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Spatial analysis of heavy metals in septic tank sewage

Hector H. Oyem[1*] and Ifeanyi M. Oyem[2]

[1*]Department of Chemical Sciences, University of Delta, Agbor, Nigeria
[2]Department of Biological Sciences, University of Delta, Agbor, Nigeria

*Corresponding author's email:
hector.oyem@unidel.edu.ng

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Heavy metals get to humans through food, water, air, and industrial activities. They arrive at septic tanks through the excretory products of man and undergo anaerobic bioremediation. Effluent and sludge sewage in both chambers of the tank contains various amounts of trace metals. The research analyses the spatial distribution of heavy metals in the effluent and sludge components of the septic tank during bioremediation. Effluent and sludge samples were randomly collected from three study locations in southern Nigeria, and composite samples were made for each of these study locations. Nine trace metals were analysed using standard methods by the American Public Health Association (APHA) for water and wastewater examination. Fe, Mn, Zn, Cu, and Cr were the most abundant. Effluent heavy metals concentrations were higher in the inlet chamber. There were strong positive correlation (r = 0.90, 0.99, and 0.99) between the effluent and sludge samples. P-values obtained from the ANOVA at a 0.05 significance level confirmed the correlation values. Multivariate charts and regression analysis gave R^2-values in the range of 0.3045 – 0.3724 in the study with a mean value of 0.33. In conclusion, the effluent and sludge samples were strongly correlated and all the metal types analysed were also present in the sludge in similar proportions. The sludge contained significant levels of trace metals with dire implications for the environment.

Keywords: Spatial, sewage, heavy metals, correlation, regression analysis.

1. Introduction

Septic tanks are anaerobic bio-digesters where human sewage is treated by acid- and methane-forming microorganisms [1]. Remediation of heavy metals in the sewage of septic tank systems is pH-sensitive [1–3]. Reports indicate that bio-precipitation of trace metals onto cell surfaces through microbial mechanisms is favoured by acidic conditions [2,4–11]. Contemporary septic tank systems consist of two chambers: the raw and the semi-treated (outlet) chambers; also known as the inlet and outlet chambers. The components of the septic tank include effluent, suspended solids, and sludge.

Sludge is the semi-solid component of sewage which contains about 57.0 % water [2]. The precipitation of solvated and insoluble chemicals leads to the production of sludge [12–15]. Contents of sludge include proteins, carbohydrates, pathogens and heavy metals [13].

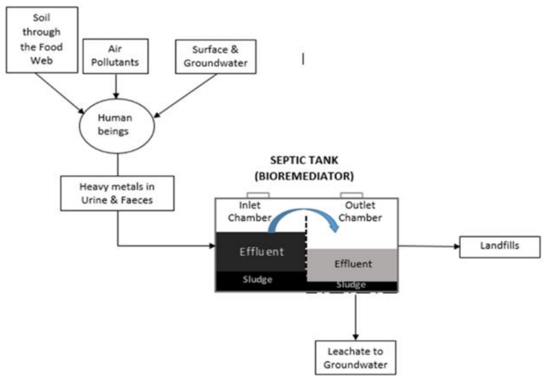


Figure 1. Schematic material balance diagram representing the pathways by which heavy metals get into man and enter the septic tank system. It also shows the layout of the septic tank bioremediation system and the partitioning of the sewage into effluent and sludge in the two chambers.

Heavy metals are elements with comparatively high densities often above 5 g/cm^3 [16–20]. Examples include arsenic, cadmium, chromium, lead, manganese, mercury, iron, and vanadium. These metals are present in the environment arising from natural and human-related activities [21,22]. They exist in all the components of the environment: air, surface and groundwater, and soil. From where they pass through plants and arrive at man through the food chain [15,23], including through air and water pollution. In this paper and for the purpose of clarity, we use the terms: heavy metals, trace metals, and dense metals interchangeably as one is synonymous with the other.

Again, man receives these metal pollutants through uptake by plants from the soil, irrigation water and airborne pollutants [24,25]. Thereby, effectively becoming a bio-accumulator of these pollutants from his environment. The body attempts to remove these pollutants by excretory processes; eventually arriving at the septic tank where they undergo bio-remediation (Figure 1).

Before now, studies relating to septic tank systems are usually and understandably focused on methanogenesis with an emphasis on energy generation from biogas.[26–30]. Others have focussed on characterizing heavy metals in water and wastewater effluent samples and comparing their concentrations with various guideline values.[22,31–34]. Until now, we have hardly found reports in the literature on studies of heavy metals in septic tank sewage systems where their concentrations and spatial distribution were studied.

In a recent study, we reported the role of *in situ* pH in the bioremediation process of heavy metals in effluents of septic tank systems, [35] and observed that the inherent pH of the system was not optimum to achieve effective bioremediation of heavy metals from the effluent. In this present study, which is a follow-up on the former, the principles of material balance is adopted to monitor the distribution of trace metals within the system, with particular emphasis on the effluent and sludge phases of the inlet chamber (raw sewage) of the septic tank to understand the spatial distribution of heavy metals in the sewage digester as a consequence of microbial remediation.

2. Materials and Methods

2.1 Sample area

Sewage samples for analysis were taken from different septic tanks in three locations of Edo and Delta States: Agbor (Location A), Benin (Location B) and Sapele (Location C) in the Niger Delta region of Nigeria (Figure. 2), these make up the study area. The soil type is characteristic of reddish-yellow ferrosols [36–39], for the Agbor and Benin sample areas except for Sapele which has the yellow-white soil of the Delta region being further south of the Niger-Delta region. In this study area, Benin City is the most metropolitan with a projected current population of 1.8m, [40] and a land area of 1,204 sq. kilometres, having a population density of 1495.0/sq. kilometres.[41] Agbor has a population of 162,594, [40] and a land area of 436 sq. kilometres. Sapele has a projected population of 240,000 spread across 580 sq. kilometres with a population density of 413.6/sq. kilometres [40,42].

