CHEMICAL CHARACTERISTICS OF HUMIC ACIDS IN RELATION TO LEAD, COPPER AND CADMIUM LEVELS IN CONTAMINATED SOILS FROM SOUTH WEST NIGERIA

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ABSTRACT

Humic acids (HAs) extracted from topsoils obtained from the highways, motor parks and auto-repair workshops in Lagos, Ibadan and Abeokuta, Nigeria were characterized by infrared spectroscopy, titration and UV-visible spectroscopy for reactive groups. The relative abundance of metals as determined by atomic absorption spectrophotometry decreased in the order Pb > Cu > Cd. Concentrations of soil total Cd (1.7 ± 0.2 to 1.9 ± 0.2 mg/kg), Cu (184 ± 25 to 360 ± 31 mg/kg) and Pb (1039 ± 123 to 1524 ± 88 mg/kg) exceeded international reference values for uncontaminated soils. Parametric and nonparametric correlations of the metal concentrations with humic acid chemical characteristics gave both negative and positive coefficients, some of which were significant at $p \leq 0.05$. Competitive trace metal complexation reactions with an equimolar solution of the cations revealed that the HAs bound more Pb (0.911 mmol/g) than Cu (0.779 mmol/g) or Cd (0.237 mmol/g). The oxygen-containing groups (carboxyl and phenol), the chromophores that absorbed in the UV-visible region and the hydrophobic nature (E_4/E_6 ratios from 4.4 ± 0.2 to 5.7 ± 0.2) of the humic acids affect soil metal sorption.

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1. INTRODUCTION

Research on heavy metal deposition and accumulation in the environment is the current attention of most environmental scientists in Nigeria. Studies often focus on heavy metal concentrations in foods, vegetables, water, hematological parameters, atmosphere and soil, giving attention only to the determination of metal concentrations or speciation in soil [1-8]. There is a scarcity of literature reports with respect to the influence of soil organic matter on the sorption of these toxic metals in soils. In Nigeria, automobile exhaust emissions account for the greater percentage of metal release, especially lead, into the atmosphere, which eventually gets deposited in the surrounding soils. Soil environments in locations associated with automobile activities like auto-repair workshops, motor parks and highways are vulnerable to contamination by metals such as lead, cadmium, copper, zinc and nickel, which often are used as major or minor additives in gasoline and auto-lubricants [8].

The understanding of heavy metal accumulation in soil is an important requirement in environmental science. Sorption of heavy metals in soil is influenced by soil factors such as clay, pH, cation exchange capacity and organic matter content [9]. Organic carbon in soil consists basically of humic substances, which are formed by decomposition of natural organic matter. Humic substances are powerful complexing and chelating entities whose sorption characteristics or properties depend on their chemical composition. In this study, cadmium, copper and lead concentrations in topsoils of auto-repair workshops, highways and motor parks in three major cities of southwest Nigeria were determined and correlated with basic chemical characteristics of humic acids extracted from the soils. Complexation of the isolated humic acids with equimolar solutions of the three cations (Cd^{2+}, Cu^{2+} and Pb^{2+}) and their subsequent desorption from the complexes by batch leaching tests using water were also carried out.

2. MATERIALS AND METHODS

2.1 Study Areas

Nigeria is located in the tropical zone between latitude 4° and 14° N and longitude 2° E with a vast area of savanna vegetation. Due to its diverse nature, the vegetation is further classified into various agro-

ecological zones, namely derived savanna, southern guinea savanna and northern guinea savanna. The soils found in the various agro-ecological zones are mostly alfisols, entisols and inceptisols, which are predominantly coarse-textured and low in organic matter (less than 4% [10, 11]). The study areas Lagos, Abeokuta and Ibadan are the respective capital cities of Lagos, Ogun and Oyo states, Nigeria and have a good network of roads.

Lagos is the most populous city in Nigeria, with an estimated human population of about 15 million. It is the commercial and industrial hub of Nigeria. Lagos was the federal capital of Nigeria until 1990. Ibadan, a cosmopolitan city, is northeast of Lagos. It is also a commercial and industrial center of southwest Nigeria with a population of about 7,560,000. Abeokuta is found north of Lagos with a human population of over 2 million [2, 11]. The motor parks, highways and auto-repair workshops are locations were automobile activities are carried out in semi-stationary and stationary designs. The construction, design and locations of motor parks and auto-repair workshops in most Nigerian cities do not always conform to official regulations. Hence they are randomly located in streets and even in residential quarters [8].

2.2. Materials

Solid humic acids used were extracted from the top-soil samples of study sites. Analytical grade chemicals utilized were hydrochloric acid (HCl), nitric acid (HNO₃), sodium hydroxide (NaOH), potassium bromide (KBr), barium hydroxide (Ba(OH)₂), calcium acetate ((CH₃COO)₂Ca), copper(II) nitrate (Cu(NO₃)₂), cadmium chloride (CdCl₂), hydrofluoric acid (HF) and lead nitrate (Pb(NO₃)₂).

2.3. Methods

2.3.1 Soil Sampling and Analysis

In soil sampling, thirty highways, ten each from Abeokuta, Ibadan and Lagos were selected. These, in addition to motor parks (five from each city) and auto-repair shops (five from each city), gave a total of sixty soil samples used in this study. At each of the 60 locations, soil samples were collected at a depth of 0 – 5 cm from various sampling points and mixed to give a composite sample, which was stored in polythene bags and transported to the laboratory. The samples were air dried for 5 days at room temperature of 299 K, crushed in a porcelain mortar and then sieved through a 2 mm mesh prior to chemical analyses. Soil mechanical analysis was carried out by the hydrometer method, while pH and electrical conduct-

ivity were measured in a 1:5 soil to water suspension with a HANNA series pH meter (M-4-24) and conductivity meter (WTW, LF 90) using standard procedures [12].

