsistent. In other words, when the wood samples volume to control medium increases the hydrogen ion concentration turns to increase. This could also be due to the acidic nature of the wood leachate (Garbowski, 2019). The pH after the exposure increased by 3–6% in all the various wood leachate samples and this may be due to gas exchanges.

The conductivity was generally high and falls between 194 – 915 μS/cm indicating the high presence of inorganic ions (Table 4). The higher wood leachate concentration had lower conductivity and generally, the conductivity decreases with increase in concentration. Most of the leachates have conductivity which was above the acceptable range for discharge into freshwater (APHA, 1992; Bhateria and Jain, 2016). The high conductivity measured in the lower leachate might be due to the ions released from the salts of the control medium and the samples. The cedrela had the highest conductivity, indicating high ions than the rest of the wood leachate samples. The variation in the conductivity observed in the various leachates could cause instability in water bodies when released into it. The Dissolved Oxygen (DO) measured among the leachates were above the acceptable limit of 6 mg/L with exception to Emire at the 100% concentration (Ferreira et al., 2008). It could also be observed that the DO of almost all the leachate samples under investigation were all within the optimal range for the survival of daphnia (Table 5). Even though the DO was within the acceptable limit, initial DO decrease after the 48 h exposure, which could be due to consumption by the daphnia and the high content of organic carbon in wood leachate.

3.2. BOD, COD and TOC analysis

The oxygen demand and total organic carbon were determined to know the level of organic pollutants within the various wood leachates. The BOD and COD for all the wood leachate sample were above the effluent discharge permissible limit of 40 mg/L and 120 mg/L respectively (Environmental Protection, 2003; Aniyikaiye et al., 2019). This indicates that there are more organic and inorganic compounds in the

Table 1. pH measured of series of dilutions of wood leachate with control medium to duckweed after exposure for 168 h and their mean and SD values (n = 3).

Concentration of leachate (%)	pH				
	Name of wood leachates				
	Mahogany	*Cedrela*	Wawa	Emire	Ceiba
Control	6.5 ± 0.1	6.5 ± 0.3	6.5 ± 0.3	6.5 ± 0.1	6.5 ± 0.1
20	6.9 ± 0.2	6.6 ± 0.3	7.5 ± 0.2	6.6 ± 0.2	9.0 ± 0.2
30	6.8 ± 0.1	6.9 ± 0.2	7.2 ± 0.1	6.4 ± 0.3	8.7 ± 0.3
45	6.8 ± 0.3	7.3 ± 0.1	7.2 ± 0.2	6.2 ± 0.1	8.6 ± 0.1
67	6.7 ± 0.1	7.2 ± 0.1	6.9 ± 0.3	6.1 ± 0.3	8.5 ± 0.1
100	5.9 ± 0.2	7.0 ± 0.2	6.7 ± 0.2	5.5 ± 0.1	8.4 ± 0.1

Table 2. pH measured of series of dilutions of wood leachate with control medium to algae after exposure for 72 h and the mean and SD values (n = 3).

Concentration of leachate (% v/v)	pH				
	Name of wood leachate				
	Mahogany	*Cedrela*	Wawa	Emire	Ceiba
Control	7.3 ± 0.2	7.3 ± 0.1	7.3 ± 0.2	7.3 ± 0.2	7.3 ± 0.2
20	6.6 ± 0.1	8.4 ± 0.2	9.0 ± 0.2	7.2 ± 0.1	11.2 ± 0.1
30	6.4 ± 0.2	9.5 ± 0.1	8.8 ± 0.1	7.1 ± 0.2	11.1 ± 0.1
45	6.4 ± 0.1	9.7 ± 0.2	8.7 ± 0.2	6.8 ± 0.1	10.2 ± 0.1
67	6.3 ± 0.2	8.6 ± 0.2	8.5 ± 0.2	6.7 ± 0.1	9.0 ± 0.1
100	5.0 ± 0.1	7.4 ± 0.1	8.3 ± 0.1	4.7 ± 0.2	8.9 ± 0.2

Table 3. pH measured of series of dilutions of wood leachate with control medium to *Daphnia* after exposure for 48 h and the mean and SD values (n = 3).

