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Changing Elemental Uptake of Roots and Leaves from Plants Grown on a Soil Variably Polluted by Crude Oil

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Abstract

A radish species was grown in a sandy loamy soil either unpolluted or polluted by increasing concentrations of crude oil added to watering solutions during one month under controlled laboratory conditions. This procedure was set to evaluate the impact of oil pollution of the substrate on the elemental uptake by plants. In summary, the increasing pollution by crude oil to the soil has not a univocal impact: the changing elemental contents in the roots and leaves of the cultivated radishes are never single trended with the amount of oil pollution, showing in turn that they are not provided by the spilled oil. The most significant elemental increase occurs in the leaves of the radishes grown in the soil polluted by 10 ml of oil and in the roots of those grown in the soil polluted by 4 ml of oil. In the detail, the significant effects of the oil pollution induce in the leaves: (1) similar behaviors for Ca, K, P, Mg, Fe and Al; (2) the highest impact on Ca, K, P, Mg and Al at the intermediate 10-ml pollution; (3) the highest uptake at the high side of pollution for Fe, Mn, Zn, Ni, Co, Cr and Pb; and (4) no significant impact on the uptake of the REEs. Oil pollution on the plant roots impacts: (1) an increased Ca, Fe, Al and Si uptake, often only in the case of the highest pollution, while P's uptake decreases; (2) an increase of the microbial population by a factor of about 2.5 at low pollution and a dramatic decrease at higher pollution; (3) an uptake of REEs only at the highest degree of pollution by a specific increase of the light REEs.

The translocation roots-to-leaves indicates a decrease of Ca to the leaves when oil pollution increases, while remaining state for K and P with a slight decrease when pollution is at its maximum. The total biomass increases in the soil at low levels of pollution, decreases at intermediate levels and remains the same at high levels of pollution. The increase of most of elements at low level of oil supply is correlated with an increase in microorganism density, which suggests that availability of elements in soil can be attributed to an increase in organic activity, which has been stimulated by the oil pollution.

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Introduction

Contamination of soils is progressively increasing all over the world because of a continuous industrial pollution, inducing modifications that affect and, therefore, alter the micro-organic communities and physical-chemical properties of soils on which plants grow, some being consumed by human populations. The specific impact of crude oil pollution on soil characteristics has already been identified and quantified in previous studies, but the conclusions are varied. For instance, Obasi et al. [1] observed a decrease in pH and of N and P of soils due to concomitant increase of the oil pollution. Alternatively, Adams and Ellis [2] observed an increase in P with time in soils affected by oil supply. Uzoho et al. [3] showed that crude oil contamination increases significantly the soil pH, up to 8.0, and reduces the available P concentrations. Onuh et al. [4] observed a decrease in the N availability with increasing level of crude oil pollution. Everett [5] demonstrated that there is a reduction in the water infiltration rate in soils polluted by oil relative to unpolluted references. For Okonokhua et al. [6], addition of oil to soils has no effect on both their pH and texture, while there was a decrease in P and an increase in Fe, Cu, Zn and Pb. Vanloocke et al. [7] also reported that soil pH tends to shift towards neutral values after hydrocarbon addition to both acidic and alkaline soils. The presence of oil in poorly drained soils can also cause anaerobic conditions [8]. In turn, a number of physiological processes in plants are affected variably by oil contamination of the soils in which they grew [9]. Inhibited seed germination in oil-contaminated soils can also be directly or indirectly related to oil contamination. Plice [10] attributed poor seed germination to the penetrating power of the volatile fraction of oil.

Therefore, oil pollution appears to affect plants in multiple ways. As reported by Baker [9], cell membranes can be damaged by penetration of hydrocarbon molecules and a reduction of photosynthesis can take place if oil enters the roots and reaches the leaves of the growing plants. Baker [11] also reported that oil pollution reduces transpiration of plants and increases or reduces their respiration ability. Oil can also inhibit germination of seeds. In turn, the critical level of oil pollution has been evaluated at about 1 kg of oil per square meter of soil [12], while Rowell [13] fixed this critical level at 8%.