Metal determination was carried out by wet digestion. A soil sample (1.0 g) was quantitatively transferred to a 125 mL Erlenmeyer flask, which had been previously soaked overnight in acidified distilled water and washed with deionized-distilled water. Aqua regia (30 mL) was added, allowed to stand overnight and then evaporated to about 4 mL. This was followed by the addition of 20 mL HNO₃: HClO₄ (1:5). Digestion was carried out in a fume hood by raising the temperature gradually to 363 K. A further 10 mL of the acid mixture was added until the solution was colorless, after which it was evaporated to about 2 mL. The digest was allowed to cool, filtered with Whatman No. 1 paper and made up to the mark in a 50 mL standard flask with deionized-distilled water. Five blank determinations were also carried out.

The concentrations of lead, cadmium and copper in the filtrate were determined with a Buck Model 210 atomic absorption spectrophotometer (Buck Scientific, USA), using air-acetylene-oxygen gas mixtures. The respective absorption lines for the metals were 288.8 nm for Cd, 324.8 nm for Cu and 282.3 nm for Pb using the respective hollow cathode lamps. The instrument was calibrated with standards diluted from commercial BDH (1000 mg/kg) stock in the concentration range 1 to 100 mg/kg for Pb, 0.02 to 1.0 mg/kg for Cd and 1 to 50 mg/kg for Cu. Samples whose metal concentrations overshot the calibration ranges were diluted and dilution factors were used in the final calculations. Sensitivity checks on the instrument were carried out with 0.2 mg Cd/kg, 15 mg Pb/kg and 2 mg Cu/kg at an absorbance value of 0.2. Recoveries of 97 ± 3% (Pb), 94 ± 2% (Cd) and 97 ± 4% (Cu) were obtained after the addition of standards.

2.3.2 Extraction of Soil Humic Acids

Humic acid was extracted by the dilute alkali method [9]. Crushed and air-dried soil samples (sieved through 2 mm mesh) were decalcified by equilibrating with 0.1 M HCl for 24 hrs at room temperature (301 ± 1 K). The dried residue was extracted with 0.1 M NaOH. The experiment was carried out in the presence of N₂ to minimize chemical changes due to autoxidation [9]. The supernatant was separated from the residue by centrifugation (450 rpm for 5 min) and decantation techniques. The alkaline extraction was repeated several times until extraction was completed, as signaled by an almost colorless alkaline extract. The black humic solution was fractionated by pH adjustment to 2.0 using 10 mL of 5 M HCl. The

system was then allowed to stand for 24 hrs at room temperature (301 ± 1 K) for the coagulated humic acid to settle at the bottom of the plastic ware. The black humic acid precipitate was separated by centrifugation and purified by re-dissolution in 0.1 M NaOH, re-precipitation by acidification (5 M HCl and 0.1 M HF) to remove possible silicates. The humic acid precipitate was finally washed several times with deionized water until the wash water tested negative to chloride and hydroxide ions by means of silver nitrate and phenolphthalein tests. The purified humic acid was then dried, ground to powder and stored at room temperature prior to chemical analysis.

2.3.3 Chemical Characterization of Soil Humic Acid

Ash content determination and infra-red analysis. Humic acid (100 mg) was combusted at 550°C for one hour in a Gallenkamp muffle furnace. The ash content was then calculated gravimetrically. The infra-red (IR) spectra were obtained by mixing 1 mg humic acid sample with 100 mg KBr and measurement of the KBr disc using a Buck Model 500 IR spectrometer (Buck Scientific, USA, scan time 3 mins) interfaced to an IBM PC and a plotter. The resulting spectra were processed using Grams/IR software for Windows$^®$ (Galactic Industries Corp., Hampshire, USA). Arithmetic Transmission and Smooth programs were used for background file and spectral resolution, respectively.

    Determination of acid group content. Acidic functional group contents were determined by titration. Total acidity groups were determined using the barium hydroxide method and the calcium acetate method was used for carboxylic acid group analysis. Phenolic hydroxyl group content was obtained by difference (total – carboxylic acid groups) [9, 13].

    Absorption in the UV-visible region. A humic acid sample (1.0 mg) was dissolved in 10 mL of 0.05 M NaHCO$_3$ solution adjusted to pH 8.5 with 0.1 M NaOH to enhance solubility. The light-brown colored solution was transferred into quartz cells and the spectrum of absorbance *vs.* wavelength (200-800 nm) was then recorded on a Helious Gamma Unicam 2000 absorption spectrophotometer equipped with Aurora for Helios Scan software. The E$_4$/E$_6$ ratio was calculated as the quotient of the absorbances at 465 nm (E$_4$) and 665 nm (E$_6$) using the solution described by Chen et al. [14].

2.3.4 Competitive Trace Metal –Humic Acid Complexation Reactions

The isolated soil humic acid was treated with an equimolar solution containing 0.1 M Cu(NO$_3$)$_2$, 0.1 M CdCl$_2$ and 0.1M Pb(NO$_3$)$_2$ at 100mg HA to 20 mL solution according to Senesi et al. [15]. The system was allowed to equilibrate at room temperature for 24 hours for maximum chemical reaction, after which it was filtered. The HA-metal complexes were air-dried, ground to fine powder and a 200 mg sample of either HA or HA-metal complex was then analyzed for Pb, Cu and Cd contents by atomic absorption spectro-photometry.