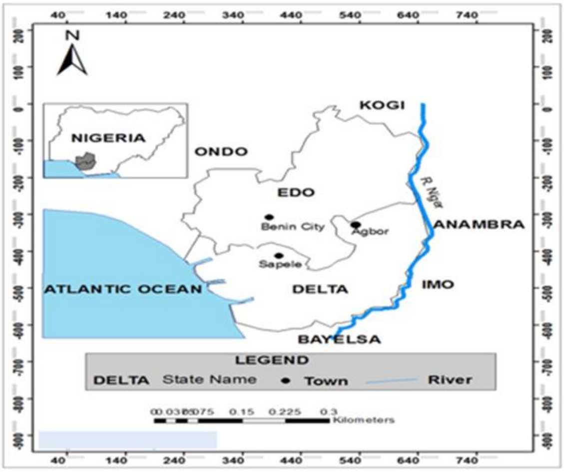


Figure 2. Map of the study area showing the three study locations A, B and C representing Agbor, Benin City and Sapele, Nigeria. (Source: Delta State Ministry of Lands and Survey, Asaba)

2.2 Sample collection

Sewage samples were collected in 1 L plastic bottles under aseptic conditions from septic tanks in the three sample locations of Agbor, Benin City, and Sapele in Edo and Delta States, Nigeria. Five sites were randomly sampled per location from each of the three sample locations. Samples were put in ice-packed coolers and then transported to the laboratory where a total of three composite samples were made for analyses according to the study locations.

These composite samples were labelled as samples A, B, and C respectively. Notably, all the septic tanks were essentially similar in design apart from the difference in the number of persons per household served per septic tank and the socio-cultural differences of the users. In all, a household is comprised of between 5 to 8 individuals. Each septic tank experienced a retention time of 4 – 6 hours daily when households would be away at school, work or sundry businesses.

2.3 pH determination

An aliquot of 1 mL of the raw chambers' separate effluent and sludge samples was obtained from each of the composite samples and put in a 10 mL sterilized beaker and then serially diluted to a final volume of 1000 mL with deionized water [43]. The pH of the samples was measured from the supernatant portion using a multi-parameter water quality portable meter, Hanna (HI 9828) by Keison Products, the United Kingdom, earlier standardized with distilled-deionized water.

2.4 Heavy metal analysis

Sewage samples were acidified with 1 mL concentrated solution of trioxonitrate (IV) acid per 100 mL of sample and then autoclaved for a period of 1 hr to solubilize the content. Heavy metal ions were analysed in triplicates together with a blank containing only distilled-deionized water. The methods were consistent with the Standard Methods for the Examination of Water and Wastewater [44], [45]. Sample heavy metal ions concentrations were obtained using a Solar Unicam atomic absorption spectrophotometer (AAS) model 969 AA manufactured by Vitech International BV, the Netherlands, previously calibrated. The mean values from three replicate readings of each heavy metal ion were obtained, from which their standard deviations were similarly calculated.

3. Results and Discussion

3.1 Hydrogen ion concentration (pH) of septic tank effluent and sludge samples

The hydrogen ion concentration of the effluent and sludge samples are presented in Table 1a and 1b. The results showed a pH range from 6.4 to 7.7.

Table 1a. The pH of septic tank effluent samples of the inlet (raw) and outlet (semi-treated) chambers in the three locations.

	Location					
	A		B		C	
Septic tank chamber	Inlet	Outlet	Inlet	Outlet	Inlet	Outlet
pH	7.7	7.3	6.9	7.3	6.5	6.7

(Oyem and Oyem, 2022)

Table 1b. pH of the septic tank sludge samples of the inlet (raw) and outlet (semi-treated) chambers in the locations.

	Location					
	A		B		C	
Septic tank chamber	Inlet	Outlet	Inlet	Outlet	Inlet	Outlet
pH	6.5	6.6	6.4	6.8	6.5	7.1

A comparison of the pH values of the effluent and sludge samples showed that the sludge component was slightly more acidic. This would favour the binding of heavy metal ions with microbes in the system [35]. The place of pH in the spatial distribution of heavy metals is significant in the septic tank system. In this paper, more focus is placed on the inlet chamber of the septic tank as we monitor the distribution dynamics of these heavy metals between the effluent and sludge components of this chamber of the septic tank where essentially sewage is held while microbes degrade organic matter. The idea is to study as well as understand the distribution of these metals between the effluent and sludge phases of the inlet chamber and use the balance in concentrations in the inlet and outlet chambers to evaluate the proportions accruing in the sludge.

Results in Table 1a showed that effluent pH conditions were basically above the neutral point (apart from location C) with averages of 7.5, 7.1, and 6.6 in locations A, B, and C in that order. In Table 1b results implied that the sludge samples tended towards the neutral pH mark of 7.0. Further analyses of the pH values of the sludge samples indicated that pH conditions varied randomly with the sludge component of the septic tank, even though acidity decreased in the following order: location A > location B > location C; with mean pH values of 6.5, 6.6, and 6.8 respectively.

3.2 Heavy metals in Effluent and Sludge samples of the inlet (raw) chamber sewage

The heavy metals profiles of effluent and sludge sewage samples were analysed for Cr, Cu, Fe, Mn, Ni, Pb, and V. The results are shown in Table 2 and 3.

Table 2. Effluent concentrations (mg/L) of heavy metals in both chambers of the sewage digesters in the study area.

