



## Heavy Metal Content and Physico-Chemical Analyses of Soils under the Litter of some Medicinal Taxa in the Luki Biosphere Reserve, Democratic Republic of the Congo

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### Authors' contributions

This work was carried out in collaboration among all authors. Authors FBM, TMF, PTM and NKN designed the study, performed the statistical analysis and wrote the protocol. Authors FBM, PBI, PKB, MMN, KNN and NKK wrote the first draft of the manuscript. Authors FBM, KNM, JBM, PKM, PMN and MNM managed the analyses of the study. Authors FBM, PKM, JBM, MMN, KNM and KNN managed the literature searches. All authors read and approved the final manuscript.

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## ABSTRACT

**Aim:** The aim of this study was to compare the texture, mineral element and heavy metal content of the soil under the litter of different selected species in relation to the plan of their pedogenesis.

**Methods:** The granulometric analysis of the various samples based on laser diffraction, measurements of Total Nitrogen and Total Organic Carbon were performed using the Elemental Analyzer, the Organic Matter content is estimated by the loss of ignition method using a Salvis furnace, In order to estimate the  $\text{CaCO}_3$  content. The same samples were heated in the oven at  $1000^\circ\text{C}$  for 60 minutes and reweighed. The carbonate content is estimated by the loss in mass during this second firing, multiplied by 2.274 which is the molecular weight ratio between  $\text{CaCO}_3$  and  $\text{CO}_2$ . The heavy metal and mineral composition was evaluated by ICP-AES and AAS.

**Results:** The granulometric analyses show a sandy texture, according to the FAO classification. The organic matter content thus observed in the soils under the litter of these four species did not show any significant difference. The concentration of major mineral elements recorded in soils under the litter of *Blighia welwitschii*, *Oncoba welwitschii*, *Zanthoxylum gillettii* and *Harungana madagascariensis* did not show any significant difference. However, potassium, phosphorus, and magnesium have a high concentration compared to calcium and sodium. This study showed that the levels of cadmium, lead, arsenic and aluminum in the various samples of litter soils of all plant species including nickel for *B. welwitschii* are above the standards set by the Canadian Council of Ministers of the Environment for soil quality. Pollution factors for aluminum are the highest. This indicates that aluminum is the most polluting metal. The low pollution factors for lead and nickel are due to the low fixation of these metals by soils.

**Conclusions:** Our study took place in the Luki Biosphere reserve and showed that the different soils under litter are polluted by very toxic and persistent heavy metals such as cadmium, lead, aluminum, arsenic and nickel. This study has revealed new aspects of heavy metal pollution. This pollution represents a serious threat to the environment in general and to humans in particular through the food chain.

**Keywords:** Litter; ICP-AES; SAA; luki; *blighia welwitschii*; *zanthoxylum gillettii*; *harungana madagascariensis*; *oncoba welwitschii*.

## 1. INTRODUCTION

Soil is one of the main components of tropical natural resources, and has been considered for several decades as the environmental interface of communities [1]. However, as a basic ecosystem, its typological diversity in the humid tropics and the complexity of the processes it supports have so far posed problems for ecologists in uncovering problems related to the availability of mineral elements for the maintenance of the physico-chemical quality of soils [2].

The overexploitation of natural resources by man, due to high population growth, has leapt forward in recent years, leading to enormous changes in ecosystems, which also have repercussions on climate, biodiversity and land use [3], thus causing pollution that progressively

and worryingly degrades the environment. With increasing industrialization, more and more heavy metals are found in the environment. Heavy metals are trace metal elements that are highly toxic and persistent in the environment [4,5].

Work on heavy metals has begun to attract the attention of scientists because of their non-biodegradability, toxicity, persistence, and their presence in food chain [6]. Heavy metals are among the widespread pollutants resulting from anthropogenic activities [7] and given the fact that they do not degrade, can migrate and accumulate in different components of natural ecosystems and eventually be incorporated into the food chain thus endangering the health of populations [8,9].

Plants grown on soil contaminated by heavy metals absorb and concentrate them through the roots and through the air. The sap then distributes them to other plant organs. Thus, heavy metals are finally found in crops and more precisely in the products harvested and consumed by humans [10].

Food thus contributes to the increase of heavy metals in the body [11]. Some metals (Zn, Fe, Cu, Cr, Co, Se) are trace elements, therefore essential for humans, but can become toxic in high concentrations. Others (Pb, Cd, Al, As, Ni), on the other hand, are toxic even at low doses and have no known metabolic function in the human body. Previous studies [12-18] have shown that exposure to high levels of zinc can cause dwarfism in children, gastrointestinal and respiratory disorders, hematological changes, liver, pancreas and kidney damage. The iron toxicity can cause digestive disorders, sometimes severe; ingestion of high doses of chromium causes severe liver and kidney disorders, abdominal pain; the ingestion of high doses of copper can cause Wilson's disease and gastrointestinal problems. The nickel toxicity can cause different types of cancer in different parts of the body while lead causes cardiovascular disease, neurological disorders, kidney damage, lead poisoning in children and cadmium causes kidney problems and increased blood pressure. Chronic exposure to cadmium leads to the development of irreversible kidney disease that can progress to kidney failure.

In the Democratic Republic of the Congo, previous studies have shown that several plant especially the medicinal species used in Lubumbashi contain heavy metals that are a danger to the environment [19-22].

Accumulating plants are generally native to a particular soil or bedrock type and can be valuable geobotanical indicators for mineral deposits. Another potential use of these accumulator plants is as an indication of anthropogenic pollution, as these plants are often among the few species capable of colonizing soils contaminated with high concentrations of heavy metals, and can thus provide insight into the extent of pollution. As a result, several detailed studies have been carried out on the phytochemistry of plants that accumulate heavy metals [23-28].