It is known for some time that most heavy metals are toxic to plants and to organisms when present in the soils in excessive concentration. In the case of crude-oil pollution, the response of microorganisms is not similar at varied pollution levels. Ekpo and Ebeagwu [13] observed that low concentrations of oil in soils (about 1%) have no noticeable effect on the micro-organic population, while the initial response of microorganisms is a reduction in their growth potential followed by an increase at high pollution level. The decreased activity of microorganisms is related to anaerobicconditions induced by the occurrence of crude oil in soils [13-15]. Sparrow et al. [16] reported

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various responses of microorganisms to oil pollution in soils, either as an inhibitor or a stimulator depending on the micro-organic species of the contaminated soil.

In summary, many published studies were focused on estimating the impact of contaminants on soil microorganisms depending on the pollution stress. Studies of either elemental accumulation or depletion with possible elemental fractionation as a response to soil pollution by oil need also to be investigated in more details. The main objective of the present contribution was then to determine the impact of different levels of crude oil added to a soil on: (1) the transfer and fractionation of chemical elements from rhizosphere to the roots and further to the leaves of radish plants, and (2) the total microorganism mass in the rhizosphere. The purpose of the experiment was a strict evaluation of the impact of increasing oil pollution on the elemental uptake of a plant type normally grown for human consummation.

Materials and Analytical Procedure

The wild radish species Raphanus *sativus* was selected for this growth experiment, the substrate consisting of a loamy soil. The amount of substrate used to grow the radish plants was about 700 grams that were stored in pots of 10 cm diameter and 10 cm depth. Approximately 140 seeds of radish plant were sprayed on the substrate of each pot. The substrates were watered with deionized water twice a week. No fertilizers were added to the substrate during the experiment.

Five sets of pots were prepared: the non-polluted reference and two times two pots with substrate polluted by respectively 2, 4, 10, 16 and 20 ml of crude oil at the first watering. After thirty days of growth, the plants were collected in living conditions for chemical analysis. The plants of each pot were picked up randomly and split into five separate batches of about 20-25 individual plants each. The obtained chemical data represent the averages of five independent analyses, the uncontaminated reference and the contaminated counterparts (Table 1, Supplementary File). Actually, the results average the chemical composition of nearly 100-120 individual plants considered collectively.

After growth, the collected plants were first washed thoroughly with deionized water, and then shaken in a gentle ultrasonic bath of mild intensity for about 10 minutes to free any solid soil particle that could have adhered at the surface of the roots. After the ultrasonic bath, the plants were washed again with deionized water, some roots taken randomly being checked by optical microscopy for any presence of soil particles. Free of solid external particles, each batch of radish samples was then prepared for the chemical analyses.

The batches of root and leave segments were first dried at 60°C for 24 hours and weighed carefully. The plants were then ashed in a Pt crucible at about 600°C for about 45 minutes. The ash was transferred into a polypropylene beaker and digested in concentrated ultra-pure HNO_3 at about 700C for about 24 hours. The solution was then slowly evaporated to dryness. To ensure complete dissolution of any remaining organic material, a few drops of $HClO_4$ were added to the material and the sample evaporated again to dryness by closing the top of the beaker. The final solution was prepared by dissolving the dried material with a known volume of 1M HNO_3.

The elemental contents of both plant segments were determined by using an Inductively Coupled Plasma-Atomic Emission Spectrometer (ICP-AES) for the major elements (Si, Al, Mg, Ca, Fe, Mn, Na, K and P) and some trace elements (Cu, Zn, Ba and Ag) and an Inductively Coupled Plasma-Mass Spectrometer (ICP-MS) for most of the remaining trace (Zr, Rb, Sr, Mo, Cr, Co, Ni, U and Th) and the rare-earth elements (REEs). Standard minerals such as B-EN and GL-O were repetitively analyzed, providing with an average analytical precision of 3% for the major elements, 5% for the trace elements and 10% for the REEs, the technical aspects of the analytical procedure following those of Samuel et al. [17]. In turn, the individual elemental uncertainty was controlled for the complete analytical procedure by analyzing five independent batches of plants. The average value of the five variances for each analyzed element are summarized in the table 1 (Supplementary File). Within the analytical spread of the mineral standards used routinely in the procedure outlined above, the analytical reproducibility can then be considered to be double-checked.

For enumeration of the bacteria population of the soil, samples were collected from rhizosphere around the radish roots, stored in sterile containers and transported aseptically to the laboratory. For enumeration of the bacterial colonies, the soil samples were serially diluted and planted in nutrient agar medium. One gram of each soil sample was transferred to 99 ml of distilled water in a sterile bottle for a 10^{-2} dilution. After mixing, 1ml was transferred from 10^{-2} bottle to another tube with 9 ml sterile distilled water to get a 10^{-3} dilution. This method was continued until a 10^{-5} dilution. Serially diluted samples were inoculated into the nutrient agar medium by the spread plate method and incubated for 24 hours at 37°C.