2.3.5 Batch Desorption of Metal from the Humic Complex

The effect of pH on the stability of Cd, Cu and Pb in the humic acid–metal complexes was evaluated by leaching fresh fractions of the HA-metal complexes with deionized-distilled water adjusted to three pH values of 1.95, 4.32 and 5.44 using 0.1 M HCl. A batch-leaching test was then conducted on 100 mg HA-metal complex using 20 mL of the water. The solution was shaken at 450 rpm for 5 minutes. This was repeated five times and the leachates of each sample were combined to give a 100 mL sample aliquot, which was analyzed for the three metal concentrations by atomic absorption spectrophotometry.

2.3.6 Statistics

Statistical analyses were performed by the use of SPSS 10.0 software for Windows$^®$. Statistical descriptions (mean, standard error, median, modes, maximum, minimum and percentiles), Pearson parametric correlations, Spearman and Kendall non-parametric correlations, paired samples T – tests and inter-element correlations were performed on the data.

3. RESULTS

3.1. Soil Characteristics

The metal concentrations obtained at the different sampling points are presented in Tables 1-3. Tables 4–6 contain a summary of topsoil characteristics of the study sites. Particle size distribution varied from 7.8 ± 0.5 to 8.3 ± 0.4 % for silt, 1.4 ± 0.1 to 1.5 ± 0.4 % for clay and 90.4 ± 0.4 to 91 ± 1% for sand. The humic acid yields ranged from 0.5 ± 0.2 to 1.6 ± 0.3%. The soil pH values were obtained in the range 6.0 ± 0.2 to 6.6 ± 0.2 for the three soil systems. Mean metal concentrations varied from 1.7 ± 0.2 to 1.9 ± 0.2 mg/kg (Cd), 184 ± 25 to 360 ± 31 mg/kg (Cu) and 1039 ± 123 to 1524 ± 88 mg/kg (Pb). The topsoils were sandy, slightly acidic and the metal concentrations

exceeded the threshold levels of 50, 1 and 50 mg/kg for Pb, Cd and Cu respectively, of Netherlands limits for uncontaminated soils as reported by Alloway [16].

Table 1 Cadmium, copper and lead concentrations (mg/kg) in topsoil from various sampling points on the highway

Sample code	City	Cd	Cu	Pb
ABHWS-1	Abeokuta	2.1	462	530
ABHWS-2	Abeokuta	2.0	560	483
ABHWS-3	Abeokuta	2.0	124	154
ABHWS-4	Abeokuta	1.9	381	1210
ABHWS-5	Abeokuta	2.1	161	540
ABHWS-6	Abeokuta	4.8	197	216
ABHWS-7	Abeokuta	1.1	195	449
ABHWS-8	Abeokuta	1.8	339	590
ABHWS-9	Abeokuta	1.1	449	694
ABHWS-10	Abeokuta	1.1	386	774
IBHWS-1	Ibadan	1.6	282	526
IBHWS-2	Ibadan	2.2	426	967
IBHWS-3	Ibadan	1.0	248	691
IBHWS-4	Ibadan	1.0	474	1081
IBHWS-5	Ibadan	1.0	401	1881
IBHWS-6	Ibadan	1.5	394	1426
IBHWS-7	Ibadan	2.0	469	1960
IBHWS-8	Ibadan	0.8	331	578
IBHWS-9	Ibadan	2.7	565	1410
IBHWS-10	Ibadan	0.8	316	1148
LAHWS-1	Lagos	1.0	401	650
LAHWS-2	Lagos	2.7	781	1240
LAHWS-3	Lagos	3.0	715	1330
LAHWS-4	Lagos	1.7	384	1900
LAHWS-5	Lagos	0.9	99	670
LAHWS-6	Lagos	2.0	437	3294
LAHWS-7	Lagos	1.0	391	560
LAHWS-8	Lagos	1.1	181	1640
LAHWS-9	Lagos	1.3	162	681
LAHWS-10	Lagos	1.8	99	1890

ABHWS = Abeokuta highway soil, IBHWS = Ibadan highway soil, LAHWS = Lagos highway soil

Due to non-availability of soil quality guidelines in Nigeria for such soil environments [8], the Netherlands system was used for comparison. Hence the reference values used were 50 mg Cu/kg, 1.0 mg Cd/kg and 50 mg Pb/kg. The degrees of contamination by the metals or metal accumulation factor given in Table 7 are for Pb (20.8 ± 2.5), Cu (7.2 ± 0.6) and Cd (1.7 ± 0.2) for highway soil, and Pb (27.0 ± 2.2), Cu (6.6 ± 1.0) and Cd (1.9 ± 0.2) for auto-repair workshops. Values for Pb (30.5 ± 1.6), Cu (3.7 ± 0.5)

and Cd (1.9 ± 0.3) were obtained in motor park soils. Significant inter-element correlations (p ≤ 0.10) were only found for the pairs of Cd/Pb and Cu/Pb in highway soils with Spearman coefficients of + 0.32 for each.

Table 2 Cadmium, copper and lead concentrations (mg/kg) in auto-repair topsoil from various sampling points

Sample code	City	Cd	Cu	Pb
ABARW-1	Abeokuta	1.0	143	1744
ABARW-2	Abeokuta	0.2	180	1690
ABARW-3	Abeokuta	3.7	234	1622
ABARW-4	Abeokuta	0.9	220	1582
ABARW-5	Abeokuta	2.5	212	1640
IBARW-1	Ibadan	2.3	725	1189
IBARW-2	Ibadan	2.0	320	1690
IBARW-3	Ibadan	2.4	204	667
IBARW-4	Ibadan	2.9	481	1190
IBARW-5	Ibadan	2.4	253	1120
LAARW-1	Lagos	1.6	800	1471
LAARW-2	Lagos	1.2	490	1490
LAARW-3	Lagos	2.3	189	579
LAARW-4	Lagos	1.8	240	1902
LAARW-5	Lagos	1.0	226	677