Several authors have reported various biochemical changes in green plants in response

to Pb: decrease in chlorophyll content [29-32], carotenoids [32], proteins [33], nitrate reductase activity [33-34], lamella and mesophyll thickness, epidermal cell size and vessel diameter [35] increase in chlorophyll a/b ratio [32] and phenol concentration [36-38]. In the presence of other metals, such as Al, Cu and Cd, increased lipid peroxidation has been observed [39-45]. Thus, in metal-contaminated plants, we expect to find symptoms of senescence, such as lower concentrations of chlorophyll, carotenoids and proteins, decreased nitrate reductase activity and increased acid phosphatase activity, lipid peroxidation and a/b ratio of chlorophyll, compared to non-metal stressed plants. Phenol detoxification is also expected.

The behavior of heavy metals in soils depends on several factors that affect their dynamics, i.e. pH, cation exchange capacity, clay content, organic matter, hence their solubilization and availability to plants [46-47]. Soil pollution by heavy metals is considered a long-term threat to the environment [48].

Overall, the present study, which is part of the "soil-plant" relations in the forest environment, aims at knowing the state of mineral and organic elements of the soils under litter of 4 plant species namely *Blighia welwitschii*, *Oncoba welwitschii*, *Zanthoxylum gilletii* and *Harungana madagascariensis*, which are used in the traditional pharmacopoeia of the Kongo Central province (precisely in the Luki Biosphere Reserve). Specific objectives were: (i) to compare the texture and mineral element content of the soil under the litter of different selected species in relation to the plan of their pedogenesis, (ii) to compare the content of heavy metals in littered soils and different parts of selected plants and (iii) to test whether there is a significant difference between physico-chemical properties, heavy metal and minerals content of the soils under the litter of these forest species and compare the results to the standards set by the Canadian Council of Ministers of the Environment (CCME) and the World Health Organization (WHO).

## 2. MATERIALS AND METHODS

### 2.1 Study Material

The study material consists of soils under the litter of *B. welwitschii* (Hiern) Radlk. (*Sapindaceae*), *O. welwitschii* (Oliv.) Gilgn. (*Flacourtiaceae*), *Z. gilletii* (De wild) P.G.

(*Rutaceae*). Waterman and *H. madagascariensis* Lam. ex Poir. (Hypericaceae) collected in November 2018, in the Luki Biosphere Reserve in the Bas-fleuve Territory, Kongo Central Province, DRC (Fig. 1). The plants were identified at the Herbarium of the Institut National d'Etudes et de Recherches Agronomiques (INERA/LUKI) and confirmed by the Laboratory of Systematic Botany and Plant Ecology, Department of Biology, Faculty of Sciences, University of Kinshasa.

## 2.2 Methods

The soil study consists of a series of successive operations carried out in the field and in the laboratory. In the field, the work was carried out in several stages and consisted in the localization of different plants and the analysis of different sites using a topographic 3D map of Kongo Central Province at 1/50000, the opening of pits, the description of profiles and finally sample collection. In the laboratory, samples were dried in an oven at 50°C followed by physico-chemical and mineral analyses. These analyses were carried out simultaneously at the

Laboratory of the Institut des Sciences de l'Environnement/Institut Forel (University of Geneva, Switzerland), the Mineral Analysis Laboratory of the Office Congolais de Contrôle/Lubumbashi (LAMI/OCC) and the Soil Analysis Laboratory of the Centre Régional d'Etudes Nucléaires de Kinshasa (CREN-K).

### 2.2.1 Sample collection

In the five randomly selected plots under litter of plants, samples were taken with an auger at 50 cm. During sampling, 2 to 6 soil cores were reconstituted in half a plastic tube. The samples were wrapped in plastic film. The cores were divided into continuous samples every 5 cm. A portion of each sample was placed in a plastic jar to be dried and finely crushed. A second portion was placed in a closed bag for sieve size analysis.

Each sample was then homogenized in a bucket, wrapped in plastic film and labelled. At the end of the sampling, these plastic films were closed and sent to the different soil analysis laboratories.