Results

After one month of growth, the height of the radish plants reached about 20 cm and the diameter of leaves about 1 cm. Comparison of the growth density and the size of the radish plants in each pot showed that there were less plants with smaller leaves when the substrate (ICP-AES) for the major elements (Si, Al, Mg, Ca, Fe, Mn, Na, K and was polluted with 10-ml of oil compared to those grown in the other pots. The first analytical step was a comparison of elements taken up by the radish plants from unpolluted substrate with those taken up by those of the substrate polluted with different quantities of oil (Table 1, Supplementary File). Difference above 5% for the major elements, 10% for the trace elements and 20% for the REEs were considered to describe increased elementary uptakes under polluted conditions, whereas negative ratios beyond these percentages outlined the opposite, that is to say reduced elementary uptakes under polluted conditions. Differences between these values, i.e. within analytical uncertainty, were considered not to be representative of elemental changes.

Effect of oil substrate pollution on the major-element uptake

In the roots

Most of the major elements increased at the 2-ml oil pollution level, except unchanged Ca, and decreasing Si and Na relative to their contents in the reference plant (Figure 1, Supplementary File). The supply of 4-ml oil results in an enrichment of all elements except unchanged Mn, and decreasing Si and Mg. The uptake of the plant roots at the 10-ml supply of oil was not determined due to a technical failure. The supply of 16 and 20 ml of oil induced an increase of all elements, except Na. Comparing the concentration variations at the different pollution levels relative to the unpolluted reference plants revealed a significant change for those plants harvested from soil polluted by 20-ml oil. Comparison of the different elemental variations induced by the oil contamination indicates that Si, Al, Fe and Ti were subjected to higher variations than the other elements.

In the leaves

Relative to the reference plants grown in the unpolluted soil, supply of 2 ml of oil induced enrichment in Si, Al, Mg, Fe, Mn and P in the leaves, not changing the concentrations of Ca, Na and K (Figure 2, Supplementary File). Supply of 4 ml of oil resulted in an enrichment of Si, Na, K, P and Mg and no change for Al, Fe, Ca and Ti (Figure 2 , and Table 1, Supplementary File). Supply of 10 ml and 16 ml of oil resulted in an enrichment for all elements. Supply of 20 ml resulted in an enrichment of Al, Ca, Fe, Mn, and Ti and a depletion of Si, Na, K and P, and no change for Mg. By comparing the change observed at the different levels of oil pollution (Figure 2, Supplementary File), the results show that a significant change occurred at the 10-ml pollution level for most elements, with a discrete change at the 4-ml pollution level. A comparison indicates that Si, Al, Fe and Ti of the leaves, as for the roots, are more sensitive to oil pollution than the other elements.

Translocation from roots to leaves

The ratio of the elemental concentrations leaves/roots has been calculated to evaluate their mobility and transfer from roots to leaves. Elements such as Ca, Mg, Mn, K and Si are characterized by a ratio equivalent to unity, while concentrations of Al, Ti, Fe and P are higher in the polluted roots than in the equivalent references. The supply of 2-ml of oil to the soil did not change the root to leave segregation of most elements, except for Al and Ti, which contents increased more in the leaves than in the roots (Table 1, Supplementary File).

Supply of 4-ml oil to the soil reduced the transfer of Si, Al, Fe and Ti to leaves and did not affect the other elements, whereas supply of 16-ml of oil increased the translocation of Si, Al, Ti, reduced that of Na, but did not affect the other elements. The impact of the 10-ml oil supply on the roots being laking, the supply of 20-ml of oil decreased the translocation of Si, Na, K, P and Ti from roots to leaves, while increasing that of Fe, not affecting those of Ca and Mg (Figure 3, Supplementary File).

Effect of oil pollution on the trace-elemental uptake

The contents of Cu, Sr, U, Co and Zn were enriched in the roots at the 2-ml contamination level, whereas Cr, Ni, Cu, Sr, Pb, U and Ba were at the 4-ml contamination, and all elements at the 16- and 20-ml contamination levels. The elements not affected by oil pollution are Cr, Sr, Ba, Th and Ni at the 2-ml pollution level, and Zn, Rb, Zr and Th at the 4-ml level. The decreasing elements due to oil pollution are Cd and Rb at the 2-ml level, and Rb, Mo and Cd at the 16-ml, and Cd at 20-ml level (Table 1, Figure 4, Supplementary File).