ABARW = Abeokuta auto-repair workshop, IBARW = Ibadan auto-repair workshop, LAARW = Lagos auto-repair workshop

Table 3 Cadmium, copper and lead concentrations (mg/kg) in motor park soil topsoil from various sampling points

Sample code	City	Cd	Cu	Pb
ABMPS-1	Abeokuta	1.3	179	1983
ABMPS-2	Abeokuta	0.8	123	1483
ABMPS-3	Abeokuta	3.7	200	1677
ABMPS-4	Abeokuta	1.5	111	1942
ABMPS-5	Abeokuta	2.3	204	1106
IBMPS-1	Ibadan	1.1	110	983
IBMPS-2	Ibadan	3.8	364	1190
IBMPS-3	Ibadan	1.1	156	1749
IBMPS-4	Ibadan	2.5	161	1746
IBMPS-5	Ibadan	1.8	219	1593
LAMPS-1	Lagos	4.8	177	1426
LAMPS-2	Lagos	1.0	235	1167
LAMPS-3	Lagos	1.0	80	1482
LAMPS-4	Lagos	1.1	37	1650
LAMPS-5	Lagos	1.4	405	1676

ABMPS = Abeokuta motor park soil, IBMPS = Ibadan motor park soil, LAMPS = Lagos motor park soil

Table 4 Summary of highway topsoil characteristics

Parameter	Mean ± SE[a]	Median	Mode	Minimum	Maximum	Percentile		
						25	50	75
pH	6.6 ± 0.1	6.7	6.7	5.5	8.1	6.2	6.7	6.9
EC[b] (µS/cm)	847 ± 100	651	2000	237	2000	464	651	1052
Silt (%)	8.3 ± 0.4	8.4	7.4	7.4	9.4	6.2	6.7	6.9
Clay (%)	1.4 ± 0.1	1.4	1.4	1.3	1.5	1.4	1.4	1.5
Sand (%)	90.5 ± 0.4	91	91	90	91	90	91	91
HA (%)	0.7 ± 0.1	0.4	0.3	0.07	3.0	0.3	0.4	0.8
Pb (mg/kg)	1039 ± 123	734	154	154	3300	555	734	1414
Cd (mg/kg)	1.7 ± 0.2	1.7	1.1	0.8	4.8	1.19	1.7	2.0
Cu (mg/kg)	360 ± 31	385	99	99	781	197	385	452
n[c]	30	30	30	30	30	30	30	30

Table 5 Summary of topsoil characteristics of auto-repair workshops

Parameter	Mean ± SE[a]	Median	Mode	Minimum	Maximum	Percentile		
						25	50	75
pH	6.0 ± 0.2	5.9	6.8	4.5	7.6	5.2	5.9	6.8
EC[b]	495 ± 53	427	220	220	290	323	427	580
Silt (%)	8.2 ± 0.4	8.4	7.3	7.3	9.3	7.4	8.4	8.9
Clay (%)	1.4 ± 0.1	1.4	1.2	1.2	1.6	1.2	1.4	1.6
Sand (%)	90.4 ± 0.4	90	89	89	92	90	90	91
HA (%)	0.5 ± 0.1	0.3	0.003	0.003	2.1	0.04	0.3	1.0
Pb (mg/kg)	1350 ± 110	1490	1690	579	1900	1120	1490	1690
Cd (mg/kg)	1.9 ± 0.2	2.0	1.0	0.2	3.7	1.0	2.0	2.4
Cu (mg/kg)	328 ± 52	234	143	143	800	204	234	481
n[c]	15	15	15	15	15	15	15	15

Table 6 Summary of motor park topsoil characteristics

Parameter	Mean ± SE[a]	Median	Mode	Minimum	Maximum	Percentile		
						25	50	75
pH	6.6 ± 0.2	6.5	6.8	5.2	8.2	6.2	6.5	6.8
EC[b] (µS/cm)	492 ± 59	394	820	258	820	280	394	715
Silt (%)	7.8 ± 0.5	7.5	6.5	6.5	9.2	7.0	7.5	8.8
Clay (%)	1.5 ± 0.4	1.5	1.4	1.4	1.6	1.4	1.5	1.6
Sand (%)	91 ± 1	91	91	89	92	90	91	92
HA(mg/kg)	1.6 ± 0.3	1.5	0.1	0.1	3.5	1.0	1.5	2.6
Pb (mg/kg)	1524 ± 88	1593	983	983	1983	1190	1593	1746
Cd (mg/kg)	1.9 ± 0.3	1.4	1.1	0.8	4.8	1.1	1.4	2.5
Cu (mg/kg)	184 ± 25	177	37	37	405	111	177	219
Sample population	15	15	15	15	15	15	15	15

[a] SE = Standard error, [b] EC = Electrical conductivity, [c] n = Sample population

3.2. Humic Acid Characteristics

Ash contents, E_4/E_6 ratios, total acidity, carboxyl and phenolic group contents are presented in Tables 8–10. Ash contents of the humic acids isolated from highway soils (HWS), auto-repair workshop soils (ARWS) and motor park soils (MPS) gave a similar mean value of 8.1 ± 0.3 (%). The UV-visible spectra of the humic acids extracted from the different soil samples were also similar. Figure 1 demonstrates the spectrum of highway soil humic acid, characterized with absorbance values that decrease with increasing wavelength. Slight shoulders at 205, 320 and 375nm at respective absorbance values of 2.634, 0.656 and 0.390 were obtained. This suggests the presence of absorbing chromophores (unsaturated and conjugated bonds) in the humic acid molecule fragment [17]. The E_4/E_6 optical ratio ranged from 4.4 ± 0.2 to 5.7 ± 0.2.