S/No.	Metal ion (Analyte)	Septic Tank Effluent Samples					
		Location A		Location B		Location C	
		Inlet (mg/L)	Outlet (mg/L)	Inlet (mg/L)	Outlet (mg/L)	Inlet (mg/L)	Outlet (mg/L)
1.	Fe	2.03 ± 0.2	0.87 ±0.02	9.41 ± 0.01	7.52 ± 0.02	8.77 ± 0.02	6.04 ± 0.02
2.	Mn	0.42 ± 0.02	0.33 ± 0.02	0.14 ± 0.02	0.10 ± 0.10	0.034 ± 0.002	0.05 ±0.02
3.	Zn	0.50 ±0.1	0.28 ±0.04	0.37 ±0.02	0.18 ±0.02	0.15 ±0.02	0.47 ±0.02
4.	Cu	0.20 ±0.1	0.16 ±0.02	0.26 ±0.02	0.17 ±0.02	0.072 ±0.002	0.063 ±0.002
5.	Cr	0.64 ±0.02	0.51 ±0.01	0.08 ±0.02	0.047 ±0.002	0.045 ±0.002	0.019 ±0.001
6.	Cd	0.44 ±0.02	0.35 ±0.04	BDL	BDL	0.031 ±0.001	BDL
7.	Ni	BDL	BDL	0.056 ±0.002	0.031 ±0.002	0.027 ±0.002	0.011 ±0.001
8.	Pb	0.83 ±0.02	0.75 ±0.02	0.062 ±0.002	0.015 ±0.002	0.093 ±0.002	0.058 ±0.001
9.	V	BDL	BDL	0.044 ±0.002	0.029 ±0.002	0.02 ±0.01	0.006 ±0.005

*BDL = Below detection limit [35].

Table 3. Sludge concentrations (mg/kg) of heavy metals in both chambers of the sewage digesters in the study area.

S/No.	Metal ion (Analyte)	Septic Tank Sludge Samples					
		Location A		Location B		Location C	
		Inlet (mg/kg)	Outlet (mg/kg)	Inlet (mg/kg)	Outlet (mg/kg)	Inlet (mg/kg)	Outlet (mg/kg)
1.	Fe	1201 ±0.1	574.0 ±2.26	3391 ±1.13	1026±2	618.4±0.2	556.4±0.2
2.	Mn	30.4 ±0.2	12.1 ±0.1	17.3 ±0.23	15.0±2.0	38.7±0.2	33.4±0.2
3.	Zn	90.6 ±0.2	42.8 ±0.1	33.9 ±0.47	22.4±0.2	59.0±2.0	51.1±0.1
4.	Cu	10.4 ±0.4	4.78 ±0.01	9.45 ±0.02	12.8±0.1	13.5±0.2	17.2±0.1
5.	Cr	1.35 ±0.02	1.24 ±0.02	1.47 ±0.02	0.64±0.02	2.62±0.01	1.04±0.02
6.	Cd	BDL	BDL	1.10 ±0.10	0.48±0.02	3.44±0.02	2.80±0.10
7.	Ni	0.95 ±0.20	0.32 ±0.02	0.81 ±0.02	0.57±0.02	1.64±0.02	1.20±0.10
8.	Pb	0.77 ±0.02	0.58 ±0.02	1.62 ±0.02	1.26±0.02	2.67±0.63	2.10±0.10
9.	V	0.86 ±0.02	0.24 ±0.02	0.77±0.02	0.52±0.02	1.52±0.02	1.14±0.02

*BDL = Below detection limit

Iron

Table 2 shows that iron (Fe) had the highest relative abundance of all the heavy metals analysed. Its average concentration value is 6.74 mg/L. Iron concentration ranged from (2.03 ± 0.20 – 9.41 ± 0.01 mg/L) (Table 2). Fe was highest in location B at 7.52 – 9.41 mg/L for the raw effluent chamber. Between the inlet and outlet chambers of the septic tank, the variations in the concentrations of heavy metal analytes indicate the extent (efficiency) of the remediation process in the system. The magnitude of this difference accounts for the amount of the metals which have been precipitated out and deposited as sludge.

Heavy metal ion contents of the sludge samples in both the inlet and outlet chambers are displayed in Table 3 showing the concentrations of the individual analytes expressed in mg/kg. In location B, which had the highest Fe concentration in the effluent, 1.89 mg/L was the difference recorded, while 2.73, and 1.16 mg/L were the values for locations C and A respectively.

Iron was the most abundant metal in the effluent and sludge component of all the dense metals studied in the three locations. It accounted for about 90.0, 98.1, and 83.4 per cent of the inlet chamber sludge heavy metals contents in the study. Fe was highest in location B and least in location C. This spatial Fe distribution tallies with the geogenic description of the sample areas given in the previous section (Section 1.2.1 above). That is, both Agbor and Benin (locations A and B) were comprised of ferrite soil types [36]; whereas, the soil in location C as has already been mentioned was yellow-white characteristic of delta regions. It is not surprising that location C recorded the least Fe composition in the study.

Zinc

Zinc values were between 0.15 – 0.50 mg/L in the raw chamber effluent with an average of 0.34 mg/L. It was between 0.18 – 0.47 mg/L in the outlet chamber, averaging 0.31 mg/L. The balance in the concentrations of Zn in both chambers is again deposited as sludge.

Zinc is next in the magnitude of its concentration in the sludge component of the study area after iron. It was highest in the inlet chamber sludge sample in location A, posting a concentration of 90.60 mg/kg, and the least value of 33.9 mg/kg in location B's inlet chamber. These results are in line with reports by several researchers who noted that sludge was a rich deposit of toxic metals [46–48].

Manganese

Manganese values in the effluent of the raw chamber for all the study locations are 0.42, 0.14, and 0.03 mg/L respectively in Table 2. It was highest in location A with a value of 0.42 mg/L. The mean value of manganese calculated for the raw chamber was 0.20 mg/L, and 0.16 mg/L in the outlet chamber. An average value of 0.04 mg/L is the balance in the concentrations of Zn in the effluents of these chambers. This translates to the amount of manganese removed during treatment at the time of the study.

Manganese is highest in the inlet chamber sludge samples of location C at 38.7 mg/kg. It ranged from a value of 17.30 mg/kg in the inlet chamber in location B to 38.7 mg/kg in the inlet chamber in location C in the order of magnitude.