Soil contamination with 2-ml (0.3%) of crude oil induced an increase of Cr, Co, Ni, Zn, Sr, Mo, Ba and Pb, a decrease of Rb, Zr and Cd, and no change for Th and U in the leaves of the plants. The contamination with 4-ml of oil increased only Zn, Sr and Ba, and decreased Cu, Rb, Cd and Th, whereas Cr, Co and U remained unchanged (Figure 5, Supplementary File). The supply of 10-ml oil to the soil increased the content of all trace elements except unchanged Th and Cd, and increasing Rb. Supply of 16-ml of oil increased Cr, Co and Zn, decreased Rb, Zr, Mo, Cd and Th, and maintained Cu, Sr, Ba, Pb and U unchanged. Supply of 20-ml of oil decreased the content of all trace elements, except unchanged Rb, Cd and Th.

Relative to the radishes from unpolluted substrate, all trace elements were taken up significantly more by the leaves at the 10-ml oil pollution, except Mo that increased at the 2-ml supply. For the roots, higher uptake in trace elements was recorded at the 20-ml pollution, except for Cu that increased most at the 2-ml contamination. In summary, the leaves/roots ratio decreased for most elements at low oil supply (2 and 4 ml) except for Cr, which ratio at the 2-ml supply exceeded the ratio of the reference plants, and for the Zn and Mo ratios that exceed those of the reference plants at the 4-ml oil supply. The supply of 16- and 20-ml of oil decreased the translocation of all trace elements from roots to leaves, except for Cr, Co, Ni and Pb, for which the leave/root ratio exceeded largely the ratio of the reference plants at the 20-ml level supply.

Effect of oil pollution on the rare-earth elemental uptake

The highest cumulative amounts of REEs taken up by the radish roots and leaves were determined in the plants grown in the soil polluted by 10- and 20-ml oil supply. Relative to those grown in the unpolluted substrate, the total change in REEs is not significant for leaves of the radishes from 4 and 16-ml pollution levels (less than 20% change) and for roots of those from 16-ml oil pollution. Also, the differences between total REEs at the successive supplies of oil are less significant for the leaves than for the roots.

Normalization of the REE concentrations from radish of the polluted soils to those from radish of the non-polluted soils outlines flat patterns for the leaves. For the roots from soils polluted by 2, 4 and 16-ml of oil, the patterns are also flat, while those of roots grown in soils with 20-ml oil pollution are enriched in middle REEs (MREEs) with negative Ce and Eu anomalies (Figure 6, Supplementary File).

Size of the viable bacterial population

The total number of bacteria in 1 g of dry-weighted soil represents the whole community of bacteria, being called the total Colony Forming Unit (CFU). The total enumeration of bacteria in the soils where radish was grown is of $112x10^4$ units in the unpolluted reference soil. This CFU almost doubled to $222x10^4$ units in the soil polluted by 2 and 4-ml of oil, while decreasing to 93 and $65x10^4$ units in the soils polluted by 10 and 16-ml oil, respectively. In the soil polluted with the highest concentration of oil (20 ml), the CFU is close to that of the reference soil at $111x10^4$ units (Figure 7, Supplementary File). The most effective concentrations of oil that enhanced the increase of the micro-bacterial density are the 2 and 4-ml supply, whereas the 10 and 16-ml supplies represent an inhibitor amount for the micro-bacterial populations. The highest rate of oil pollution did not change the CFU.

Discussion

Elemental availability in soils depends on their physical and chemical properties, on their micro-organic populations, on the requirements of the grown plants, as well as on the potential uptake by the plant roots. Alternatively, toxic ions present in a soil may affect plants by damaging root cells, leading in turn to a reduction in the elemental availability by inhibition of water and nutrients transport to the root cells. Modification of contaminated rhizosphere can induce a reduction in the biomass and a decrease in the production of ligands that may affect the mobility of elements because of: (1) the occurrence or absence of mobilizing chelator factors, and (2) a modification in the production of reducing or oxidizing agents. As shown above, radishes harvested from soils polluted by 2 and 16-ml of crude oil show no significant ($p > 0.05$) variation in heavy metal concentrations when compared with the reference plants grown in the equivalent non-polluted soil, which is to the opposite to 4 and 10-ml oil pollution that induced an increase of elements, and to the 20-ml pollution that reduced the contents in Si, K, P and Na in the leaves.