Table 7 Degree of contamination of soils by Pb, Cd and Cu using Netherlands reference values for uncontaminated soils

	Metal	Mean ± SE	Median	Mode	Minimum	maximum	Percentile		
							25	50	75
HWS	Cd	1.7 ± 0.2	1.7	1.1	0.8	4.8	1.1	1.7	2.0
	Cu	7.2 ± 0.6	7.7	2.0	2.0	15.6	3.9	7.7	9.1
	Pb	20.8 ± 2.5	14.7	3.1	3.1	65.9	11.1	14.7	28.3
ARWS	Cd	1.9 ± 0.2	2.0	1.0	0.2	3.7	1.0	2.0	2.4
	Cu	6.6 ± 1.0	4.7	2.9	2.9	16	4.1	4.7	9.6
	Pb	27.0 ± 2.2	29.8	23.8	11.6	38.0	22.4	30.0	33.8
MPS	Cd	1.9 ± 0.3	1.4	1.1	0.8	4.8	1.1	1.4	2.5
	Cu	3.7 ± 0.5	3.5	2.2	0.7	8.1	2.2	3.5	4.4
	Pb	30.5 ± 1.6	31.9	33.5	19.7	39.7	23.8	31.9	34.5

HWS = Highway soil, ARWS = Auto-repair workshop soil, MPS = Motor park soil, SE = Standard error of mean

Table 8 Characteristics of humic acids extracted from highway soil of southwest Nigeria

Parameter	Mean ± SE	Median	Mode	Minimum	Maximum	Percentile		
						25	50	75
Total acidity (meq/100g)	855 ± 57	750	750	550	1450	650	750	1025
Carboxyl group (meq/100g)	23 ± 1	21	20	17	39	19	21	26
Phenolic group (meq/100g)	832 ± 56	730	530	526	1430	632	730	1005
E_4/E_6 ratio	4.4 ± 0.1	4.3	4.3	402	465	4.3	4.4	4.5
Ash (%)	8.1 ± 0.3	7.8	7.7	7.6	9.4	7.7	7.8	8.6
Sample population	30	30	30	30	30	30	30	30

SE = Standard error of mean

Table 9 Characteristics of humic acids extracted from auto-repair soils of southwest Nigeria

Parameter	Mean± SE	Median	Mode	Minimum	Maximum	Percentile		
						25	50	75
Total acidity (meq/100g)	388 ± 6	400	400	352	410	354	400	410
Carboxyl group (meq/100g)	285 ± 7	290	320	240	320	250	290	320
Phenolic group (meq/100g)	103 ± 3	107	90	80	112	99	107	110
E_4/E_6 ratio	4.9 ± 0.1	4.9	5.0	4.8	4.9	4.9	4.9	5.0
Ash (%)	8.1 ± 0.3	7.8	7.6	7.6	9.4	7.7	7.8	8.6
n	15	15	15	15	15	15	15	15

SE = Standard error of mean, n = sample population

Table 10 Characteristics of humic acids extracted from motor park soils of southwest Nigeria

Parameter	Mean ± SE	Median	Mode	Minimum	Maximum	Percentile		
						25	50	75
Total acidity (meq/100g)	432 ± 5	428	420	420	475	420	428	432
Carboxyl group (meq/100g)	321 ± 2	320	320	310	340	315	320	330
Phenolic group (meq/100g)	111 ± 4	102	100	100	145	100	102	120
E_4/E_6 ratio	5.7 ± 0.1	5.7	5.4	5.43	5.97	5.5	5.6	5.7
Ash (%)	8.1 ± 0.3	7.8	7.6	7.6	9.4	7.7	7.8	8.6
n	15	15	15	15	15	15	15	15

SE = Standard error of mean, n = sample population

3.2.1 Infra-red Absorption Spectra

Infra-red spectroscopy (IR) gives information about the presence of various atomic and functional groups in a substance [17]. Major features of the IR spectra in Fig. 2 are a broad band at 3533 – 3416 cm^{-1} due to OH

stretching of phenols and alcohols. A hydrocarbon skeleton was identified at 2907 cm^{-1}. The band at 1631 cm^{-1} was attributed to an alkene conjugated to C=C or C=O. It could also be due to the carboxylate ion. The strong absorption at 1037 cm^{-1} represents C-O of alcohol and polysaccharide and the bands below 1000 cm^{-1} (915 cm^{-1} and 687 cm^{-1}) suggest the presence of substituted aromatics [17].
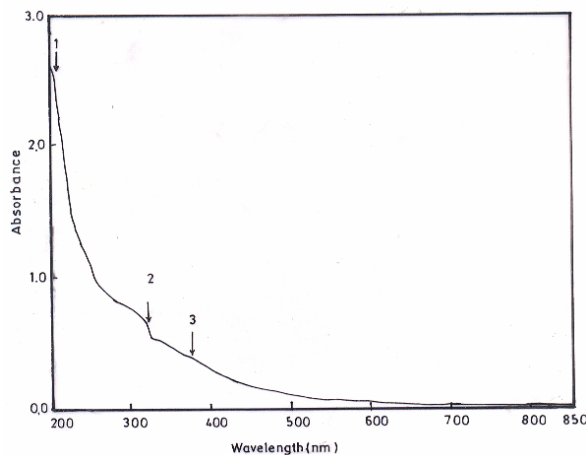

Figure 1 UV-visible absorption spectrum of humic acid extracted from the highway
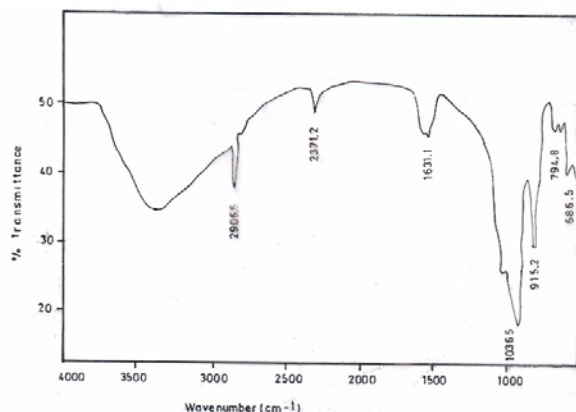
Figure 2 A typical infra-red spectrum of humic acid isolated from a highway topsoil, Nigeria.