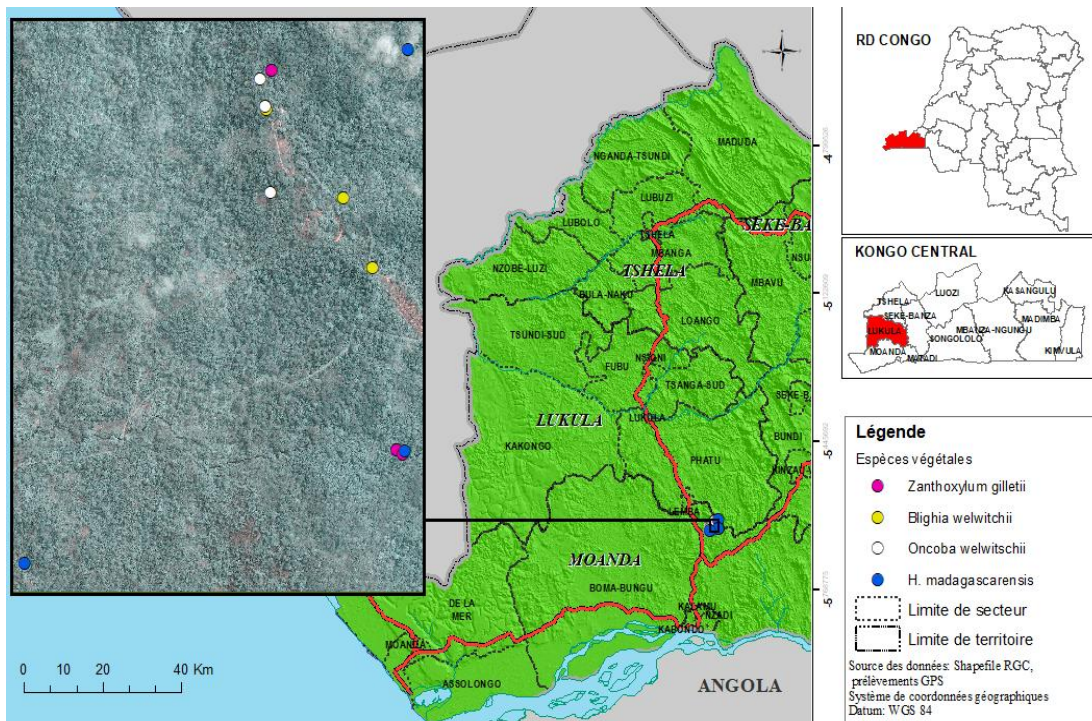


Fig. 1. Soil sampling sites in soils under the litter of the plants under study

## 2.2.2 Physical analysis

### 2.2.2.1 Granulometric analysis

The granulometric analysis of the various samples was performed at the Forel Institute using a Coulter<sup>®</sup>LS100 laser diffractometer (Beckman Coulter, Fullerton, USA). This method, based on laser diffraction, establishes the granulometric spectrum of the sample particles of which size ranges from 0.4 µm to 900 µm, which was followed by dispersion during 5 minutes by sonication in deionized water [48].

### 2.2.2.2 Chemical analyses

#### (i) pH-water

Mix 10 g of sieved soil in 25 ml of distilled water under stirring for 16 h and take the pH with a glass electrode.

#### (ii) Water content

Empty containers were weighed and numbered. Moist samples were placed in these containers, weighed, placed in an oven at 60°C for 48 hours and then weighed again. The water content of each sample was deduced from the formula:  

$$T_{\text{water}} = \frac{(Ph - Ps)}{(Ps \cdot 100\%)}$$

Where  $T_{\text{water}}$  is the water content of the sample, Ph the weight of the wet sample and Ps the weight of the dry sample.

#### (iii) Organic matter (OM) and carbonate (CaCO<sub>3</sub>) contents

The organic matter content is estimated by the loss of ignition method [Loss Of Ignition, LOI] using a Salvis furnace (AG Emmenbrücke, Lucerne, Switzerland), according to the procedure described in detail by [40]. Briefly, 1 to 5 g of dry, crushed and sieved soil is weighed, heated at 550°C for 60 min and then weighed again. The organic matter content of each sample is estimated by mass loss. In order to estimate the carbonate content, samples were fired in the oven at 1000°C for 60 minutes and reweighed. The carbonate content is estimated by the loss in mass during this second firing, multiplied by 2.274, which is the molecular weight ratio between CaCO<sub>3</sub> and CO<sub>2</sub>.

The calcium carbonate content of the first sample is measured by acid etching (HCl 1M) of the dry soil and crushed in carbonate bombs. The

carbonate content of each sample is measured by the bomb from the pressure exerted by the CO<sub>2</sub> from the vaporized calcium carbonates according to the following reaction :  $2\text{HCl} + \text{CaCO}_3 \rightarrow \text{CaCl}_2 + \text{H}_2\text{CO}_3 \rightarrow \text{H}_2\text{O} + \text{CO}_2 + \text{CaCl}_2$  [40].

#### (iv) Measurement of Total Nitrogen (TN) and Total Organic Carbon (TOC)

Measurements of Total Nitrogen (TN) and total organic carbon (TOC) were performed using a PE 2400 Series II CHNS/O Elemental Analyzer (Perkin Elmer, USA). For each sample, two capsules containing between 3 and 8 g of crushed dry soil were prepared. One of the two capsules was acidified by adding 50 µl of H<sub>3</sub>PO<sub>4</sub> (5%). The capsules were placed in the PE® 2400 Series II CHNS/O Elemental Analyzer to undergo complete combustion at 975 °C and then reduced to CO<sub>2</sub>, H<sub>2</sub>O, N<sub>2</sub> and SO<sub>2</sub> at 500°C. TN measurements were performed on non-acidified capsules and TOC measurements were performed on acidified capsules.

## 2.2.3 Mineral analysis by ICP-AES and AAS of the soils

The determination of minerals content was carried out using water and nitric acid method while the analysis was by Inductively Coupled Argon Plasma Optical Emission Spectrometry (ICP-AES) and Atomic Absorption Spectrometry (AAS) [49-50]. Then 0.3 g of soil under litter of each plant was diluted in 5mL of distilled water while the mixture was placed in PM60 Teflon bombs (Analytikjena 40Bar), then heated at 60°C, and 10 mL of nitric acid (65% HNO<sub>3</sub>) (Merck) were added. The mixture was allowed to react for 30 minutes at room temperature in the bombs that were covered with caps and then stripped with HNO<sub>3</sub>/H<sub>2</sub>O (v/v, 1:1). The bombs were then placed in the high frequency microwave mineralizer (Analytikjena AG TOPwave: 2.5 Ghz, Germany) controlled by microcomputer by choosing the Vegetable Leave mode as a digestion mode at 180°C, 50bar for 1 hour. At the end of mixing, the digester stopped by letting the bombs rest for 3 hours until completely cooled. The cooled analyte is thus carefully transferred from bombs by filtration on Whatman filter paper, to 50 mL volumetric flasks previously stripped. The initial volume was diluted to 50 mL with distilled water and 13 mL of analyte were placed in 15 mL conical flask previously stripped for reading by Inductively Coupled Argon-Plasma Atomic Emission

Spectrometry (ICP-AES) (Optima 8300 Perkin Elmer, USA). The analysis was performed in triplicate.

The content of these different heavy metals are studied (Pb, Al, As, Ni, Cd) and minerals (P, Ca, Mg, Na, Co, K, Zn, Mn, Se, Cr) by Induction Coupled Plasma Atomic Emission Spectroscopy (ICP-AES) and also by Atomic Absorption Spectroscopy (AAS) the Fe and Cu.

An aliquot part of the solution (Analyte) is sucked into the instrument (AAS/ICP-AES) for the determination of metals/minerals, namely P, Ca, Mg, Na, Co, K, Fe, Zn, Mn, Cu, Se, Pb, Al, As, Ni, Cd and Cr.