In fact, the changing elemental contents in the roots and leaves of the radishes cultivated here are never single trended: the impact of oil supply is not univocal as they increase and decrease when the input of oil pollution increases. For instance, the increase of most of elements in the roots and leaves of radishes from 10-ml oil polluted soil may be due to an important elemental availability and mobilization. On the other hand, the decrease of the total micro-mass relative to the unpolluted soil suggests an intolerance to changing physical properties of the soil polluted at intermediate pollution levels relative: (1) to the low level of pollution at which the total micro-mass increased, and unlike (2) the high level of oil pollution (20-ml) at which the micro-mass remains similar to that of the reference soil. Abosede [18] reported that supply of oil reduces the porosity, aeration and water infiltration capacity of the soil. Crude oil contamination can also increase the soil pH up to 8.0, and reduce available P concentrations [19].

Alternatively, the increase of most elements in the roots and leaves at low pollution level is correlated with an increase in microorganism density, suggesting in turn that increasing element availability in soils can be attributed to an increased organic activity stimulated by the supply of oil. At the intermediate pollution level, the availability of elements exceeds the availability of elements in the unpolluted soil; it is correlated with a decrease in the total content of microorganisms. Such decrease can be attributed to a change in the soil, for instance a reduction of aeration and a change of pH. Acidity in soils can reduce the microbial activity as reported by Ziervogel et al. [20], and enhance the dissolution of minerals [21], which can be the reason of the increased elemental availability at intermediate levels of oil pollution. Njoku et al. [22] reported that crude oil pollution makes soils acidic, Banwart et al. [23] showing that the lower pH in soils are correlated with a greater biological productivity.

The translocation of elements from roots to leaves expressed as a ratio leaves/roots concentrations showed that Si, Mg, Ca, Mn and K are of the same order of magnitude in the variably polluted and unpolluted radishes, whereas elements such as Al, Fe, Ti, Na and P are more accumulated in the polluted roots. Only Mn from equally distributed elements between roots and leaves was affected by oil pollution especially at the 16-ml level, since it was more accumulated in leaves than in roots. Among the other elements, Al and Fe were less translocated from roots to leaves at the 4-ml oil spill relative to the reference plants. At high levels of pollution, Al and Fe contents increased more in the leaves than in the roots, while Na increased more in the roots than in the leaves. Investigation of the capacity of the reference radish to transfer trace elements from roots to leaves indicates that Zr, Rb and Th were more accumulated in the leaves than in the roots. Elements such Cu, Cd and Pb were equally distributed in leaves and roots, while Cr, Co, Ni, Sr, Ba and U were more accumulated in the roots than in the leaves. Supply of oil in the soil increased the translocation of Cr, Ni, Co and Pb from roots to leaves, but reduced those more accumulated in the roots of the reference plants.

Mobility increase of some elements and decrease of others from roots to leaves could result from a competition for the use of the complexing agents. The diverse elemental behavior might also be due to their physiological function in the plant cells. The elements, which mobility increased from the roots to the leaves at a high pollution level may yield specific roles in physiological leave activities, such as in photosynthesis and respiration. Among these elements are Cr and Ni. The stable forms of Cr are the trivalent Cr(III) and the hexavalent Cr(VI) species. With no essential function in plants, Cr can interfere with other elements like Fe and Mo, Shanker et al. [24] reporting that Cr toxicity in plants originate from its action as an oxidizing agent, as well as from formation of free radicals during Cr(VI) to Cr(III) reduction that occurs inside the cells.

Compared to the reference plants, the bacterial count is high at the 2 and 4-ml level of oil pollution, but it decreases at the 10-ml and 16-ml supply, which could indicate that either some toxic elements were released at this level of pollution or the mobility of some elements was reduced. The aeration decrease by oil pollution may also have reduced the bacterial activity. Such impact was not obvious at low level of pollution, but could become significant at the higher 10-ml and 16-ml pollution levels.