The carbonyl absorptions of humic acids isolated from motor park soils and auto-repair workshops were observed at 1717 cm^{-1} and 1673 cm^{-1}; 1636 cm^{-1} due to C=O of ketone, aldehyde, quinone and carboxylate ion and the vibrations for aliphatic hydrocarbons were identified at 2935 cm^{-1} and 1483 cm^{-1} (HA-ARWS). The corresponding peaks for HA-MPS were detected at 2933 cm^{-1}, 2658 cm^{-1} and 1383 cm^{-1}. The bands at 1599 cm^{-1}, 882 cm^{-1} and 773 cm^{-1} confirm the

presence of substituted aromatics in HA-ARWS and the corresponding absorptions for HA-MPS were found at 912 cm^{-1}, 797 cm^{-1} and 755 cm^{-1}.

3.2.2 Reactive Groups

The acid group contents of the isolated humic acids, presented in Tables 8-10 vary from 381.2 ± 0.2 to 855.2 ± 3 meq/100g for total acidity, 230.7 ± 0.1 to 325.5 ± 0.1 meq/100g for carboxylic acid and 105.3 ± 0.1 to 833 ± 2 meq/100g for phenolic hydroxyl groups. From the mean values, humic acids from auto-repair workshops and motor park soils contained similar PhOH/COOH compositions of 27%:73% and 26%:74%, respectively. By contrast, humic acids extracted from the highway soils contained more phenolic (97%) than carboxylic acid (3%) of the total reactive groups.

3.3. Humic Acids and Soil Metal Concentration

3.3.1 Correlation between Humic Acid Chemical Characteristics and Soil Metal Level

Negative and positive trends were obtained for parametric correlation (Pearson) and non-parametric correlations (Spearman and Kendall). Significant coefficients (Table 11) were obtained (p ≤ 0.05) for Cd and acidity groups (total and phenolic groups) of humic acids isolated from highway soils with values ranging from + 0.37 to + 0.48. Positive associations were also obtained for Pb and acidity groups of HAs extracted from motor park topsoils, with the coefficients varying from + 0.16 to + 0.95 but only those between the metal and phenolic groups were significant at p ≤ 0.05. Significant correlations (both negative and positive) were also found for the metal concentrations (especially Cu) and E$_4$/E$_6$ at the 0.01 and 0.05 levels (Table 11).

3.3.2 Trace Metal Binding of the Extracted Highway Soil Humic Acid

Results show that the metal concentrations bound by the humic acids were 0.779 mmoles/g HA of Cu, 0.911 mmoles/g HA of Pb and 0.237 mmoles/g HA of Cd. The humic acid gave higher binding for Pb^{2+} than for the other two metals, as presented in Table 12. The metal binding capacity of the humic acid was thus in the order Pb^{2+} > Cu^{2+} > Cd^{2+}, which agrees with the work of Stevenson [18].

The desorbed metal concentrations obtained from leaching tests (Table 13), indicate that metal leaching from the HA-metal complex is pH dependent. The

percentage of leached metal with respect to the bound metals ranged from 1.58 x 10^{-4} to 1.91 % (Table 14), suggesting reduced solubility of the complexes. This is consistent with literature reports that humic acids form relatively water insoluble complexes with heavy metals [9].

Table 11 Correlation coefficients for soil metal levels and humic acid chemical characteristics

HA characteristics at the sites	Lead			Copper			Cadmium		
	Pearson	Spearman	Kendall	Pearson	Spearman	Kendall	Pearson	Spearman	Kendall
Highways									
Total group	+0.12	+0.15	+0.11	+0.07	+0.04	+0.02	+0.46*	+0.48*	+0.40*
Carboxyl	-0.10	-0.12	-0.06	+0.01	+0.21	+0.18	+0.14	+0.06	-0.01
Phenolic	0.13	+0.13	+0.07	+0.01	-0.02	-0.02	+0.46*	+0.47*	+0.37*
E_4/E_6 ratio	+0.44	-0.28	+0.26	-0.05	-0.14	-0.09	+0.53*	+0.30	+0.25
Motor parks									
Total group	+0.64	+0.80	+0.67	-0.11	+0.40	+0.33	-0.39	-0.20	0.00
Carboxyl	+0.16	+0.45	+0.41	+0.64	+0.45	+0.41	0.28	-0.00	0.00
Phenolic	+0.95*	+0.95*	+0.91*	-0.39	-0.11	-0.18	-0.39	-0.63	-0.55
E_4/E_6 ratio	-0.39	-0.20	0.00	-0.70	-1.00**	-1.00**	-0.87	-0.80	-0.67
Auto-repairs									
Total group	+0.20	+0.30	+0.20	-0.65	-0.50	-0.40	-0.68	-0.60	-0.40
Carboxyl	+0.16	+0.30	+0.20	-0.68	-0.50	-0.40	-0.54	-0.60	-0.40
Phenolic	+0.03	-0.10	0.00	+0.51	-0.30	-0.20	0.09	-0.20	-0.20
E_4/E_6 ratio	+0.25	+0.03	+0.20	+0.89*	+0.90*	0.80*	+0.03	+0.40	0.40