The calibration of the ICP-AES was performed using the standard prepared from the commercially available standard multi-element solution 3 at two points (1mg/L and 2.5mg/L, Perkin Elmer, USA). The most appropriate wavelength, gaseous argon flow, plasma stabilization and other ICP-AES instrument parameters for minerals were selected and measurements were made in the linear range of the standards used for calibration.

Working conditions were: Instrument: ICP-AES (Optima 8300 Perkin Elmer, USA); Power of Rf: 1500Watt; Plasma gas flow (Ar): 8 L/min; Nebulizer: 0.70 L/min; Auxiliary gas flow (Ar): 0.2 L/min; Viewing size: 5-22 mm; Copy and playback time: 1-5s (maximum 45s); Flow time: 1s (maximum 10s); View: Radial.

Calibration of the AAS was performed using a working standard prepared from commercially available metal/mineral standard solutions (1 mg/L, Perkin Elmer, USA). The wavelength, weakened cathode lamp current, gas mixture flow rate, gap width and other parameters of the AAS for metals/minerals were selected as specified in the instrument manual, and a background correction was used in the determination of metals/minerals. Measurements were made within the linear range of working standards used for calibration [49-50].

Working conditions were: Instrument: AAS (Analyst 400 Perkin Elmer, USA); Flame temperature: 2800°C, Acetylene pressure: 0.9-1.0bar, Atmospheric pressure: 4.5-5 bar, Playback time: 1-10 seconds (maximum 60 seconds), Flow time: 3-4 seconds (maximum 10 seconds).

The concentrations of all minerals were expressed in mg/kg dry weight of the sample. Each value is the mean of the determination in three replicates  $\pm$  standard deviation.

#### 2.2.4 Statistical analysis

Means and standard deviations were calculated using Excel. The comparison of the means of the granulometric analysis, the physico-chemical, the mineral and heavy metal parameters was carried out by an analysis of variance (ANOVA) using the STATISTIX software. The level of significance retained is 5%.

### 3. RESULTS

#### 3.1 Physico-chemical Analyses

##### 3.1.1 Granulometric analysis

The granulometric analysis of soils under the litter of the identified plant species are presented in Table 1.

The elements in this table showed that under these plant species, the soils have a sandy structure. The recorded clay contents showed very low values of this soil component under these plant species. The average values of these grades are shown in Fig. 2.

The minerals in this figure showed that the soils underneath the litter of each plant species under study have evolved so much that they are in a very advanced stage of alteration. With at least 90% sand and 19% clay, these soils are in the category of sandy-clay soils according to the FAO classification.

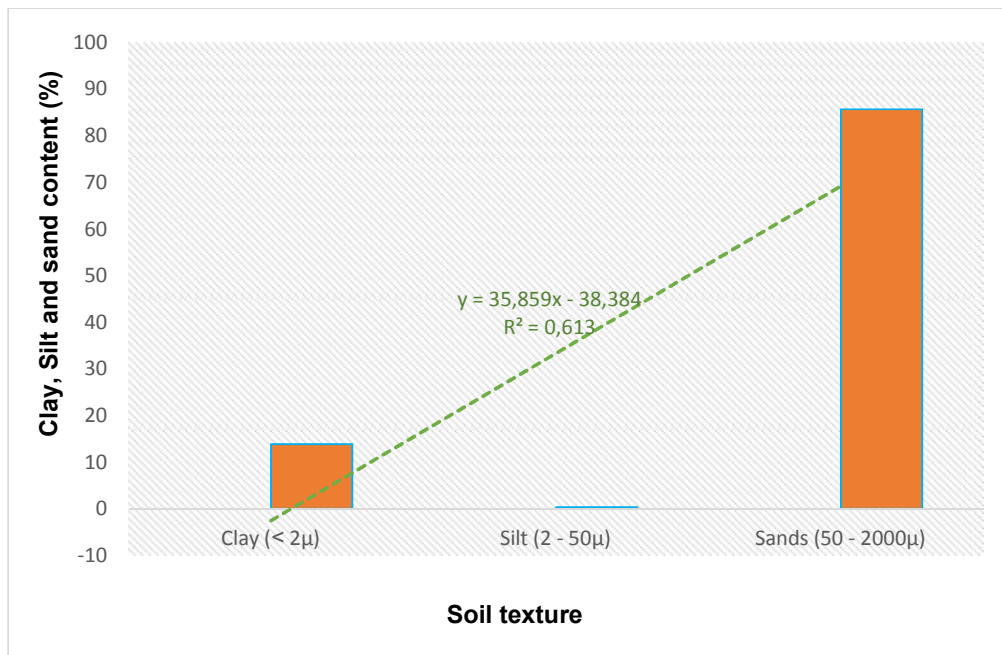
##### 3.1.2 Chemical analysis

The pH-water, Water content, Organic matter (OM) and Carbonate ( $\text{CaCO}_3$ ) contents and measurement of Total Nitrogen (TN) and Total organic Carbon (TOC) composition of the soils and the litter of its plant species are shown in the table.

The elements listed in this table showed that the pH of the soil under the litter of these species is acidic. It varied from pH 4.9 for soils under the litter of *H. madagascariensis* to pH 5.7 for soils under the litter of *B. welwitschii* and *Z. gillettii*.

**Table 1. Particle size analysis of sub-bedded soils**

N°	Plant species	Percentage by volume		
		Clay (< 2µ)	Silt (2 - 50µ)	Sands (50 - 2000µ)
1.	<i>B. welwitschii</i>	15.00 ± 0.06b	0.28 ± 0.09c	84.72 ± 3.96
2.	<i>O. welwitschii</i>	11.00 ± 0.10a	0.58 ± 0.08b	88.42 ± 2.35
3.	<i>Z. gilletii</i>	13.30 ± 0.05b	0.25 ± 0.09c	86.45 ± 5.929
4.	<i>H. madagascariensis</i>	16.40 ± 0.18b	0.62 ± 0.04a	82.98 ± 3.714



**Fig. 2. Evolution of the average clay, silt and sand contents in soils under bedding**

For moisture content, the soils under the litter of *O. welwitschii* have a moisture content of 45.63% slightly higher than the litter soils under other plant species. This was followed by soils under *Z. gilletii* (44.99%), *H. madagascariensis* (39.21%) and *B. welwitschii* (35.37%).