Concentration of leachate (% v/v)	pH				
	Name of wood leachate				
	Mahogany	*Cedrela*	Wawa	Emire	Ceiba
Control	7.8 ± 0.1	7.8 ± 0.1	7.7 ± 0.1	7.7 ± 0.1	7.7 ± 0.1
20	6.9 ± 0.2	7.4 ± 0.1	7.8 ± 0.2	7.2 ± 0.3	7.9 ± 0.2
30	6.8 ± 0.1	7.4 ± 0.2	7.7 ± 0.1	7.1 ± 0.2	8.1 ± 0.1
45	6.5 ± 0.2	7.1 ± 0.1	7.6 ± 0.1	6.8 ± 0.1	8.0 ± 0.3
67	6.5 ± 0.1	7.1 ± 0.1	7.6 ± 0.2	6.1 ± 0.1	7.9 ± 0.2
100	6.1 ± 0.1	6.9 ± 0.1	6.9 ± 0.1	5.1 ± 0.2	6.7 ± 0.1

Table 4. Conductivity measured of series of dilutions of wood leachate with control medium to duckweed after exposure for 168 h and the mean and SD values (n = 3).

Concentration of leachate (% v/v)	Conductivity (μS/cm)				
	Name of wood leachate				
	Mahogany	*Cedrela*	Wawa	Emire	Ceiba
Control	915 ± 2	915 ± 7	915 ± 2	915 ± 4	915 ± 2
20	432 ± 6	754 ± 10	912 ± 2	831 ± 7	897 ± 6
30	427 ± 1	652 ± 7	882 ± 8	786 ± 3	843 ± 6
45	352 ± 4	537 ± 8	856 ± 13	736 ± 3	842 ± 4
67	278 ± 2	384 ± 4	832 ± 2	612 ± 4	837 ± 1
100	194 ± 3	273 ± 2	784 ± 4	492 ± 9	814 ± 4

Table 5. DO measured of series of dilutions of wood leachate with control medium to daphnia after exposure for 48 h and the mean and SD values (n = 3).

Concentration of leachate (% v/v)	Dissolved Oxygen (mg/L)				
	Name of wood leachate				
	Mahogany	*Cedrela*	Wawa	Emire	Ceiba
Control	7.8 ± 0.1	7.8 ± 0.1	7.7 ± 0.1	7.7 ± 0.1	7.7 ± 0.1
20	6.9 ± 0.2	7.4 ± 0.1	7.8 ± 0.2	7.2 ± 0.3	7.9 ± 0.2
30	6.8 ± 0.1	7.4 ± 0.2	7.7 ± 0.1	7.1 ± 0.2	8.1 ± 0.1
45	6.5 ± 0.2	7.1 ± 0.1	7.6 ± 0.1	6.8 ± 0.1	8.0 ± 0.3
67	6.5 ± 0.1	7.1 ± 0.1	7.6 ± 0.2	6.1 ± 0.1	7.9 ± 0.2
100	6.1 ± 0.1	6.9 ± 0.1	6.9 ± 0.1	5.1 ± 0.2	6.7 ± 0.1

sample and hence will affect the quality of water and kill aquatic organisms when discharged without treatment.

The BOD measured ranges between 101-268 mg/L (Table 6), and averagely from the various wood leachate was more than 2 times the permissible limit. The COD from the values measured (Table 6) averagely was 3 times that of the permissible level. This shows that there were more chemicals within the samples that were not oxidized (Rex et al., 2016). The BOD and COD ratio of 0.3 indicates that it can be treated biologically, but must be seeded (Khaled and Gina, 2014). The TOC measured was also above the permissible limit indicating the presence of more organic carbon within the sample. This could promote high oxygen demand and hence affect lives in the aquatic environment (Kannepalli et al., 2016).