Lead

Lead ranged from 0.062 – 0.830 mg/L in the effluent samples of the inlet chamber. Lead was next to Fe in the abundance in the study area. Its values varied from 0.015 – 0.750 mg/L in the semi-treated chamber in the study area.

A mean value of 0.33 mg/L was recorded in the effluent for Pb in the raw chambers in the study. While an average value of 0.28 mg/L was obtained in the outlet chamber in Table 2. This translates to 0.05 mg/L being the net value of Pb precipitated out of the effluent of the raw chamber.

The sludge sample lead concentrations were significantly high in all the sample locations. 2.34 mg/kg was the highest reported Pb in the study for the inlet chamber sludge samples, and this was obtained in location C (Sapele area). Meanwhile, the figures of Pb in the raw chamber sludge samples in the study location B and A is 1.62 and 0.77 mg/kg. That is, an average of 1.58 mg/kg Pb was contributed to the sludge in the inlet chambers in the study area.

Copper

Copper had the highest concentration in the effluent samples of the inlet chamber in location B posting a value of 0.26 mg/L, above the 0.20, and 0.07 mg/L obtained in the other two locations (A and C) respectively. Cu was similarly observed in concentrations of 0.16, 0.17, and 0.06 mg/L in the outlet chamber effluent samples in the study area. A value of 0.18 mg/L was calculated as the average value of Cu in the inlet chambers in the study. Meanwhile, 0.13 mg/L was obtained as the mean for Cu in all the effluent samples of the outlet chamber in this study.

The sludge sample in the inlet chambers had Cu levels of 10.4, 9.45, and 13.5 mg/kg for the three study locations in alphabetical order.

Chromium

Analysis of the data for chromium showed that chromium (Cr) concentration was 0.64 mg/L, 0.084, and 0.045 mg/L in the inlet chamber effluent samples in locations A, B and C respectively. Whereas, the values were 0.51, 0.047, and 0.019 mg/L respectively in locations A, B, and C in the outlet chamber effluent samples. These account for 0.130, 0.037, and 0.026 mg/L, as the differences in Cr values in both chambers. Similarly, these values represent the levels of Cr considered to be present in the sludge of the inlet chamber of the septic tank. A mean value of 0.07 mg/L of Cr was considered to have been deposited in the sludge in the inlet chamber of the study area.

Total chromium had the following values in the inlet chamber: 1.35, 1.47, and 2.62 mg/kg for locations A, B, and C.

Cadmium

Cadmium was observed to be below the limit of detection (BDL) in several places in locations B and C. It recorded values of 0.44 mg/L, below the detection limit in location B, and 0.03 mg/L respectively in effluents of the inlet chamber. It was 0.35, BDL, and BDL respectively in the outlet chamber.

The following values of cadmium were reported in the inlet chambers of the three locations, as below the detection limit (BDL), 1.10, and 3.44 mg/kg of three locations (A, B, C) in that order.

Nickel

Nickel (Ni) was BDL in the inlet chamber effluent samples in location A. Effluent samples Ni values in the raw chamber were 0.06 and 0.03 mg/L in locations B and C. Outlet effluent Ni concentrations in locations A, B, and C was BDL, 0.03, and 0.01 mg/L. This implies that 0.03 and 0.02 mg/L of Ni were the mean values in both chamber effluents.

In the inlet chambers' sludge samples from all the study locations, Ni concentrations were reported as 0.95 mg/kg in location A, 0.81 mg/kg in location B, and 1.64 mg/kg in location C. Ni concentration was highest in location C at 1.64 mg/kg and least at 0.81 mg/L in location B of all the three sample locations.

Vanadium

The least abundant heavy metal in the study was vanadium. Although it was not detected in location A, values of 0.04 and 0.02 mg/L were however recorded in locations B and C.

Vanadium concentrations of 0.86, 0.77 and 1.52 mg/kg were observed in the raw sludge samples in locations A, B, and C.

From the foregoing, it is apparent that a dynamic correlation exists between the concentrations of dense metals in the septic tank effluent and sludge components. This dynamic relationship accounts for the distribution of these metals in the system – inlet (effluent and sludge) and the outlet chamber effluent. In the outlet chamber, percolation is high if hydraulic conductivity is similarly high depending on the soil texture. Therefore, sludge and other materials in this chamber would be expected to gradually seep away.

3.3 Correlational Analysis of trace metals in effluent and sludge samples

Table 4. Results of A) the correlational, B) two-tail analysis of variance (ANOVA) with p-value at 0.05 level of significance computed for the effluent and sludge samples heavy metals concentrations in location A using Statistical Package for the Social Sciences (SPSS) software.

A) Correlational analysis

	Effluent Conc. (mg/L)	*Sludge Conc. (mg/kg)*
Effluent Conc. (mg/L)	1	0.896541259
Sludge Conc. (mg/kg)	0.896541259	1

B) ANOVA

	df	*SS*	*MS*	*F*	*Significance F*	*P-value*
Regression	1	1007326	1007326	28.67538	0.001058547	**0.001058547**
Residual	7	245900.2	35128.6			
Total	8	1253226				

Table 5. Results of A) the correlational, B) two-tail analysis of variance (ANOVA) with p-value at 0.05 level of significance computed for the effluent and sludge samples heavy metals concentrations in location B using Statistical Package for the Social Sciences (SPSS) software.

A) Correlational analysis

	Effluent Conc. (mg/L)	*Sludge Conc. (mg/kg)*
Effluent Conc. (mg/L)	1	0.999558439
Sludge Conc. (mg/kg)	0.999558439	1

B) ANOVA

	df	*SS*	*MS*	*F*	*Significance F*	*P-value*
Regression	1	10163249.92	10163249.92	7921.173678	5.95413E-12	**5.95413E-12**
Residual	7	8981.339427	1283.04849			
Total	8	10172231.26				

Table 6. Results of A) the correlational, B) two-tail analysis of variance (ANOVA) with p-value at 0.05 level of significance computed for the effluent and sludge samples heavy metals concentrations in location C using Statistical Package for the Social Sciences (SPSS) software.