The organic matter (OM) content evolves in almost the same way in all these soils. A particularity for the soils under the litter of *Z. gilletii* (25.27% of O.M.), *H. madagascariensis* (22.56% of O.M.) and *O. welwitschii* (21.74% of O.M.).

Calcium carbonate also evolves in almost the same way in these soils. It varies from 0.21% in soils under the litter of *Z. gilletii* to 0.27% in soils under the litter of *O. welwitschii*.

**3.1.3 Mineral analysis of soils under the litter by AAS and ICP-AES**

**3.1.3.1 Concentration of major minerals (mg/kg) on soils under litter**

The concentration of major soil elements of each plant species under study are presented in the figures.

The elements reported in Figs. 3 and 4 show that in soils under *B. welwitschii* litter, phosphorus has a concentration of 49.5 mg/kg, followed by potassium (41.5 mg/kg), sodium (40.4 mg/kg), magnesium and calcium each have a concentration of 33.58 mg/kg and 31.49 mg/kg respectively. While for soils under *O. welwitschii* litter, the concentration of magnesium is 58.03 mg/kg, followed by potassium (56.88 mg/kg), and phosphorus (53.37 mg/kg); finally calcium (38.42 mg/kg) and sodium (24.23 mg/kg).

**Table 2. Chemical composition of soils under litter**

Plant species	pH	TH <sub>2</sub> O (%)	OM (%)	CaCO <sub>3</sub> (ppm)	TOC (%)	TN (%)	C/N (%)
<i>Blighia welwitschii</i>	5.7 0± 0.40	35.37 ± 5.87	15.93 ± 0,78	0.21 ± 0.03	0.07 ± 0.001	0.28 ± 0.02	0.25
<i>Oncoba welwitschii</i>	5.30± 0.56	45.63 ± 3.59	21.74 ± 0.86	0.27 ± 0.07	0.97 ± 0.03	0.16 ± 0.02	6.06
<i>Zanthoxylum gillettii</i>	5.70± 0.32	44.99 ± 4.26	25.27 ± 2.16	0.21 ± 0.10	1.21 ± 0.03	0.09 ± 0.001	13.50
<i>H. madagascariensis</i>	4.90 ± 0.20	39.21 ± 4.40	22.56 ± 0.87	0.24 ± 0.03	0.32 ± 0.32	0.13 ± 0.08	2.46

**Table 3. Concentration of heavy metals on soils under litter of all plants**

	Ni (ppm)	Cd (ppm)	Al (ppm)	As (ppm)	Pb (ppm)
<i>B. welwitschii</i>	64.58 ± 2.04	14.97 ± 2.66	2893.00 ± 107,62	2508.73 ± 85.10	737.10 ± 29.56
<i>O. welwitschii</i>	33.86 ± 0.92	14.04 ± 2.17	1709.33 ± 61.26	1333.50 ± 139.54	428.23 ± 23.73
<i>Z. gillettii</i>	23.53 ± 1.04	15.26 ± 2.89	1019.00 ± 31.43	328.67 ± 45.11	211.40 ± 4.01
<i>H. madagascariensis</i>	45.53 ± 3.01	4.28 ± 0.83	1872.67 ± 89.67	521.33 ± 20.58	359.90 ± 32.48
CCME	50	1,4	1,4	80	70

Legend: Cd = cadmium; Pb = Lead; Ni = nickel; Al = Aluminum; As = arsenic; Ppm = parts per million = mg.kg<sup>-1</sup>; CCME = canadian council of ministers of the environment



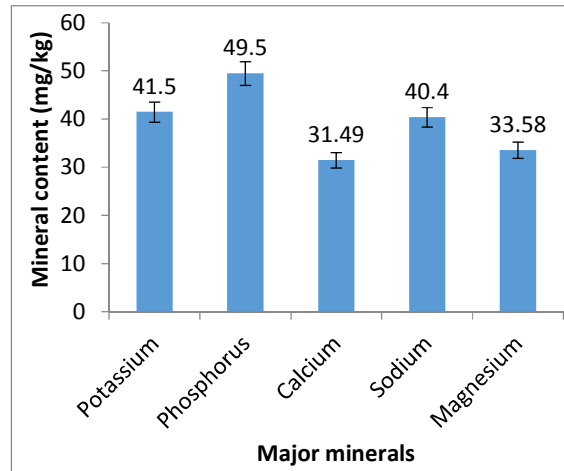


Fig. 3. Concentration (mg/kg) of major minerals in soils under the litter of *B. welwitschii*

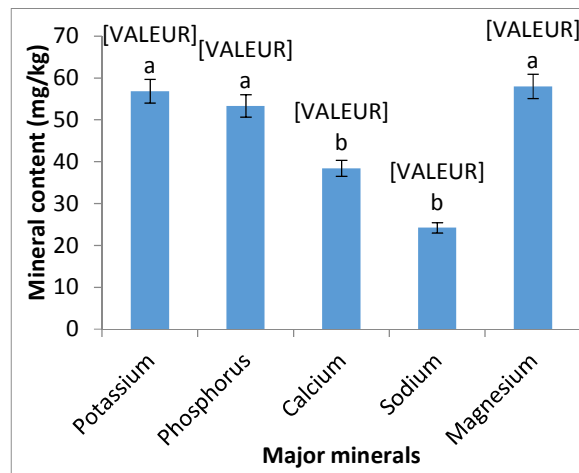


Fig. 4. Concentration (mg/kg) of major minerals in soils under the litter of *O. welwitschii*

