Original Research Article

Characterization of soil toxicity in Kinkazi, Kitombe and Thiende villages affected by oil activities in Muanda and potential risk of chemical poisoning

ABSTRACT

Aims: Discharges from oil exploitation contain very toxic and persistent organic and inorganic environmental pollutants. The present study aims to characterize the ecotoxicity of the soils of these three villages subjected to oil exploitation activities.

Study design: This research consists of four parts, an introduction with literature review, a description of the study environment, the methods used and finally the results obtained.

Place and Duration of Study: In order to assess the impacts due to oil exploitation activities on the soil, a study was carried out on six soil samples taken from the villages Kinkazi, Kitombe and Thiende during the month of November 2022 in the territory of Muanda, Kongo Central, D.R. Congo.

Methodology: The soil samples were prepared in the laboratory according to standard procedures and a series of ecotoxicological tests were carried out with the analysis of some physico-chemical parameters using a Multiparameter. The data collected was processed and analysed with Excel 2010 software.

Results: The soils of the Kinkazi, Kitombe and Thiende villages, like dumping grounds for pollutants from oil activities, are and represent a risk of chemical poisoning for the population. The inhibitory dose (ID_{50}) varies from 26.25 to 36 g/g indicating the toxicity of these soils to *Hibiscus asper* seeds and the germination index obtained gives an average of around 58.7%.

Conclusion: The ecotoxicological impact of oil activities on the soil represents a risk of chemical poisoning for the local population of Muanda given that these polluted soils are primarily used for agriculture and the species *Hibiscus asper* is one of the most popular species for crops market gardeners in the region.

Keywords: Petroleum activities, soils, Toxicity, Hibiscus asper, Muanda, Kongo-Central, D.R. Congo

1. INTRODUCTION

Oil has been exploited in the territory of Muanda in the D.R. Congo since 1975, first by the American Chevron in collaboration with Teikoku-Unocal and Fina and then by the Franco-British company PERENCO Rep.in 2000. Alongside exploitation in the maritime zone (offshore) which began in 1975, exploitation activities in the land zone (onshore) only really began in 1981 [1].

Oil extraction generates discharges that the population accuses of being at the root of water, air and soil pollution, the destruction of food crops, the scarcity of fishery species resources and the problemspublic health [2].In the villages of Kinkazi, Kitombe and Thiende in the territory of Muanda in the D.R. Congo, oil exploitation activities are concentrated, which are accompanied by damage to the environment and damage to the health of local populations.

Some operating sites raise the question of environmental risk through their function of “storage” of by-products or industrial waste [3], and the contamination of the environment that follows.

Indeed, this soil contamination by organic and inorganic pollutants from petroleum activities can influence the ecological balance and sometimes lead to the destruction of ecosystems as well as the erosion of biodiversity[4], due to the release of derivativestoxic, in particular PAH's and other Pop's and heavy metals.Consequently, their accumulation in the animal and plant organism represents a risk for public health[5].This risk is all the higher since, considering the generally carcinogenic and mutagenic properties of molecules such as PAHs [6] and trace metals, consumption is the main route of exposure to these pollutants, exceeding thefrom other exposure pathways such as the nasal and dermal routes.However, the soils of Muandaare largely used in an agricultural context, with market gardening.

It is therefore urgent to control soil pollution in order to preserve its fertility, its productivity [3,7], the organisms that live there and thus protect biodiversity while respecting the criteria of sustainable development.

Furthermore, the management of soil biological resources must be considered as an essential element for the sustainability of this ecosystem [8].The biological quality of soils refers to the abundance, diversity and activity of organisms involved in the functioning of these ecosystems. Thus, the objective of this study is to determine the toxicity of this soil for the *HibiscusAsper* species in order to assess its impact on the biological functioning of this ecosystem.