A) Correlational analysis

	Effluent Conc. (mg/L)	***Sludge Conc. (mg/kg)***
Effluent Conc. (mg/L)	1	0.995763396
Sludge Conc. (mg/kg)	0.995763396	1

B) ANOVA

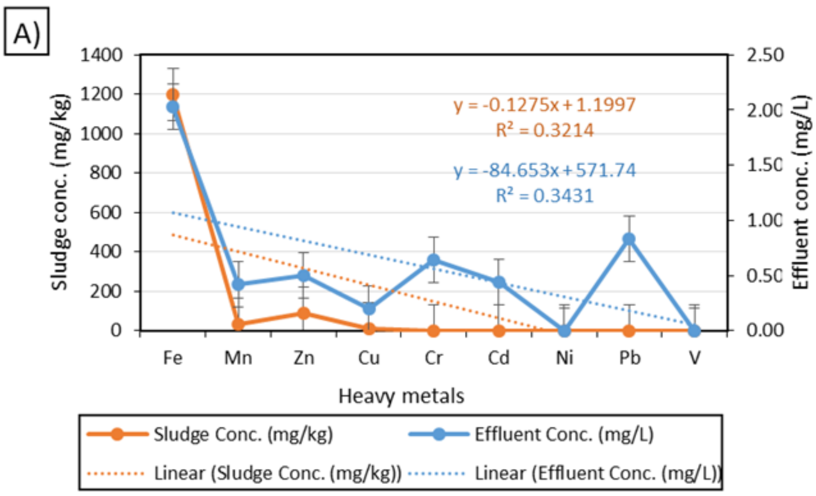
	df	*SS*	*MS*	*F*	*Significance F*	*P-value*
Regression	1	323812.0623	323812.0623	820.8871846	1.62297E-08	**1.62297E-08**
Residual	7	2761.261813	394.4659733			
Total	8	326573.3241				

Tables 4, 5 and 6 display the results of the correlation and analysis of variance (ANOVA) at a 0.05 significance level for trace metal concentrations in both the effluent and sludge samples from septic tanks in the study area. Correlation data reveal a high degree of relationship between the effluent heavy metal concentrations and the concentrations in the sludge component. The implication is that there was a dynamic interface between the effluent and sludge phases of the system. This further demonstrates that metallic ions were indeed precipitated out of the effluent, and cascaded down to the bottom of the tank where they accumulated in sludge. The correlation values were high in all the sample locations with values of r = 0.89, r = 0.99, and r = 0.99 respectively recorded for location A, B, and C in that order.

A two-tailed analysis of variance (ANOVA) computation (Table 4B, 5B, and 6B respectively) proved that both the effluent and sludge data were independently exclusive of each. It tells us that these two data sets have means that are statistically different from each other even though they are highly correlated.

The p-values (in green) obtained in this study (Tables 4b, 5b, and 6b) correspond with the F-significance values also in Table 4b, 5b, and 6b) illustrating in numerical terms the significance of the Pearson's correlation r-values earlier reported above. P-values test the significance of the correlation between a set of variables. In this study, p-values were resolved at a 0.05 level of significance. And all p-values obtained in Tables 4b, 5b, and 6b were relatively lower than the 0.05 significant value. Therefore, we conclude that the correlation between the effluent and sludge heavy metal concentrations was highly significant.