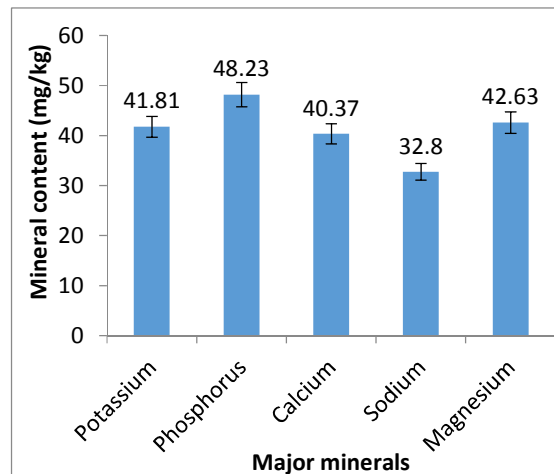


Fig. 5. Concentration (mg/kg) of major minerals in soils under the litter of *Z. gillettii*

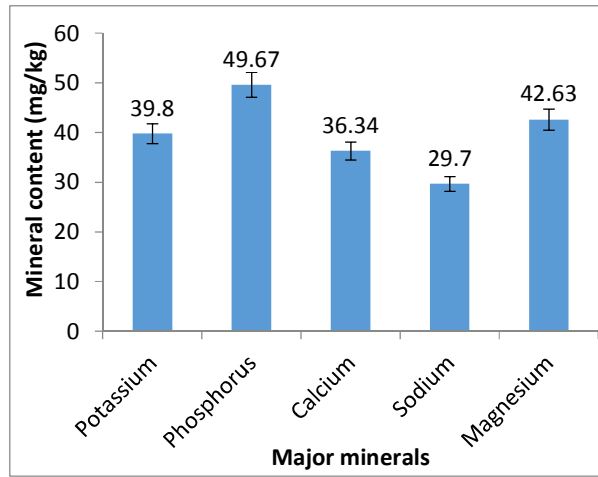


Fig. 6. Concentration (mg/kg) of major minerals in soils under the litter of *H. madagascariensis*

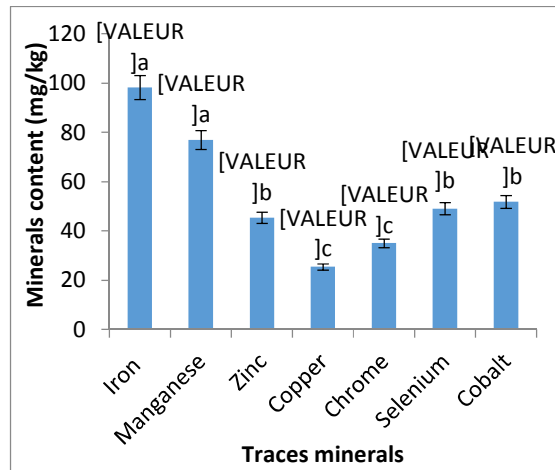


Fig. 7. Concentration (mg/kg) of traces minerals in soils under the litter *B. welwitschii*

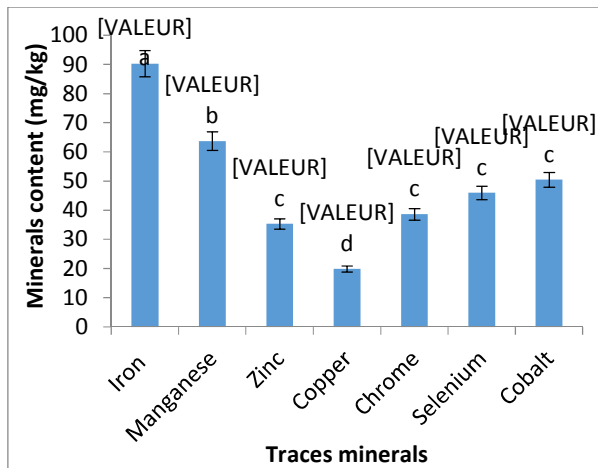


Fig. 8. Concentration (mg/kg) of traces minerals in soils under the litter *O. welwitschii*

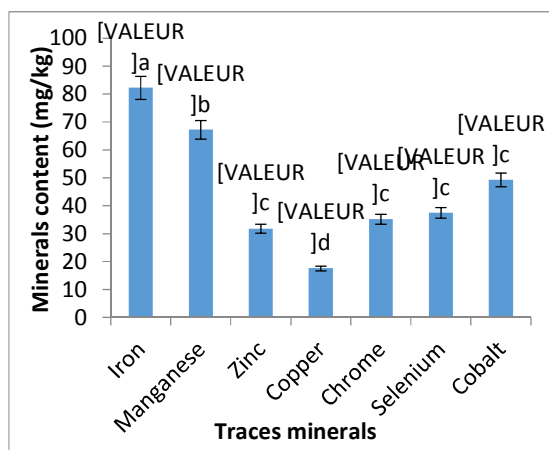


Fig. 9. Concentration (mg/kg) of traces minerals in soils under the litter *Z. gillettii*

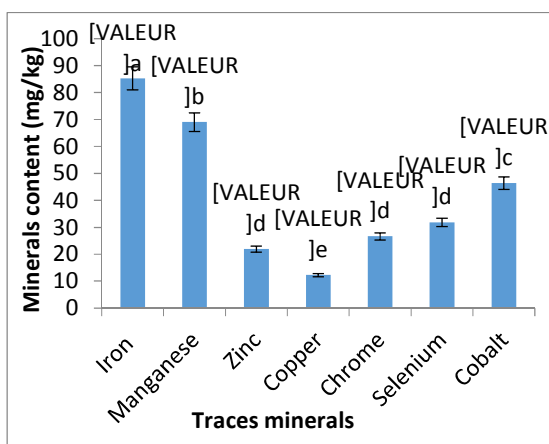


Fig. 10. Concentration (mg/kg) of traces minerals in soils under the litter *H. madagascariensis*