1.1. Assumption

The soils of the villages Kinkazi, Kitombe and Thiende in the territory of Muanda constitute directly or indirectly the dumping grounds for hydrocarbons exploited by the local oil company, and this affects the ecosystem balance of these environments.

1.2. Objective

The objective pursued in this study is to analyze the toxicity of the soils of the villages Kinkazi, Kitombe and Thiende in Muanda with respect to the specimen of *Hibiscus asper* (one of the most cultivated vegetable species and intended for human consumption).

Specifically, this study consisted of characterize the polluted soil matrix; determine the degree of toxicity of these soils and define a decision support tool.

2. STUDY SITES, MATERIAL AND METHOD

The villages Kinkazi, Kintombe and Thiende are part of the area affected by oil activities in the territory of Muanda (Image 1), located in the west of the country, in the province of Kongo-Central between the meridian 12° and 12°30' East and the parallels 5°30 and 6° South, i.e. a quadrilateral 40 km long and 10 km wide for an area of 6,000 square kilometers, of which just over 1,000 square kilometers is offshore [3].

Oil exploitation activities in these villages are also encountered in all the other villages of Muanda, sometimes close to dwellings, that is to say less than 10 meters for some, and on the outskirts of villages.They extend over an area of 400 square meters, which represents a land area of nearly 10% of the 4265 square kilometers of Muanda territory.Thus, the proximity of these activities makes the risk of pollution by pollutants from this type of activity very high.

2.1. Methodology

2.1.1. Observation

The organoleptic observation made on the sampling sites alerts to possible contamination by petroleum products.Indeed, the observation focused on the signs of contamination and this from visual clues such as the blackish coloring of the soil and the pronounced odor (olfactory index).Also, the results obtained during this observation made it possible to formulate the first hypotheses on possible pollution and therefore the need to take samples to assess the nature and concentration of contaminants by means of in-depth laboratory analyses.

2.1.2. Collection of soilsamples :

The soils intended for the bio-tests were sampled using a steel auger, placed in previously sterilized 1 kg tinted glass bottles, then stored in a cooler with accumulators maintaining the temperature around 4°C,then sent to the laboratory for analysis.

As part of this study, six composite soil samples were taken from three villages (see Fig. 1).

2.1.2.1. Sampling conditions

The samples were taken according to the procedure proposed by the Genosol platform, which describes how to take representative soil samples from the sampling point [9].

Process:

• **Sampling period**: in order to aim for optimal conditions, the samples were taken in soil that was neither too dry nor too wet.To do this, a period of two days apart was observed after the rain (after runoff) to sample.

• **Sampling conditions**: the samples taken were treated and stored separately to avoid any form of contamination of the samples and/or between them.To do this, only clean equipment was used, washed and sterilized beforehand;then change the equipment when the sampling site is changed.

2.1.3. Physico-chemicalcharacterization of soils

The physico-chemical analyzes focused on measuring the hydrogen potential (pH) and temperature.This choice is justified by the direct implication of these parameters on the acute ecotoxicity test.Indeed, the pH acts on the bioavailability of contaminants, i.e. organic and inorganic pollutants from petroleum activities whose toxicity is assessed here, while the temperature can influence the volatilization of hydrocarbons [10] both above and below ground[11].

Thus, the measurement of pH-water, pH-KCl and temperature was obtained using a multi-parameter (in a soil/solute suspension for pH).

2.1.4.Ecotoxicological analysis with the species *Hibiscus asper*

To assess the toxicity of the soils of Muanda contaminated by organic and inorganic pollutants resulting from the oil exploitation activities of the place, we used as a model (bio-indicator) the seeds of *Hibiscus asper* (Sorrel of Guinea).The test specimens were collected in a capsule containing 29 seeds in the experimental garden of the Department of Biology of the Faculty of Science at the University of Kinshasa.Then, the recovered seeds were soaked in water in order to recover the viable seeds, those floating (non-viable) were eliminated and those submerged (viable)

were recovered for the test, in accordance with the approach followed by[12].