3.3. Determination of total phenol

The phenol is one of the major compound attributed for causing toxicity in wood leachate (Rex et al., 2016, Svensson et al., 2013; Taylor et al., 1996). The phenols determined in the various wood leachates were all above the permissible limit of 1 mg/L in waste water (Hussain et al., 2015). Mahogany was 70 times higher than the permissible limit, which indicates its level of toxicity to the various aquatic organisms (Table 7). The difference in the various concentrations of phenols in the wood leachates could be due to their nature and chemical composition because concentration of extractives varies from tree species to tree species (Nascimento et al., 2013). The various concentrations of phenols measured corresponded with the level of toxicity of the various wood

Table 6. BOD, COD and TOC values for the various wood leachate samples and also shown at the mean and SD values (n = 3).

Wood Leachate Samples	BOD (mg/L)	COD (mg/L)	TOC (mg/L)
Mahogany	268.2 ± 4.0	1397.1 ± 7.0	699.0 ± 5.2
Cedrela	121.7 ± 6.0	329.7 ± 4.0	167.2 ± 6.1
Wawa	31.0 ± 3.0	89.5 ± 8.0	112.5 ± 3.3
Emire	103.3 ± 6.0	505.3 ± 6.0	246.0 ± 4.3
Ceiba	101.1 ± 6.0	476.6 ± 2.5	200.0 ± 6.5

Table 7. Phenol concentration in wood samples using spectrophotometry method and values are mean ± SD (n = 3).

Wood Leachate Samples	Phenol Concentration (mg/L)
Mahogany	76.7 ± 7.0
Cedrela	7.1 ± 0.2
Wawa	8.7 ± 0.7
Emire	40.1 ± 3.0
Cedrela	7.1 ± 0.2

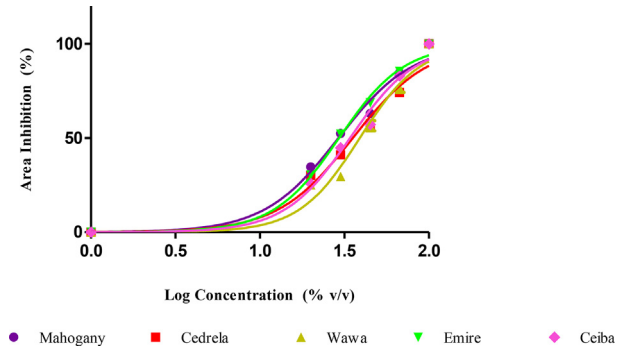

Figure 1. Percentage median Inhibition Concentration (%IC_{50}) of area of fronds after 7 days of exposure of duckweed to various dilutions of wood leachates.

leachates (Taylor et al., 1996). Also, the presence of phenols affect the development, growth and survival of aquatic organisms.

3.4. Ecotoxicity assay

The toxicity of wood leachate to duckweed was determined by observing the reproduction, area and chlorophyll content of the fronds (ISO, 2005). The duckweed on the 3rd day of exposure showed increased in frond numbers in the lower sample concentrations with wawa, *cedrela* and ceiba doubling in frond numbers with an initial number of 10 fronds (Wawa-23, *Cedrela*-21, Ceiba-22, Emire-18, and Mahogany-17). However, at higher concentrations of leachate, there was less reproduction of fronds especially in emire and mahogany (emire-14 and mahogany-13) this could be due to a lack of nutrients to support growth (Radić et al., 2011; Rajaram, 2016). There was growth in the frond areas in all the concentrations, but the high area growth rate was observed in the lower concentrations. After the third day of exposure, chlorosis was seen in the 67 and 100% v/v of the emire and the mahogany leachate (Ziegler et al., 2016). However, after the 7th day of exposure, there were great changes in the duckweed morphology in the various wood leachate. Some of the fronds showed both chlorosis and necrosis, others had black spots on the fronds and some with elongated roots. This occurred because of the presence of phenols and also lack of nutrients in some of the set up depending on the medium available (Tao et al., 2005).