Table 4. Heavy metal pollution factors for soils under the litter

	Ni (ppm)	Cd (ppm)	Al (ppm)	As (ppm)	Pb (ppm)
<i>B. welwitschii</i>	1.29	10.69	2066.43	31.36	10.53
<i>O. welwitschii</i>	0.68	10.03	1220.71	16.67	6.12
<i>Z. gillettii</i>	0.47	10.90	727.86	4.11	3.02
<i>H. madagascariensis</i>	0.91	3.06	1337.14	6.51	5.14

Cd = cadmium; Pb = lead; Ni = nickel; Al = Aluminum; As = arsenic; Ppm = parts per million =  $mg.kg^{-1}$ ; the pollution factor is without unit

The minerals shown in Figs. 5 and 6 show that the phosphorus concentration is always higher in soils under the litter of *Z. gillettii* (48.23 mg/kg) and *H. madagascariensis* (49.67 mg/kg), followed by the magnesium concentration 42.63 mg/kg respectively for soils under the litter of *Z. gillettii* and *H. madagascariensis*. It is noted that the sodium concentration is low in the soils under the litter of these two plant species.

### 3.1.3.2 Concentration of traces minerals (mg/kg) on soils under litter

The concentration of trace minerals in soils under the litter of each plant species under study are presented in the figures.

Figs. 7 and 8 show that iron concentration is high in soils under the litter of *B. welwitschii* (98.33 mg/kg) and *O. welwitschii* (90.33 mg/kg).

Followed by the manganese concentration 77.01 mg/kg for the soils under the *Blighia welwitschii* litter and 63.72 mg/kg for the soils under the *O. welwitschii* litter. Copper, zinc, and chromium are trace minerals with low concentrations in soils under the litter of these two species. As for cobalt, it has a concentration of 51.91 mg/kg in the soil under the *B. welwitschii* litter and 0.5 mg/kg in the soil under the *O. welwitschii* litter.

The minerals in Figs. 9 and 10 show that in soils under *Z. gillettii* litter, the iron concentration is high (82.33 mg/kg) relative to other elements, followed by manganese (67.29 mg/kg). The same trend is also observed in soils under the litter of *H. madagascariensis*. Iron and manganese are the trace minerals that have a high concentration. While zinc and copper have low concentrations in these soils.

### 3.1.3.3 Concentration of heavy metals (mg/kg) on soils under litter

The concentration of heavy metals of different soils is presented in the table.

Pollution factors are presented in Tables 3-7. These factors are used to evaluate the pollution of the soil under study by heavy metals. The pollution factor is the ratio between the average concentration of a metal in the soil analyzed and the value of the standard for the same metal. This factor makes it possible to specify the degree of pollution. A soil is more polluted by a metal when it has a higher pollution factor for that metal. A metal is more polluting when it has a higher pollution factor.

If the pollution factor exceeds 1, it indicates that the average concentration of the metal in the soil being analyzed exceeds the value of the standard. This implies that the sample is polluted.

The aluminum has very high pollution factors in the littered soil of all plants. Nickel has very low pollution factors (less than 1) except at the level of soils under litter of *B. welwitschii*. Lead has higher pollution factors than nickel but lower than cadmium, except for the litter of *H. madagascariensis*. Pollution factors for *H. madagascariensis* (reference species) exceed 1 except for nickel, and are lower than for *B. welwitschii* and *O. welwitschii* but higher than for *Z. gillettii*.

In general, the descending order of pollution is as follows:

- At the level of soils under litter of the different species: *B. welwitschii* (higher factors) > *O. welwitschii* > *H. madagascariensis* > *Z. gillettii* (lower factors).
- Heavy metals in soils: Aluminum (higher factors) > Arsenic > Cadmium > Lead > Nickel (lower factors).

## 4. DISCUSSION

### 4.1 Granulometric Analysis

The granulometric analysis shows a sandy texture, according to the FAO classification. Clay shows significant difference in the soils under the litter of *O. welwitschii*, compared to the soil under the litter of other species. Soils under the litter of *O. welwitschii*, *Z. gillettii* and *H. madagascariensis* show a higher silt content than the soil under the litter of *B. welwitschii*. Although studies on the influence of agricultural practices on the properties of tropical soils [51] rarely show a change in soil texture, the present study highlighted the influence of *O. welwitschii*, *Z. gillettii* and *H. madagascariensis* species on the content of grain size fractions by following slices of simple biodynamic solum samples from reference pits.

Clay is the most active granulometric fraction because not only does it associate with humified organic matter, but it also has a high water retention capacity and contributes to the fixation of cations and anions on its exchange sites with plant species [52-53] Clay sufficiently influences the texture and thus the behaviour of a soil. In this case, in the American triangle, beyond 20% the term clay is used in the denomination of textural classes at the prefix or suffix level [54]. The same applies to Congolese soils with the difference that here, we consider rather the content of fine elements (clay + silt), with the lower limit of 15%. As for the role of silts and sands, it must be taken into account when the sample contains less than 35% clay [55]. The silt content, which is very low and characteristic of ferrallitic soils [56] is numerically similar, but significantly very different in different soils under different litters of studied plants.

### 4.2 Chemical Analysis

The pH results observed these soils in the slightly acidic soil category. This acidity is due to the transfer of acidic elements, especially oxides

and hydroxides of iron and aluminum. These metals are found in large quantities in tropical soils and induce an aluminic and ferric acidity that can lower the pH of the soil. Similar results were found by Yemefack et al. [57] in Southern Cameroon. These authors found that soils under certain plant species had pH values ranging between 4.5 and 5.7 and that the distribution of horizons from surface to depth influenced the decrease in pH values. Moreover, some plant species in the tropics have substances that acidify soils after decomposition. Lowering the pH promotes the mobility of trace metal elements, particularly by dissolving metal salts or destroying the retention phase. Conversely, raising the pH causes immobilization through the formation of insoluble compounds or an increase in cation exchange capacity [58]. Moreover, raising the pH and the presence of OH ions allows the formation of insoluble metal hydroxides that limit the migration of TME to the plant and to the water table and surface water [59].