Six kilograms of potentially polluted soils taken from the vicinity of the oil installations in Muanda formed the basis from which the mixtures were prepared with a dilution matrix, natural soil, recommended in the AFNOR X 31-202 standard [13].Ten individuals of *Hibiscus asper* were placed in each pot for the toxicity test.Each concentration was repeated five times.The following manifestations were observed: emergence of the radicle, elongation of the stem, rotting of the seed.

Process:

• **Mixture (concentration)**: The technique consists in preparing different soil mixtures to study the effect of 5 concentrations, 25, 50, 75, 100% of diluted sample (or not, in the case of 100% and 0%, that is to say the control solution);that is, each pot contained a given concentration.First 100% contaminated soil, 75% contaminated soil + 25% mixed soil, 50% contaminated soil + 50% mixed soil;25% contaminated soil + 75% mixture soil, 0% contaminated soil + 100% control soil.The test is repeated 3 times.

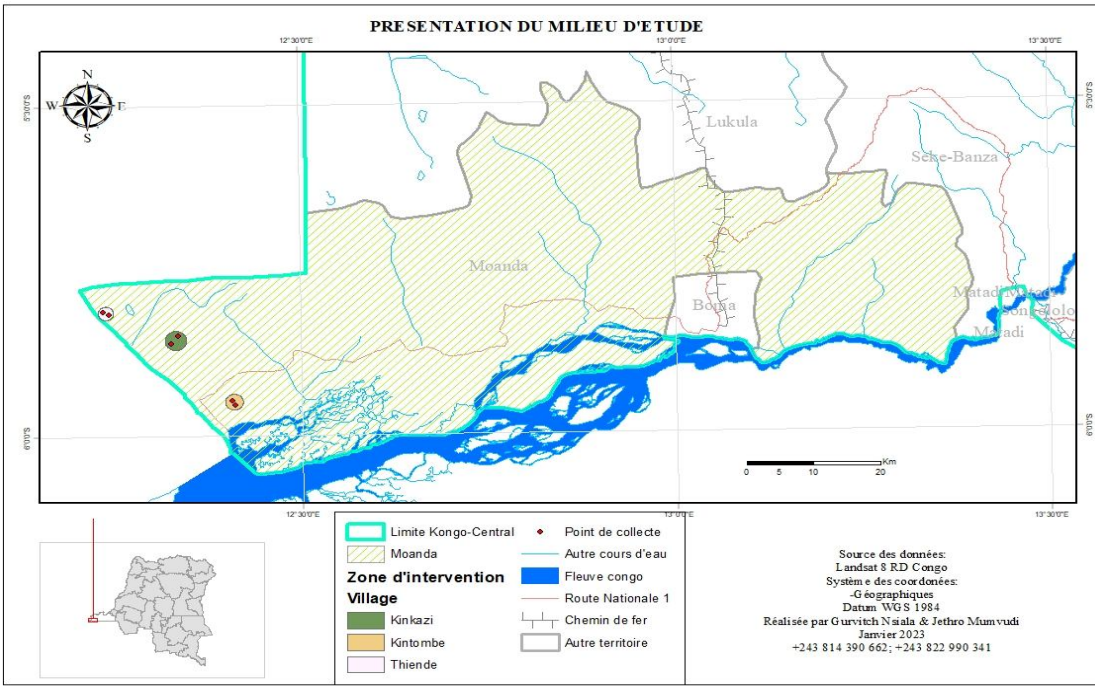
• **Bio-indicator**: we therefore put 3 times ten *Hibiscus asper* seeds in each mixture (concentration), i.e. 30 seeds per concentration; which allowed us to observe them for 7 days according to the toxicity test of inhibition of seed germination by a potentially polluted matrix [14].The technique consists of observing the emergence of the seed radicle daily during watering.