**Correlation is significant at the 0.01 level (2-tailed), *Correlation is significant at 0.05 level (2-tailed).

Table 12 Amount of metals bound by humic acids extracted from highway soil

Process	Copper (µmoles)	Lead (µmoles)	Cadmium (µmoles)
Before complexation	2.3 ± 0.1	ND	ND
After complexation	781.3 ± 0.1	911 ± 2	237 ± 1
Bound metal	779	911	237

n = 15 for each mean value

Table 13 Amount of metal leached from the humic - metal complex

Water pH	Pb (µmoles)	Cd (µmoles)	Cu (µmoles)
1.95	1.53 ± 0.01	0.033 ± 0.01	14.87 ± 0.03
4.32	0.004 ± 0.001	0.023 ± 0.001	0.006 ± 0.001
5.54	0.0014 ± 0.001	0.011 ± 0.001	0.003 ± 0.001
n	15	15	15

n = sample population

Table 14 Metal leached from the humic acid- metal complexes at room temperature

Water pH	Pb (%)	Cd (%)	Cu (%)
1.95	1.68 x 10^{-1}	1.39 x 10^{-2}	1.91
4.32	4.20 x 10^{-4}	9.70 x 10^{-3}	7.70 x 10^{-4}
5.54	1.58 x 10^{-4}	4.46 x 10^{-3}	3.80 x 10^{-4}

4. DISCUSSION

4.1. Soil Metal Level

The relative abundance of the metals in topsoils of motor parks, auto-repair shops and highway soils was in the order Pb > Cu > Cd, which agrees with previous work [18]. The geology of the study areas, especially Ibadan, has low enrichment of Pb and minimal weathering potentials, being mainly Precambrian branded gneiss of granitic, migmatitic quartzite [8]. The high concentrations of Pb obtained at all sites suggest that the source of lead in the environment is essentially anthropogenic. Thus, Pb is derived mostly from exhausts of vehicles, which in Nigeria still run on gasoline of Pb concentrations averaging 0.25 to 0.66 g per liter [19, 20]. Maduka [20] reported national consumption of 25 million liters per day and a 75% emission rate of Pb particulate and an estimated 2800 metric tons of vehicular gaseous Pb emission are deposited in urban areas of Nigeria annually as reported by Onianwa et al. [8].

Metal concentrations in this study (Cd (1.7 ± 0.2 to 1.9 ± 0.2 mg/kg), Cu (184 ± 25 to 360 ± 310 mg/kg) and Pb (1039 ± 123 to 1524 ± 88 mg/kg)) were in agreement with previous reports on trace metal concentrations found in topsoils in the vicinities of auto-repair workshops, gas stations and motor parks in Ibadan, which ranged from 21 to 5070 mg/kg for Pb, 15 to 2150 mg/kg for Cu and 0.09 to 4.10 mg/kg for Cd [8]. In Osogbo city, southwest Nigeria, the metal levels found in auto-repair workshops were 729.5 ± 110.9 mg/lkg (Pb), 4.58 ± 1.01 mg/kg (Cd), 116.6 ± 26.8 mg/kg (Cu) and in motor parks, they were obtained as 519.0 ± 73.0 mg/kg (Pb), 3.6 ± 0.8 mg/kg (Cd) and 37.9 ± 12.5 mg/kg (Cu) [21]. Li [22] reported that although lead emissions have dropped drastically with the phase-out of tetraethyl-lead as a fuel additive in Canada, accumulations up to 1628 mg/kg soil were found in highway topsoils.

The highest value of lead (3294 mg/kg) for highway soils in this study was found in Lagos. This could be attributed to high vehicular traffic volume of greater than 1840 ± 132 per hour in Lagos roads used by up to 15 million people. Abeokuta recorded reduced Pb contamination since only one site had a metal level exceeding 1000 mg/kg (Table 1). The vehicular traffic volume per hour in Abeokuta, inhabited by less than 20% of Lagos population, is given as 1509 ± 75 to 1752 ± 344, 1518 ± 109 to 1735 ± 167 and 1509 ± 159 to 1840 ± 132 vehicles per hour for the morning (8-9am), afternoon (12-1pm) and evening (5-6pm), respectively [2]. Previous reports stated that the levels of metals at the high density roads were significantly higher than the corresponding levels of medium and low density roads [21].

According to the UK Department of Environment Interdepartmental Committee on the Redevelopment of Contaminated Land (DOE-ICRCL) as found in Alloway [16], values for uncontaminated soils are given as 0 – 1 mg/kg for Cd, 0 – 100 mg/kg for Cu and 0 – 500 mg/kg for Pb. Soils containing 1 - 3 mg Cd/kg, 100 – 200 mg Cu/kg and 500 – 1000 mg Pb/kg are slightly contaminated. Areas referred to as contaminated soils usually have 3 - 10 mg Cd/kg, 200 - 500 mg Cu/kg and 1000 - 2000 mg Pb/kg. Heavy contamination is recorded when the soil contains 10 - 50 mg Cd/kg, 500 - 2500 mg Cu/kg and 2000 - 10,000 mg Pb/kg.

As a consequence of these values, our study sites could be classified as being slightly contaminated with Cd. Some of the sampling points gave no cadmium contamination with values less than 1.0 mg/kg. Only 6.6% (2 out of 30 sampling points) of highway soils are contaminated with Cd and about 20% (3 out of 15 sampling points) were recorded for motor park soils (Tables 1 and 3). A great majority of the sampling points were contaminated by Cu and Pb. This agrees with the reports of Onianwa et al. [8] for such areas in Nigeria, stating that Cu and Pb were the significant pollutants found.

The study sites are designed as open spaces in the country. Trigger concentrations for an open space for heavy metals are 1500 mg Pb/kg, 15 mg Cd/kg and 140 mg Cu/kg [16]. About 37% of the 60 sampling points exceeded the Pb trigger concentration, 87% exceeded the value for Cu and none exceeded that of Cd.