The fronds numbers in wawa quadrupled, *cedrela* and ceiba tripled in the 20 and 30% v/v this was observed because at these concentrations more nutrients are available to duckweed which enable their reproduction. But not so much reproduction of frond numbers observed in all the wood samples at concentrations 67 and 100% v/v and also, there was no growth in the frond area (Figure 1). This could be due to a lack of nutrients to support their growth. The colour of some of the leachate could be the effective of retardation in reproduction, because they prevented

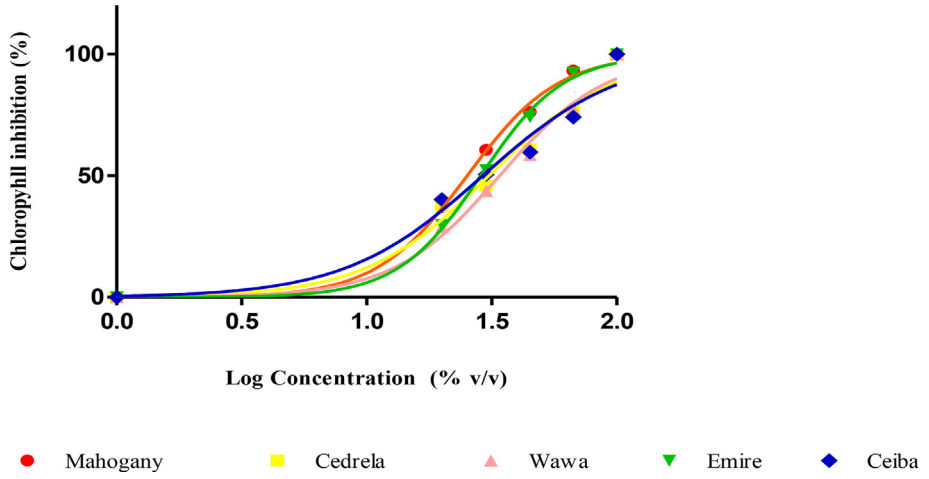

Figure 2. Percentage median Inhibition Concentration (%IC_{50}) of chlorophyll content of fronds after 7 days of exposure of duckweed to various dilutions of wood leachates.

Table 8. %IC_{50} of the Duckweed (% Area Inhibition and % Chlorophyll Inhibition), algae and *Daphnia* inhibition of the various wood leachates. Also, shown are the 95% confidence interval and the coefficient of regression.

Types of wood	Duckweed Toxicity					
	% Area Inhibition			% Chlorophyll Inhibition		
	IC_{50}	95% CI	R^2	IC_{50}	95% CI	R^2
Mahogany	29.2	27.0–31.5	0.97	25.2	24.3–26.2	0.99
Cedrela	36.1	32.9–39.7	0.96	30.8	27.8–34.2	0.96
Wawa	34.0	31.6–36.7	0.97	34.0	31.6–36.7	0.97
Emire	29.4	28.1–30.7	0.99	28.6	27.7–29.4	0.99
Ceiba	34.1	31.8–36.6	0.97	29.1	25.6–33.1	0.95
	% Algae Inhibition			% *Daphnia* immobilization		
	IC_{50}	95% CI	R^2	IC_{50}	95% CI	R^2
Mahogany	27.7	25.9–29.7	0.98	21.5	18.5–24.9	0.94
Cedrela	40.5	37.7–43.6	0.97	35.3	31.3–39.9	0.92
Wawa	41.5	37.8–45.6	0.95	55.6	53.3–58.1	0.98
Emire	30.6	28.6–32.8	0.98	29.8	26.7–33.3	0.96
Ceiba	38.6	37.1–40.3	0.99	42.4	37.7–47.6	0.91