After 7 days, the germinated seeds were counted in the different dilutions to determine the effect on germination, and the number of shoots was reduced to five.The results obtained made it possible to calculate the ID_{50} using the Karber and Behrens equation[15].

$$ID_{50} = \mathbf{ID}_{100} - \frac{\Sigma(\mathbf{ab})}{\mathbf{n}}$$

with the following parameters:

- n = Average of seeds per batch,
- a = Mean inhibition between doses,
- b = Difference between two successive doses.



and the results of the ID_{50} give a value around 29.25 g/g.

3. RESULTS AND DISCUSSION

3.1. Results

3.1.1. Physico-chemical parameters

Some indicator parameters of the physico-chemical properties of the soil, representative for an ecotoxicological study, are given in table 2 below.

Table 1 on the physico-chemical parameters shows that all the samples reveal a more less neutral pH (6.69 ± 0.11 to 7.94 ± 0.13).Regarding temperature, the results revealed an average around 24.3 ± 0.64 °C, with variations ranging from 23.6 ± 0.22 to 25.1 ± 1.29 °C.

3.1.2. The biotest

The results of the effect of soil pollution by oil activities on plant biodiversity are illustrated by the tables below:

As seen tin table 2, the result of the germination index obtained is around 62% of *Hibiscus asper* seeds.In 100% of the polluted matrix, the results showed 40% germination;in 75 and 50% of the polluted matrix half of the seeds germinated while in 25% we observed 70% germination. The ID_{50} of this soil is around 36 g/g testifying to its toxicity for *Hibiscus asper* seeds.

In the table 3, the observation made on the soil of the "K2" site reveals in its raw concentration (100% potentially polluted soil) 70% germination of *Hibiscus asper* seeds, followed by concentrations of 75% and 50% with respectively 40% and 50% germination, then the concentrations of 25% and 0% (control soil) of which 70% and 90% germination were observed in each of the respective concentrations.The germination index is 64% while the ID_{50} results reveal a value around 34.5 g/g.

The table 4 shows that the response of the *Hibiscus asper* species whose seed germination was tested on the soil of the "KTM1" site revealed in its raw concentration (100% soil) and 75% an average around half of the germinated seeds, andthe other concentrations (50% and 25%) allowed 30% and 60% seeds to germinate respectively.In addition, the germination index is around 54%

The soil of the “KTM2” site in Kitombe village in Muanda, as seen in the table 5, reveals in its raw concentration (100% soil) and 75% an average of sprouted seeds around 40%, followed by the concentration of 50% with half of sprouted seeds, then the concentration of 25% with 70% of germinated seeds, and finally in the control soil all the seeds subjected to the experiment germinated, that is to say 100%.The germination index is estimated at around 60%. This soil showed its lethal toxicity (ID_{50}) against *Hibiscus asper* seeds from the value of 34.5 g/g.

In table 6, the soil of site “TH1” in Thiende village in Muanda was found to be highly toxic to Hibiscus asper seeds.Indeed, in the raw concentration, 30% of seeds germinated and but in the concentration of 75% of soil, there was 10% germination.In addition, in the concentration of 50% and 25% of this soil, we had 70% and 60% of germinated seeds respectively, while on the control soil we observed 70% germination;the germination index being around 48%.The ID_{50} is around 26.25 g/g indicating the toxicity of this soil to *Hibiscus asper* seeds.

As seen in the table 7, the soil of the “TH2” site in the Thiende village in Muanda reveals in its raw concentration (100% soil) toxicity inhibiting the germination of 40% of *Hibiscus asper* seeds.In the concentration of 75%, 50% and 25% an average of seeds germinated respectively around 50%, 70% and 60%.On the control soil there were 80% of germinated seeds.In short, the germination index gave a value of around 64% and the effluent showed its lethal toxicity (ID_{50}) towards *Hibiscus asper* seeds around 36 g/g.