Roadsides, auto-repair workshops and motor parks in Nigeria are characterized by improper disposal of liquid and solid wastes. The metals are released during combustion and spillages of petrol and lubricants. Tyres and scraps are usually incinerated at these sites, various automobile oils (waste engine oil, transmission oil and brake fluids), which contain these metals as additives, are thrown onto the soils by the vulcanizers, panel-beaters, servicemen, auto-electricians, unskilled drivers and mechanics who work at the study sites. These processes contribute to the elevated Cd and Cu concentrations at the sites. Contaminated soils are easy sources of transmission of these metals through run-offs and the food chain to man. Thus, a considerable potential hazard exists for humans as these metals could be leached by urban run-offs and transported to nearby groundwater [16].

4.2. Soil Humic Acid Characteristics and Metal Complexation

Titration, infra-red and UV-visible spectroscopies

provide evidence and are great contributions to our knowledge of the chemical structure of humic acid. They suggest the presence of multiple bonds such as alkene, alkene with extended conjugation or conjugated to C=O and aromatic structures in the HA molecule. The optical parameter E_4/E_6 ratio expresses molecular condensation. A value $\geq$ 5.0 indicates a reduced hydrophobic structure while that $\leq$ 5.0 suggests an increased hydrophobic structure [9, 13]. E_4/E_6 ratios in the range of 3.8 to 6.4 reported in the literature suggest that the values obtained in this study $(4.4 \pm 0.2$ to $5.7 \pm 0.2)$ are characteristic of humic acid and that the molecules are not entirely hydrophilic. The IR spectra agree with previous studies [9, 13, 23-28].

Hydrophobic skeletons such as phenols are capable of influencing metal sorption in soil. The major ligand sites for a metal ion in humic acids are the oxygen-containing functional groups. These nucleophilic sites as quantified by titration (23 ± 1 to 321 ± 2, 114 to 832 ± 56 and 388 ± 6 to 855 ± 57) meq/100 g for RCOOH, PhOH and total acidity are similar to those obtained for some humic acids from Nigerian soils containing 340 meq/100 g COOH, 560 meq/100 g PhOH and 900 meq/100 g total acidity as reported by Eneje and Mbagwu [27].

These reactive groups are known to produce chemical changes in a wide variety of metals. The functional groups enable the humic acids to form water insoluble complexes with heavy metals, resulting in their accumulation in soil. Humic substances (HSs) are the most widespread natural polymers on earth. One of the main functions of HSs in the biosphere is the re-distribution of metals and organic matter in soils and groundwater. Metal ions tend to sequester humic substances, restricting their migration to neighboring media [9].

The complexes are usually formed by the exchange reaction between the hydrogen of the acidic functional groups and the metal cations. The chemical reaction involves simultaneous participation of the adjacent carboxyl and phenolic groups in the humic acid to form either a 1:1 or 2:1 chelate complex [18, 29-36]. The presence of these oxygen-containing functional groups revealed by infra-red absorptions is consistent with those found in humic acids from Nigeria as reported by Adekunle and Onianwa [25] and outside the country [26-28].

The relative stabilities of humic acid–metal complexes have been reported as Cu > Pb > Cd [35] or Pb > Cu > Cd [18]. The binding capacities of the humic acids in this study were obtained as 0.78 mmoles Cu^{2+}/g and 0.91 mmoles Pb^{2+}/g, showing that more Pb was bound compared to copper. This is consistent with the work of Logan et al. [32], which gave the maximum binding capacities as 0.564 mmoles Pb^{2+}/g and 0.188 mmoles Cu^{2+}/g for humic acid extracted from peat. The desorption of the three metals from the complexes using water, which was less than 2.0%, confirms the stability of humic acid-metal complexes in water [9].

Spearman, Pearson and Kendall correlations performed on the data showed that there was no defined pattern in the associations between soil metal concentration and humic acid characteristics, consistent with reports that humic acids tend to form complexes that are different for each metal and also depend on soil conditions such as pH, cation exchange capacity and clay mineral fraction [24]. Organic matter not only forms complexes with these metals but it also retains them in exchangeable forms, affecting each metal differently. Some metals are bound and rendered unavailable while others are bound and readily available [9, 13]. This implies that the readily available metals are easily transported or dispersed, explaining the negative correlations obtained in the study. Barancikova and Makovnikova [24] also obtained negative and positive Spearman correlation coefficients for heavy metal concentrations and humic acid chemical characteristics (acidity groups and E_4/E_6 ratios) which varied with metal type.

5. CONCLUSIONS

The topsoils obtained from the highways, auto-repair workshops and motor parks located in Lagos, Ibadan and Abeokuta cities of southwest Nigeria contained elevated concentrations of Cd, Cu and Pb. Their relative abundance in each site decreased in the order Pb > Cu > Cd. About 32% and 87% of the sampling points exceeded the trigger concentrations of 1500 mg Pb/kg and 140 mg Cu/kg, respectively. The chemical characteristics of the humic acids isolated from the sites are consistent with values obtained in Nigeria and elsewhere.

Spearman, Pearson and Kendall correlation coefficients showed some significant negative and positive associations ($p < 0.05$) between the humic acid chemical characteristics and the soil metal concentrations. The isolated humic acids bound to metals in the order of Pb > Cu > Cd. Batch desorption tests with water (adjusted to varying pH values) indicate low likelihood of significant leaching, implying that under ideal conditions metals complexed to the humic acids have reduced mobility. In conclusion, the humic acids from the study sites exert some influence on the sorption of Pb, Cd and Cu in soils.

6. REFERENCES

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