3.4 Regression Analysis and R-square values of effluent/sludge proportions



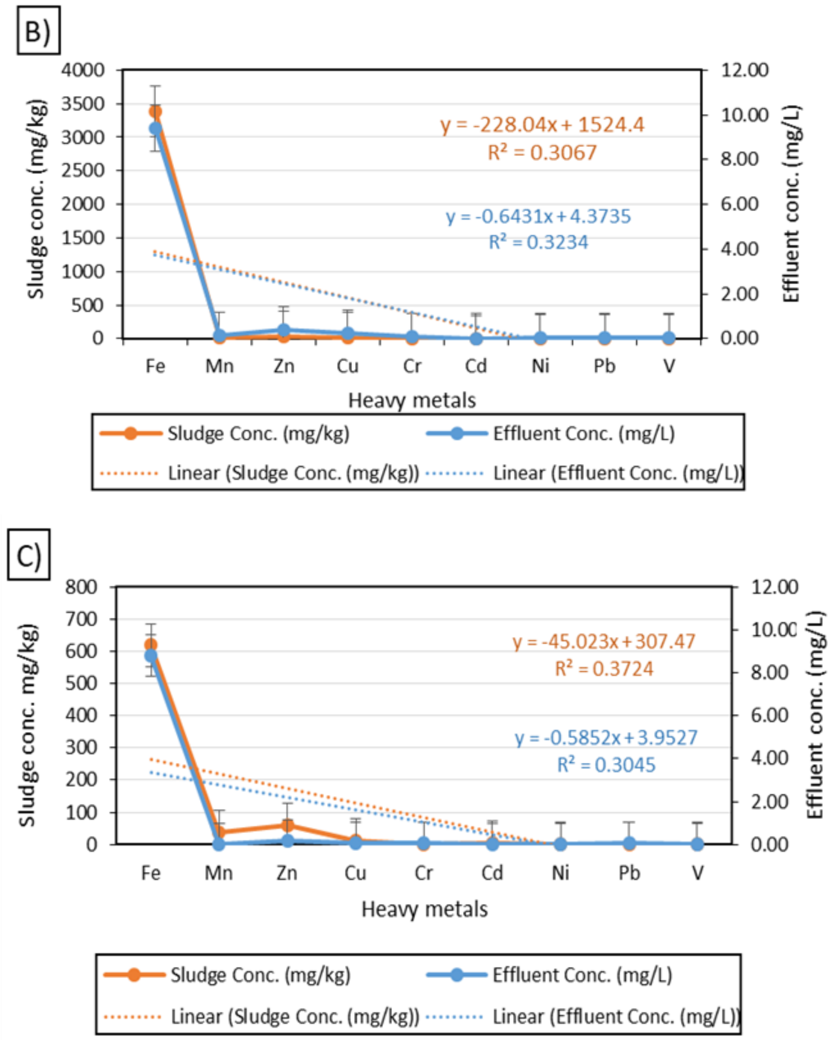


Figure 3. Multivariate chart of heavy metals types (x-axis) vs concentration of heavy metals in sludge y-axis (left) with orange colour and concentration of heavy metals in the effluent (right) with blue colour; the dotted lines (orange and blue represent lines of best fit). Inset (right) is the regression equation and the R-square values; underneath is the chart legend. All coordinates indicated by error bars. (A) Location A (Agbor), (B) Location B (Benin City), (C) Location C (Sapele).

Regression analysis of data from the trace metals concentrations for the effluent and sludge were determined and the result is presented in Figures 3a, 3b, and 3c representing the different sample locations A, B, and C consecutively. These results further show a relationship between the effluent and sludge concentrations in the septic tanks in this study. This is further exemplified in the values of the correlation analysis obtained in the previous section 1.3.2. (Tables 4a, 5a, and 6a). Correlation analysis typically identifies the relationship between two variables in a study. This analysis was considered necessary because of the difference in the concentration units of the effluent and sludge samples.

In each of the charts above, the R-square values were relatively constant. R-square statistically speaking is a measure of the proportion of the variation between two sets of variables. These R-square constants had a range of values between 0.3045 – 0.3724 in the study, with a mean value of 0.33 determined from the individually pooled R-square values for each of the study locations. The R-square values for the variations in the effluent and sludge heavy metal concentrations are essentially similar. Also, from the multivariate charts in Figures 3a, 3b, and 3c, we can tell that there is a higher proportion of heavy metals yet in the effluent than in the sludge samples. The similarity in the R-square values is considered an indication of the connection between the heavy metal concentration of the effluent and the sludge samples.

4. Conclusion

Heavy metals' spatial distribution in the effluent and sludge components of septic tank systems was studied to understand and account for the distribution of the heavy metals within the system. Findings confirm that these metals were indeed bio-precipitated out of the effluent component of the raw chamber during the process of pH-controlled microbial remediation. From where they gradually cascaded under the influence of gravity onto the bottom of the tanks as a component of sludge. Results also showed that sludge samples in the anaerobic bioreactors contained a high concentration of various trace metals. A dynamic relationship was established between the dense metals in both phases of the inlet chamber in the septic tanks. R-square values recorded in the study showed that at a given pH condition, the variations in the concentrations of the heavy metals in the effluent and sludge components of a septic tank system are approximately constant. The trace metal concentrations in both phases of the septic tank system were strongly correlated. All the heavy metals analysed in the effluent were similarly observed in the sludge, *albeit*, at lower concentrations. Thereby demonstrating that these heavy metals emanated from the effluent component of the system. There was always a higher proportion of heavy metals present in the inlet chamber than there is in the outlet chamber. Nonetheless, the inlet chamber sludge contained significant proportions of heavy metal concentrations with serious implications on the environment in respect of groundwater and landfills contamination.

Conceptualization

HHO and IMO developed the methodology. IMO picked the samples. HHO prepared and wrote the original draft. IMO proofread the article.

Conflict of Interest

The author declares that there is no conflict of interest.

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Data Availability Statement

All relevant data are included in this article.

References

1. Abatenh, E.; Gizaw, B.; Tsegaye, Z.; Misganaw, W. The Role of Microorganisms in Bioremediation - A Review. *Open J. Environ. Biol.* **2017**, *2* (1), 038–046. https://doi.org/10.17352/ojeb.000007.
2. Ademoroti, C. M. A. *Standard Methods for Water and Effluents Analysis*; Foludex Press Limited: Ibadan.
3. Tang, Y. Z.; Gin, K. Y. H.; Aziz, M. A. The Relationship between pH and Heavy Metal Ion Sorption by Algal Biomass. *Adsorpt. Sci. Technol.* **2003**, *21* (6), 525–538. https://doi.org/10.1260/026361703771953587.
4. Canovas, D.; Cases, I.; de Lorenzo, V. Heavy Metal Tolerance and Metal Homeostasis in Pseudomonas Putida as Revealed by Complete Genome Analysis. *Environ. Microbiol.* **2003**, *5*, 1242–1256.
5. Delangiz, N.; Varjovi, M. B.; Lajayer, B. A.; Ghorbanpour, M. Beneficial Microorganisms in the Remediation of Heavy Metals. In *Molecular Aspects of Plant Beneficial Microbes in Agriculture*; 2020; pp 417–423. https://doi.org/10.1016/b978-0-12-818469-1.00034-1.
6. Hassan, M. A.; Salleh, A.; Milow, P. Characterization of the Adsorption of the Lead (II) by the Nonliving Biomass Spyrogyia Neglecta (Hasall) Kutzing. *Am. J. Biochem. Biotecnol.* **2009**, *5* (2), 75–83. https://doi.org/10.3844/AJBBSP.2009.75.83.
7. Igiri, B. E.; Okoduwa, S. I. R.; Idoko, G. O.; Akabuogu, E. P.; Adeyi, A. O. E. I;. Toxicity and Bioremediation of Heavy Metals Contaminated Ecosystems from Tannery Wastewater: A Review. *J. Toxicol.* **2018**, 1–16. https://doi.org/2568028.
8. Jin, X.; Streett, D. A.; Dunlap, C. A.; Lyn, M. E. Application of Hydrophilic-Lipophilic Balance (HLB) Number to Optimize a Compatible Non-Ionic Surfactant for Dried Aerial Conidia of Beauveria Bassiana. *Biol. Control* **2008**, *46* (2), 226–233. https://doi.org/10.1016/j.biocontrol.2008.03.008.
9. Osman, G. E. H.; Abulreesh, H. H.; Elbanna, K.; Shaaban, M. R.; Samreen; Ahmad, I. Recent Progress in Metal-Microbe Interactions: Prospects in Bioremediation. *Journal of Pure and Applied Microbiology*. **2019**, pp 13–26. https://doi.org/10.22207/JPAM.13.1.02.
10. Rose, J.; Jiménez Cisneros, B.; Adegoke, A. A.; Stenstrom, T.-A. Septic Systems. In *Water and Sanitation for the 21st Century: Health and Microbiological Aspects of Excreta and Wastewater Management (Global Water Pathogen Project)*; Rose, J. B., Jiménez-Cisneros, B., Eds.; Michigan State University: Michigan, 2016; pp 1–16. https://doi.org/10.14321/waterpathogens.59.
11. Yuncu, B.; Sanin, F. D.; Yetis, U. An Investigation of Heavy Metal Biosorption in