Thus, at the end of the Biotest carried out for seven days in the laboratory, it appears that the soils of the different sites "K1, K2, KTM1, KTM2, TH1 and TH2" in the territory of Muanda are toxic on different scales.

Table 1. Physico-chemical parameters relating to the temperature and the reaction of the soils of all the sites

Parameters	Soil samples					
	K1	K2	KTM1	KTM2	TH1	TH2
pH Water	7,57 ± 0,03	7,29 ± 0,89	7,94 ± 0,13	7,72 ± 0,08	7,27 ± 0,10	7,38 ± 0,06
pH KCL	7,28 ± 0,19	7,51 ± 0,07	7,61 ± 0,10	7,4 1± 0,06	6,69 ± 0,11	7,2 ± 0,04
Temperature	25,1 ± 1,29	24,4 ± 0,27	24,5 ± 0,27	23,6 ± 0,22	24,4 ± 1,01	23,8 ± 0,77

Table 2. Number and percentage of germinated seeds of *Hibiscus asper* on contaminated soil of the K1 site (Village Kinkazi)

Contaminated soils in g*	Dilution matrix in g*	Observation							Total	% sprouted seeds	% ungerminated seeds
		Day1	Day2	Day3	Day4	Day5	Day6	Day7			
60	**0**	0	0	0	2	1	1	0	4	40	60
45	**15**	0	0	1	1	2	1	0	5	50	50
30	**30**	0	0	1	0	3	1	0	5	50	50
15	**45**	0	0	3	0	1	3	0	7	70	30
0	**60**	0	0	8	1	1	0	0	10	100	0

g: gram ; % : percentage*

Table 3. **Number and percentage of germinated seeds of *Hibiscus asper* on contaminated soil of the K2 site (Village Kinkazi)**

Contaminatedsoils in g*	Dilution matrix in g*	Observation							Total	% sproutedseeds	% ungerminatedseeds
		Day1	Day2	Day3	Day4	Day5	Day6	Day7			
60	**0**	0	0	2	4	1	0	0	7	70	30
45	**15**	0	0	2	1	1	0	0	4	40	60
30	**30**	0	0	3	1	1	0	0	5	50	50
15	**45**	0	0	3	1	2	1	0	7	70	30
0	**60**	0	0	7	1	2	0	0	9	90	10

g: gram ; % : percentage*

Table 4. Number and percentage of germinated seeds of *Hibiscus asper* on contaminated soil of the KTM1 (Village Kitombe)

Contaminatedsoils in g*	Dilution matrix in g*	Observation							Total	% sproutedseeds	% ungerminatedseeds
		Day1	Day2	Day3	Day4	Day5	Day6	Day7			
60	**0**	0	0	1	1	3	0	0	5	50	50
45	**15**	0	0	2	2	1	0	0	5	50	50
30	**30**	0	0	3	0	0	0	0	3	30	70
15	**45**	0	0	5	1	0	0	0	6	60	40
0	**60**	0	0	6	1	1	0	0	8	80	20

g: gram ; % : percentage*

Table 5. Number and percentage of germinated seeds of *Hibiscus asper* on contaminated soil of the KTM2 site (Village Kitombe)

Contaminatedsoils in g*	Dilution matrix in g*	Observation							Total	% sproutedseeds	% ungerminatedseeds
		Day1	Day2	Day3	Day4	Day5	Day6	Day7			
60	**0**	0	0	1	2	1	0	0	4	40	60
45	**15**	0	0	2	1	1	0	0	4	40	60
30	**30**	0	0	2	3	0	0	0	5	50	50
15	**45**	0	0	3	3	0	1	0	7	70	30
0	**60**	0	0	9	1	0	0	0	10	100	0

g: gram ; % : percentage*

Table 6. Number and percentage of germinated seeds of *Hibiscus asper* on contaminated soil of the TH1 site (Village Thiende)