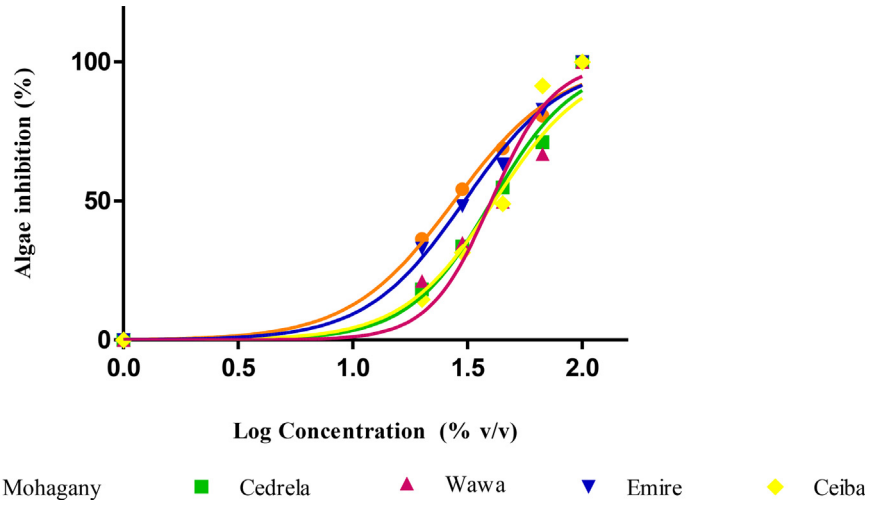

Figure 3. Percentage median Inhibition Concentration (%IC_{50}) of algae growth rate after 3 days of exposure to various dilutions of wood leachates.

transmission of light which is necessary to the growth of plants (Ziegler et al., 2016). The chlorophyll content of the fronds decreased with an increase in the concentration of the various wood leachate (Figure 2), which corresponds to the growth rate of the duckweed in the various wood leachate concentrations. Hence, more chlorophyll content was measured in the lower concentrations because there were increase in fronds and size of area. The %IC_{50} of duckweed toxicity (Table 8) for both the area and chlorophyll ranges between 24.4%–36.0% indicating confirmed (Pooja et al., 2017). This proves that averagely more than 70% of the duckweed used for the toxicity test was negatively affected by the leachates. Furthermore, the leachate has the ability to cause toxicity to duckweed when release into aquatic body without treatment. The high levels of BOD, COD and TOC measured in the various wood leachates indicating high levels of organic and inorganic compounds could also contribute to the retardation in the growth of the duckweed (Tao et al., 2005). The mahogany showed the highest IC_{50} of 24.3 and 29.2% in chlorophyll and area among all the wood leachates and this could be due to its chemical composition (Garbowski, 2019; Svensson et al., 2013).

The growth rate of algae was observed in the lower concentration among all the wood leachate (Figure 3). In addition, the wawa and *cedrela* leachate had the same growth rate to the control, and this could be due to available nutrients such as phosphorus and nitrogen (Taylor and Carmichael, 2003). The high growth rate of algae experienced in the lower concentrations of wawa and *cederela* could be due to the transparency of the leachate, organic content and proportion of mixture of the wood leachate to the control medium (O'Hare et al., 2018; Garbowski, 2019; Svensson et al., 2013). According to (CEES, 2019) water chemistry, nutrient, mixing conditions, turbidity can greatly influence algal growth. However, the lower growth rate recorded in the higher concentrations (45, 67 and 100% v/v) especially in the emire and mahogany, could be due the colour of the landfill leachate, because it could prevent penetration of light to promote algal growth, which depicted a very lower algal growth rate at high concentrations of these wood samples (Singh and Singh, 2015). The reduction in the volume of control medium to sample would have been a contributing factor to the low growth rate of algal in the higher concentrations, because of lack of nutrients (McLachlan, 2011; Laohaprapanon et al., 2012). The high content of both organic and inorganic compounds in the leachates affected the growth rate of the algae and this seen in the higher concentrations. The presences of high phenols concentration greatly affected the growth of algae in the various wood leachates because it affects chlorophyll formation and also induced particular structure alterations which could include vanishing, or reduction, of nucleolus and swelling of vacuoles, which may end up in cell death (Duan et al., 2017; Taylor and Carmichael, 2003). High % IC_{50} of algae observed in mahogany and Emire 27.7 and 30.6% respectively (Table 8), because of their chemical constituent, leachate colour and organic matter. The high % IC_{50} obtained after the exposure indicated that the wood leachates toxicity to algae is on a higher side and when released without necessary assessment and possible treatment it could cause imbalance in the aquatic system. Generally, the various wood leachates exhibited confirmed and potential toxicity to all the bio-indicators used for the test (Pooja et al., 2017).