Relation to C/N Ratio of Activated Sludg. *J. Hazard. Mater.* **2006**, *137*, 990–997.
12. Diep, P.; Mahadevan, R.; Yakunin, A. F. Heavy Metal Removal by Bioaccumulation Using Genetically Engineered Microorganisms. *Front. Bioeng. Biotechnol.* **2018**, *29*. https://doi.org/https://doi.org/10.3389/fbioe.2018.00157.
13. Zhang, Xudong. Ye, Peng. Wu, Yajun. Guo, Zhen. Huang, Yuan.Zhang, Xingtao. Sun, Yuncong. Zhang, H. Research on Dewatering Ability of Municipal Sludge under the Treatment of Coupled Acid and Microwave. *Hindawi* **2021**, *2021*, 1–11. https://doi.org/https://doi.org/10.1155/2021/71611815.
14. Qadir, M. S.; Abed, M. F.; Al-Hadad, K. A. Environmental risk assessment of heavy metals contamination in the sediment of mandali dam reservoir eastern iraq. *Niger. Soc. Phys. Sci.* **2022**, *4* (2), 543–555. https://doi.org/10.5281/zenodo.7433083.
15. Ajah, D. N.; Agboeze, E.; Ihedioha, J. N.; Hukwudi-Madu, E.; Chime, C. C. Levels of Zinc (Zn), Copper (Cu), Iron (Fe), and Cadmium (Cd) in Soil, Rice Stalk, and Oryza Sativa Grain in Ishiagu Rice Field, Ebonyi State, Nigeria; Human Health Risk. *Niger. Soc. Phys. Sci.* **2022**, *4*, 891. https://doi.org/DOI:10.46481/jnsps.2022.891.
16. Ali, H.; Khan, E.; Ilahi, I. Environmental Chemistry and Ecotoxicology of Hazardous Heavy Metals: Environmental Persistence, Toxicity, and Bioaccumulation. *Journal of Chemistry*. 2019, pp 1–14. https://doi.org/10.1155/2019/6730305.
17. Briffa, J.; Sinagra, E.; Blundell, R. Heliyon Heavy Metal Pollution in the Environment and Their Toxicological Effects on Humans. *Heliyon* **2020**, *6*, e04691. https://doi.org/10.1016/j.heliyon.2020.e04691.
18. Chu, D. Effects of Heavy Metals on Soil Microbial Community. In *IOP Conference Series: Earth and Environmental Science*; 2018; Vol. 113, pp 1–6. https://doi.org/10.1088/1755-1315/113/1/012009.
19. Hawkes, S. J. What Is “Heavy Metals”? *J. Chem. Educ.* **1997**, *74* (11), 1374.
20. Sayo, S.; Kiratu, J. M.; Nyamato, G. S. Heavy Metal Concentrations in Soil and Vegetables Irrigated with Sewage Effluent : A Case Study of Embu Sewage Treatment Plant , Kenya. *Sci. African* **2020**, *8*, e00337. https://doi.org/10.1016/j.sciaf.2020.e00337.
21. Ibrahim, M.; Ibrahim, N. The Relations Between Concentration of Iron and the pH of Ground Water (Case Study Zulfi Ground Water). *Int. J. Environ. Monit. Anal.* **2016**, *4* (6), 140. https://doi.org/10.11648/j.ijema.20160406.11.
22. Tchounwou, P. B.; Yedjou, C. G.; Patlolla, A. K.; Sutton, D. J. Molecular, Clinical and Environmental Toxicicology Volume 3: Environmental Toxicology. *Mol. Clin. Environ. Toxicol.* **2012**, *101*, 133–164. https://doi.org/10.1007/978-3-7643-8340-4.
23. Nkwunonwo, U. C.; Odika, P. O.; Onyia, N. I. A Review of the Health Implications of Heavy Metals in Food Chain in Nigeria. *Sci. World J.* **2020**, *2020*, 1–11. https://doi.org/10.1155/2020/6594109.
24. He, Z.; Yang, X.; Stoffella, P. Trace Elements in Agroecosystems and Impacts on the Environment. *J Trace Elem Med Biol.* **2005**, *19* (2–3), 125–140.
25. Usman, Z. U.; Mukesh, Y.; Vandana, S.; Sharma, J. K.; Sanjay, P.; Sidhartha, D.; Anil, S. K. Microbial Bioremediation of Heavy Metals: Emerging Trends and Recent Advances. *Res. J. Biotechnol.* **2020**, *15* (1), 164–178.
26. Holliger, C.; de Laclos, H. F.; Hack, G. Methane Production of Full-Scale Anaerobic Digestion Plants Calculated from Substrate’s Biomethane Potentials Compares Well with the One Measured on-Site. *Front. Energy Res.* **2017**, *5* (JUN), 1–9. https://doi.org/10.3389/fenrg.2017.00012.
27. Jiménez-Ocampo, U. E.; Vargas, A.; Moreno-Andrade, I. Methane Production from Food Waste Using a Feedback Control Strategy in a Sequencing Batch Reactor. *Water Sci. Technol.* **2021**, *84* (8), 1969–1980. https://doi.org/10.2166/wst.2021.370.
28. Klassen, V.; Blifernez-Klassen, O.; Wibberg, D.; Winkler, A.; Kalinowski, J.; Posten, C.; Kruse, O. Highly Efficient Methane Generation from Untreated Microalgae Biomass. *Biotechnol. Biofuels* **2017**, *10* (1), 1–12. https://doi.org/10.1186/s13068-017-0871-4.
29. Okoro, E. E., Okafor, I. S., Igwilo, K. C., Orodu, K. B., Mamudu, A. O. Sustainable Biogas Production from Wastein Potential States in Nigeia. *Altern. Source Energy* **2020**, *38* (14), 627–643. https://doi.org/10.1080/02589001.2020.1825650.
30. Romero, H. I., Vega, V., Feijoo, D. V., Sacramento, C. Methane Production through Anaerobic Co-Digestion of Tropical Fruits Biomass and Urban Solid