Contaminatedsoils in g*	Dilution matrix in g*	Observation							Total	% sproutedseeds	% ungerminatedseeds
		Day1	Day2	Day3	Day4	Day5	Day6	Day7			
60	0	0	0	1	1	1	0	0	3	30	70
45	15	0	0	1	0	0	0	0	1	10	90
30	30	0	0	2	3	2	0	0	7	70	30
15	45	0	0	3	2	1	0	0	6	60	40
0	60	0	0	6	1	0	0	0	7	70	30

g: gram ; % : percentage*

Table 7. Number and percentage of germinated seeds of *Hibiscus asper* on contaminated soil of the TH2 site (Village Thiende)

Contaminatedsoils in g*	Dilution matrix in g*	Observation							Total	% sproutedseeds	% ungerminatedseeds
		Day1	Day2	Day3	Day4	Day5	Day6	Day7			
60	**0**	0	0	2	1	2	1	0	6	60	40
45	**15**	0	0	0	3	1	0	1	5	50	50
30	**30**	0	0	1	2	3	1	0	7	70	30
15	**45**	0	0	3	2	1	0	0	6	60	40
0	**60**	0	0	6	1	1	0	0	8	80	20

g: gram ; % : percentage*

3.2. Discussion

3.2.1. Physico-chemical parameters

The temperatures varied between 23.6 ± 0.22 to 25.1 ± 1.29 °C and this below 30°C as a preference.As far as pH is concerned, the soils all have a more or less neutral pH which migrates within the range of 6.69 ± 0.11 to 7.94 ± 0.13.According to [16] the pH of our soils is slightly lower than that of agricultural pH (7.5<ph<8.5).Moreover, these pH values varied very significantly from one site to another (*P* = .000); however, those of temperature did not vary significantly between sites (*P* = .941).Note that thephysico-chemical characteristics of the soils mentioned above could influence the behavior of pollutants in the soil, among others the volatilization of hydrocarbons when the soil temperature increases and their bioavailability as a pollutant as the pH decreases.

These observations are supported by the study by [17], on the characterization of the contamination of banks by hydrocarbons, the results of which showed that the physico-chemical parameters of a soil influence the absorption and migration of contaminants.In addition, a study [18] underline the fact that the presence of hydrocarbons in the soil increases the concentration of the latter in organic compounds and this increase can affect its physical and chemical properties such as pH and temperature.

3.2.2. Biotests

From a methodological point of view, the numerous surveys on the ground having made possible the organoleptic findings such as: the blackish color, the pronounced odor, the oily characteristics of the soil, etc., have made it possible to formulate the hypotheses on a possible risk of pollution of severalsites in different villages in the territory of Muanda in the Democratic Republic of Congo.To verify these risks, we carried out biotests on a few soil samples taken from these sites.The results obtained on the inhibitory dose (ID_{50}) vary from 26.25 to 36 g/g indicating the toxicity of these soils to *Hibiscus asper* seeds.

The results of the germination index obtained range from 48% to 64%, with an average around 58.7% of germinated seeds.The values of the germination index obtained varied highly significantly from one site to another (*P* = .001).These results are very close to those found by[12] who obtained a germination index of around 58.24% of *P. lentiscus* seeds on a substrate polluted with hydrocarbons.

According to these results, it seems that the germination was not affected by the hydrocarbon pollution, even though the germination rates are not so famous.This could be explained by the resistance of the seeds of this species to this type of pollution.

We recorded rather surprising germination rates in the samples containing the large concentrations of polluted soil.Indeed, overall the germination rate obtained in the raw concentration ranges from 40% to 70%, this could be due to the hydrocarbons which prevented the infiltration of water contained in the soils, therefore it allows the soil a contentin water which is a preliminary element for the triggering of the germination process.