Immobilization of daphnia was observed in mahogany and emire at the 100% v/v after 24-hour observation, but generally, more immobilization of daphnia realised after 48-hours. Death of all the daphnia was recorded in 100% v/v in almost all the wood samples except wawa, which had about 40% of daphnia surviving (Figure 4). The response of daphnia to the wood leachate was high to the extent that at 67% v/v death was observed in mahogany, ceiba and *cedrela*. The high death could be due to the presence of organic and inorganic compounds because the dissolved oxygen measured after the experiments in the various leachates were above 6 mg/L (Hingston et al., 2001; Weis and Weis, 2002). The pH after exposure was 5.1–8.1 (Table 3) and the lowest pH measured in emire and mahogany, which was below the optimum pH for survival and growth of daphnia. This might have contributed to the immobilization of daphnia in these leachates (Mahassen et al., 2011). However, some of the daphnia survived at lower concentrations in wawa, cedrela, and ceiba leachate and this could be due to optimal conditions within these samples that promoted survival and growth. The high phenols concentration within the wood leachates could might be the main cause of toxicity as reported in other research, because it caused biotoxicity that is destroying cells and tissues of organisms (Bandow et al., 2018; Taylor and Carmichael, 2003). The %IC_{50} was very high in

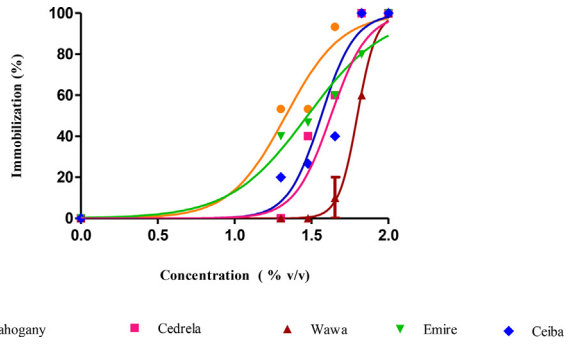

Figure 4. Percentage median Inhibition Concentration (%IC_{50}) of *Daphnia* after 24 h of exposure to various dilutions of wood leachates.

mahogany 21.5% and lowest in wawa 55.6%, indicating both confirmed and potential toxicity of the leachate to daphnia (Pooja et al., 2017).

One-way analysis of variance (ANOVA- $P < 0.05$) was used to mutually assess the difference between the toxicity among the various wood leachate. It was established that there is no significant difference ($P < 0.01$) between the toxicity of the various wood leachate.

4. Conclusion

The tropical wood leachates exhibited various levels of toxicity to the test organisms under study. The high BOD, COD and TOC could be a contributing factor, but the main cause of toxicity in the various wood leachates may be the high levels of Phenols. The mahogany leachate was the most toxic and Wawa the least toxic among all the various wood leachates studied. The effect of wood leachate to the test organisms followed the same trend of toxicity level: Mahogany > Emire > Ceiba > *Cedrela* > Wawa. The toxicity ranged between 21.1 and 55.6%, which indicated both conformed and potential toxicity to aquatic organisms. The highest $\%IC_{50}$ was obtained in daphnia, 21.5 % that indicated that it is more sensitive to the wood leachates than the other aquatic organisms. This study has confirmed that tropical wood leachate is toxic to aquatic organisms. The result obtained will help African countries like Ghana, which has decided to add value to its timber before exportation to know the need to treat leachate before disposal. The construction industries will also be aware of which wood to use for a particular project to prevent contamination of water bodies. It can be used by policy makers in environmental management policy and also will help environmental engineers in design of wood leachate treatment facility. The information obtained will help other researcher in their toxicity test, for further study on tropical wood leachates.

Declarations

Author contribution statement

Lyndon N.A Sackey: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

Vladimir Kočí: Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data.

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Competing interest statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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