The water content observed in the soils under the litter did not show any significant difference between these different species. However, the water content obtained in soils under the litter of *O. welwitschii* (45.63%) and *Z. gilletii* (44.99%) were higher than those in soils under the litter of *B. welwitschii* (35.37%) and *H. madagascariensis* (39.31%). These results are justified by the fact that the decomposition of the leaves of the species *O. welwitschii* and *Z. gilletii* increases the moisture in the soils, as the leaves of these species accumulate large amounts of water in their tissues compared to the other two plant species. Van Wambeke [60] had long argued that tropical soils under the litter of some plant species have water contents of more than 30%. The tropical dense forest, because of its ecological diversity and density, intercepts quantities of water during rainy episodes, resulting in the regulation of infiltration and evapotranspiration as well as an increase in soil moisture.

Organic matter is the very important element capable of providing, after mineralization, the essential mineral elements in soils. The organic matter content thus observed in the soils under the litter of studied species did not show any significant difference. However, soils under the litter of *O. welwitschii*, *Z. gilletii* and *H. madagascariensis* showed higher organic matter contents than soils under the litter of *B.*

*welwitschii*. The average organic matter content thus observed (21.36%) in the soils under the litter of these species indicates a good decomposition of these litters.

Furthermore, the organic matter (%) varies between 15.93 and 25.27. It is higher in the surface samples (10-20 cm) and this is explained by the presence of humus in this surface horizon. Soils under *B. welwitschii* litter have the lowest value. These findings are consistent with those of Segalen [61], which already reported a rather modest amount of OM for ferrallitic soils. The presence of organic matter in soils is responsible for the appearance of certain physico-chemical properties (e.g., soil bulk density and water content) [62]. The increasing these contents is accompanied by an improvement in structure, ease of water infiltration, and increased water retention capacity [63]. In addition, organic matter, with its colloidal properties, element binding and chelating properties, plays an important chemical role in soils: it releases nutrients after mineralization and increases the cation exchange capacity [62].

### 4.3 Mineral Content

Mineralization is a very important process in soils, because through it, matter passes from the organic state to the mineral state. This passage is always accompanied by the release of a large number of mineral elements in several forms and at different concentrations. The concentration of major mineral elements recorded in soils under the litter of each plant species by the spectrometry method AAS and ICP-AES did not show any significant difference. However, potassium, phosphorus, and magnesium have a high concentration compared to calcium and sodium. While the concentration of these mineral elements is significantly different in soils under *O. welwitschii* litter. Phosphorus, magnesium and potassium were found to be more concentrated in these soils than calcium and sodium. These results are justified by the fact that this species concentrates phosphorus, magnesium and potassium in the tissues during its mineral nutrition; and after the leaves and branches fall, the decomposition-mineralization process enriches the soils with these elements.

The concentration of trace minerals in soils under the litter of these four plant species showed significant differences. Iron concentration is high in soils under the litter of *B. welwitschii* (98.33 mg/kg), followed by soils under the litter of *O.*

*welwitschii* (90.33 mg/kg), *H. madagascariensis* (85.33 mg/kg) and finally *Z. gilletii* (82.33 mg/kg). As for manganese concentration, the soils under the litter of *B. welwitschii* showed a high concentration (77.01 mg/kg), followed by the soils under the litter of *H. madagascariensis* (69.11 mg/kg), *Z. gilletii* (67.29 mg/kg), and finally the soils under the litter of *O. welwitschii* (63.72 mg/kg). These results are justified by the fact that tropical soils have a high concentration of certain metallic elements, such as iron, aluminum, manganese, etc., which can be found in the soil under *O. welwitschii* (63.72 mg/kg). These trace elements usually combine with oxides and hydroxides to form chelate groups consisting of iron and aluminum oxyhydroxides. Manganese can replace magnesium in the weathering complex. The consequence of this phenomenon is a decrease in the pH of the soil. These results are similar to those found by Sombo et al. [64] in Batéké Plateau soils. These authors stated that iron and manganese as well as other trace minerals, notably copper, and zinc are still present in the soils and at different concentrations. During mineral nutrition, these elements accumulate in plant tissues and can return to the soil surface through the life cycle upwelling mechanism.

Our study showed that the levels of cadmium, lead, arsenic and aluminum in the various samples of litter soils of all plant species including nickel for *B. welwitschii* are above the standards set by the Canadian Council of Ministers of the Environment for soil quality [44]. Pollution factors for aluminum are the highest. This indicates that aluminum is the most polluting metal. The low pollution factors for lead and nickel are due to the low fixation of these metals by soils.

## 5. CONCLUSION

The objective of this study was to compare heavy metal content, the texture and the mineral element contents of soils under the litter of *B. welwitschii*, *O. welwitschii*, *Z. gilletii* and *H. madagascariensis* in relation to the plan of their pedogenesis. Different findings showed that physico-chemical analyses alone, significant differences in the mineralogical and granulometric composition of a soil under different plant species can be found. It was circumscribed within the framework of the soil-plant relationship, which is an indicator par excellence of the knowledge of the pedogenetic status of the different soils evolving in the humid

tropics. Thus, each plant species influences the soil with regard to its biological, chemical and biochemical nature. The results obtained showed that the soils under the litter of the above-mentioned plant species are slightly acidic with a sufficient organic matter content to enrich different horizons with major mineral elements as well as trace minerals. Among the major minerals, phosphorus, potassium and magnesium have a high concentration in these soils compared to the other elements. While for trace elements, iron and manganese have a very high concentration compared to the other elements. However, the difference in soil concentration of these elements is not statistically significant between soils under the litter of these four plant species. This study has revealed new aspects of heavy metal pollution. This pollution represents a serious threat to the environment in general and to humans in particular through the food chain. The study should be extended to the human organism in order to assess the effects of heavy metals on health and to develop health and nutritional monitoring programs for populations exposed to them.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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