Waste. *Energy Reports* **2020**, *6* (8), 351–357. https://doi.org/10.1016/j.egyr.2020.11.17.0.

31. Abdeldayem, R. A Preliminary Study of Heavy Metals Pollution Risk in Water. *Appl. Water Sci.* **2020**, *10* (1), 1–4. https://doi.org/10.1007/s13201-019-1058-x.
32. Kinuthia, G. K.; Ngure, V.; Beti, D.; Lugalia, R.; Wangila, A.; Kamau, L. Levels of Heavy Metals in Wastewater and Soil Samples from Open Drainage Channels in Nairobi, Kenya: Community Health Implication. *Sci. Rep.* **2020**, *10* (1), 1–13. https://doi.org/10.1038/s41598-020-65359-5.
33. Mehr, M.P., Hekmati, M., Shayesteh, K. Investigation of Heavy Metals in Drinking Water: A Systematic Review in Iran. *J. Adv. Environ. Heal. Res.* **2020**, *8* (3), 216-224. https://doi.org/10.22102/jaehr.2020232374.1170.
34. Mohod, C. V; Dhote, J. Review of Heavy Metals in Drinking Water and Their Effect on Human Health. *Int. J. Innov. Res. Sci. Eng. Technol.* **2013**, *2* (7), 2992–2996.
35. Oyem, H. H.; Oyem, I. M. Heavy Metals pH-Mediated Microbial-Remediation in Septic Tank Effluents. **2022**, *4* (1), 15–35.
36. Avwunudiogba, A. A Comparative Ananlysis of Soil and Nutrient Losses on Maize Plant with Different Tillage Practices in Ikpoba River Basin of South-Western Nigeria. *Niger. Geogr. Journal, New Ser.* **2000**, *3* (4), 199–208.
37. Oyem, H. H., Oyem, I. M., and Ezenweali, D. Temperature, pH, Electrical Conductivity, Total Dissolved Solids and Chemical Oxygen Demand of Groundwater in Boji-Boji Agbor/Owa Area and Immediate Suburbs. *Res. J. Environ. Sci.* **2014**, *8* (8), 444–450.
38. Oyem, I. M., Oyem, H. H., Oyem, M. N., Usese, A. I., Ezeweali, D., and Obiwulu, E. N. O. An Investigation into Groundwater Contamination in Agbor and Owa Communities in Nigeria. *Sacha Journals Environ. Stud.* **2015**, *5* (1).
39. Oyem, H. H., and Oyem, I. M., and Usese, A., I. Iron, Manganese, Cadmium, Chromium, Zinc, and Arsenic Groundwater Contents of Agbor and Owa Communities of Nigeria. *Springerplus* **2015**, *4*, 104. https://doi.org/10.1186/s40064-015-0867-0.
40. National Bureau of Statistics. Demographic Statistics. In *Alpacas, Sheep, and Men*; 1977; pp 219–228. https://doi.org/10.1016/b978-0-12-527850-8.50022-5.
41. Britannica, T. E. of E. Benin City. *Britannica*; Encyclopedia Britannica, 2019.
42. Ugbomeh, B. Household Size and Socio-Economic Resources: A Case Study of Ika North East Local Government Area of Delta State of Nigeria. *African Res. Rev.* **2011**, *5* (1), 226–238. https://doi.org/10.4314/afrrev.v5i1.64522.
43. Ademoroti, C. M. A. *Standard Methods for Water and Effluents Analysis*, First.; Foludex Press Limited: Ibadan, 1996.
44. APHA. *Standard Methods for the Examination of Water and Waste Water*, 20th ed.; American Public Health Association: Washington, DC, 1995.
45. United Nations Children’s Fund. *Handbook on Water Quality.*; New York, 2008.
46. Agoro, M. A.; Adeniji, A. O.; Adefisoye, M. A.; Okoh, O. O. Heavy Metals in Wastewater and Sewage Sludge from Selected Municipal Treatment Plants in Eastern Cape Province, South Africa. *Water (Switzerland)* **2020**, *12* (10), 1–19. https://doi.org/10.3390/w12102746.
47. Duan, B.; Zhang, W.; Zheng, H.; Wu, C.; Zhang, Q.; Bu, Y. Comparison of Health Risk Assessments of Heavy Metals and As in Sewage Sludge FromWastewater Treatment Plants (WWTPs) for Adults and Children in the Urban District of Taiyuan, China. *Int. J. Environ. Res. Public Heal.* **2017**, *14*, 1194.
48. Steinhardt, G. C.; Egler, C. J. *Home & Environment*; 2018.