Fractionation of the rare-earth elements during uptake

Concentration and distribution of REEs depend on many factors in the plant-soil systems, including mineral composition, physical and chemical parameters (temperature, pH, Eh, chemical composition, ion complexation of the interstitial solutions, porosity, microbial activity, etc) of the soil around the plant roots, as well as the type of plants grown in polluted substrates [25, 26]. Here, the amount of REEs in the plants from substrates variably polluted by 4 and 16-ml oil remains similar to that in the unpolluted plants. Although the leaves collected from plants grown in soils polluted by 2, 10 and 16-ml of oil are enriched in REEs relative to the unpolluted plants the increase is significant at the 10-ml pollution level as for the other elements. There is no direct connection between REE uptake and pollution level. Unlike for the leaves, the total REEs taken up by the roots increased at each level of pollution with an important increase at the 20-ml supply, as for most elements. No relation between REE uptake and reduction in microorganisms from substrate or in ligands is noticeable in the plant segments.

As shown above, the REEs of the leaves yield flat distribution patterns relative to those of the unpolluted reference plants. Those in roots are flat when the level of oil pollution is less than 20 ml. When the level of pollution reaches 20 ml, the REE patterns of the roots are characterized by a decrease in the middle REEs relative to those of the unpolluted reference plants, which can be explained by a probable dissolution of phosphate minerals from initial soil, such as apatite that could not be detected by XRD analysis of the bulk soil, but is suggested by the high P content in the elemental analysis. P minerals are of interest in the study of REE mobility in soils due to their preferential complexation [27, 28]. Among the potential complexation agents that plants use for the transfer of REEs from soil to plants there are organic and inorganic ligands, such as NO_3^-, Cl^-, SO_4^{2-}, PO_4^{3-}, etc …, PO_4^{3-} playing also an important role in REE bio-accumulation processes. Furthermore, plants need also P for their normal development, photosynthesis (ATP/ADP), storage and transfer of energy, and respiration among various other functions. Ding et al., [29] reported that inorganic P in plants is able to raise several hundred times the concentration of PO_4^{3-}in soil solutions with an important interaction between PO_4^{3-} and REEs. Previous studies [29, 30] also reported that REE fractionation causing middle REE (MREE) enrichment in wheat roots results exclusively from phosphate precipitation occurring in or on the roots. Ishikawa et al. [31] also demonstrated the coexistence of REE ions (La^{3+}, Gd^{3+}, and Yb^{3+}) with phosphate in root-type portions of rice and pea, as observed in the roots of maize roots [32].

Conclusions

The main conclusion of this study is that the pollution by increasing amounts of crude oil to a soil is not univocal. In fact, the changing elemental contents in the roots and leaves of the cultivated radishes are never single trended: they variably increase and decrease during a continuously increasing spill of crude oil. In turn, the significant effects of an oil pollution in a soil on which radishes were grown can be summarized by following elemental variations in the plant leaves: (1) Ca, K, P, Mg, Fe and Al behave similarly in the radishes from unpolluted and polluted soils; (2) the intermediate 10-ml oil pollution has the highest impact on the Ca, K, P, Mg and Al contents; (3) the highest uptake is at the high oil pollution for Fe, Mn, Zn, Ni, Co, Cr and Pb; (4) the uptake of the REEs seems not to be affected significantly.

The most important increase of elements occurred in the leaves of the radishes grown in the soil polluted by 10-ml of oil and in the roots of those grown in the soil polluted by 4-ml of oil. The significant effects of oil pollution on the plant roots include: (1) an increased Ca, Fe, Al and Si uptake, often only in the case of the highest pollution, whereas P's uptake decreases; (2) an increase of the microbial population by a factor of about 2.5 at low oil pollution and a dramatic decrease at higher pollution; (3) the uptake of REEs seems only to be affected significantly at the highest degree of oil pollution by an increase of the light REEs.

The translocation roots-to-leaves indicates a decrease of Ca in the leaves when oil pollution increases, while homogeneous for K and P with a slight decrease when pollution is at its maximum. The total biomass increased in the soil at low levels of oil pollution, but decreased at intermediate levels and remains the same at high levels of pollution. The increase of most elements at low level of oil supply is correlated with an increase in microorganism density, which suggests that availability of elements in soils can be attributed to an increase in organic activity, which could have been stimulated by the pollution of oil. At last but not least, the impact of oil pollution is significant and sets serious problems for understanding the transfer of contaminants to the plants.

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Competing Interests

The author declares that they have no competing interests.

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