Similar to us, the results of a study by [19] indicate that seed germination of *P. lentiscus* was not affected by oil pollution.Because the seeds of this species do not exhibit dormancy and only require the removal of the pulp and a short period of heavy rain to germinate.Thus, according to the same authors, this characteristic could play a role in favor of its resistance to hydrocarbon pollution because the seeds germinated before the hydrocarbons exerted their toxicities. As for the seeds of *Hibiscus asper*, although these pollutants do not exert an acute toxicity on their germination, they could reveal a subchronic or chronic toxicity according to the life cycle of the species.These observations are confirmed by a study conducted by [20], which showed that plant species such as agropyron, sunflower, clover and safflower, could well germinate on soil contaminated with hydrocarbons and see their growth be disturbedlater.

However, the fact that the seeds have germinated increases poses another environmental problem.In addition to the damage caused to the balance of receiving ecosystems, this problem goes beyond and could affect humans at the top of the food chain.According to [21] when fuels are spilled in soil, they do not migrate through soil profiles due to their hydrophobic properties but they will be retained on the soil surface and in the arearooting of most plant species.In this way, crops will be able to assimilate pollutants through their root system.

However, the fact that the seeds have germinated increases poses another

environmental problem.In addition to the damage caused to the balance of receiving ecosystems, this problem goes beyond and could affect humans at the top of the food chain.Still according to [21] when fuels are spilled in soil, they do not migrate through soil profiles due to their hydrophobic properties but they will be retained on the soil surface and in the arearooting of most plant species.In this way, crops will be able to assimilate pollutants through their root system.

That said, the risk of chemical poisoning will be great due to the consumption of vegetables grown on contaminated soil.As demonstrated by [22], hydrocarbons could penetrate through the cell membranes of a plant and cause their accumulations in one or more of its parts.Then, the hydrophobic compounds will pass directly into the cells by passive diffusion through the membranes.More polar molecules can be passively or actively absorbed via water flows, and be transported into the xylem or phloem [23].

This is what leads to the bioaccumulation of pollutants and represents for the local population of Muanda, a risk of chemical poisoning given these polluted soils have first an agricultural vocation and the species *Hibiscus asper* is among the mostpopular for market gardening in the region.By ripple effect, when it rains, the mixture of rainwater and overflows from boreholes extends over habitable areas and floods crop fields.This pollution, which is mainly due to leaching directly and or indirectly to spills, voluntary or involuntary, of hydrocarbons by the oil exploitation installations located in the villages of Muanda, in particular the village of Kinkazi, Kitombe and Thiende, also affects the environment in itstogether that the health of the population a chemical risk.

It should be noted that the territory of Muanda, apart from the three aforementioned villages, is the sector with the greatest concentration of oil exploitation facilities.The surroundings are affected by traces of pollution, so it would be interesting to repeat this same type of characterization study in the other villages of Muanda affected by oil exploitation activities.

4. CONCLUSION

The evaluation carried out within the framework of this study made it possible to determine the degree of toxicity of the soils of several sites in the villages of Muanda in the Democratic Republic of Congo.In view of the results obtained, it appears that this environment presents a chemical risk because the oil exploitation activities are close to residential houses and crop fields and, among other damages, dump pollutants into the ground;these pollutants do not exert immediate toxicity on the germination of seeds (*Hibiscus asper* seeds), but can be accumulated by them and cause a toxic reaction in humans, future consumers of these vegetable crops.

Given the current situation, it is therefore to be feared that this pollution will spread further if appropriate measures are not taken and rigorously applied on the ground.Because, in addition to the inhibition of seed germination, the presence of toxic agents in a soil can affect the growth of a plant and lead to a reduction in the biomass of the roots, the length/size of the roots, the stems,and leaves and premature senescence of leaves and, further, the mortality of humans following the consumption of contaminated vegetables.It would be interesting in future studies to study other physico-chemical and biological parameters to show the effect of the pollutants encountered in these sites, in the medium and long term, on the behavior of natural ecosystems.It would also be necessary to analyze the presence and content of pollutants, organic and inorganic, resulting from these oil activities in the plant tissues of market garden crops in the area.

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