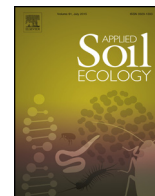




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Viewpoint

Reflections on soil contamination research from a biologist's point of view

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ABSTRACT

Soil contamination is one of the most important threats to soil health. The different aspects treated by scientists dealing with soil contamination are: characterization, impact, remediation, monitoring and prevention. Traditionally, soil contamination research was biased towards Chemistry. Here, from a biologist's point of view, we emphasize the need to incorporate new approaches to soil contamination research by suggesting proposals of research development for these five aspects of soil contamination research, in an attempt to promote discussion on these issues.

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1. Introduction

In 400 B.C., the father of medicine Hippocrates considered the “health of the soil” as a relevant factor to human health (Krupenikov et al., 2011). Since then, many researchers have shown the relationship between the soils' status and human health (Brevik and Sauer, 2015). The term “soil health” refers to the capacity of soil to function as a vital living system to sustain biological productivity, promote environmental quality, and maintain plant and animal health (Doran and Zeiss, 2000). This “health” metaphor provides the opportunity to incorporate well-known medical concepts when disseminating the importance of preserving the health of our soils. Actually, the different aspects treated by scientists dealing with soil contamination, one of the most important threats to soil health, are similar to well-known medical concepts: characterization ≈ clinical analysis; impact ≈ diagnosis; remediation ≈ therapy; monitoring ≈ checkup; and prevention of contamination ≈ disease prevention.

Here, we emphasize the need to incorporate new approaches to soil contamination research by suggesting proposals of research development for these five aspects of soil contamination research.

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2. Characterization: consider biological tools

There is an important need to characterize potentially contaminated soils in order to identify risks for ecosystems and human health. The study of potentially contaminated sites usually involves different phases: past historical activities, orography, hydrogeological study of the area, analysis of soil physicochemical properties (pH, organic matter content, texture, etc.) and, most important, chemical determination of the presence and concentration of inorganic and organic contaminants in a set of selected sampling points. In this respect, most environmental legislations are based on total concentrations of contaminants, which are employed to establish regulatory limits for specific soil uses (agricultural, recreational, urban, industrial, etc.). However, total concentration values have well-known limitations for assessing the adverse impact of soil contaminants (Hooda, 2010). Then, it has been widely proposed to use “bioavailable concentrations” to better assess the impact of soil contaminants (Vasseur et al., 2008). The term “bioavailability” refers to the “contaminant fraction which is freely available to cross an organism's cellular membrane from the medium the organism inhabits at a given point in time” (Semple et al., 2004). A high bioavailability of a contaminant is judged as a negative factor from an environmental point of view; nonetheless, a high value of bioavailability might be positive from other points of view such as, for instance, when dealing with

bioremediation initiatives as it will facilitate the degradation of the contaminant. Besides, other factors not related to the contaminants themselves can actually modify their chemical bioavailability (see below): thus, for instance, the chemotactic movement of bacteria towards some organic compounds increases their actual bioavailability (chemotaxis-enhanced bioavailability), which in turn can have a beneficial effect on their bioremediation when those organic compounds are contaminants (Krell et al., 2013). But the concept of bioavailability is somewhat vague and, above all, it depends on dynamic processes, thus resulting in fluctuating values of bioavailable concentrations. Within the term bioavailability, there is, for instance: (i) *chemical availability*, which depends on physicochemically-driven processes like adsorption, desorption, diffusion, etc. (which, in turn, are controlled by contaminant and soil specific parameters like pH, cation exchange capacity, hydrophobicity, aqueous solubility, clay and organic matter content, and so on); (ii) *biological availability*, which depends on physiological-driven uptake processes controlled by species-specific parameters (e.g., anatomy, surface-volume ratio, habitat preference, feeding strategy); and (iii) *toxicological availability*, which depends on internal allocation processes by organism-specific parameters (e.g., metabolism, detoxification, storage capacity, excretion) (Loibner et al., 2006). This miscellany of concepts can generate confusion, hampering the possibility of incorporating bioavailable concentrations in the regulatory frameworks.

In any case, for the determination of contaminants in potentially contaminated soils, so far, the utilization of chemical techniques has been the norm. Nevertheless, we want to emphasize that biological tools can today play a relevant role in this matter. Interestingly, the relative abundance of the class 1 integron-integrase gene (*int1*) has been reported as a proxy for anthropogenic contamination (i.e., a universal marker of selective pressures imposed by anthropogenic contamination) (Gillings et al., 2015). The *int1* gene is linked to genes conferring resistance to antibiotics, disinfectants and heavy metals, which means that it might be an excellent de facto measure of the general level of resistance to selective agents that are most likely to be present in contaminated areas (Gillings et al., 2015). Then, the relative abundance of *int1* is a priori a good candidate for a biological screening of the presence of contaminants in soil (i.e., like a canary in the coal mine), prior to a chemical characterization for specific contaminants. It must be emphasized that biological parameters have an important advantage over chemical parameters, i.e. biological parameters have an integrative character (Epelde et al., 2009a), overcoming one of the main limitations of chemical characterizations, that is, that direct detection for all contaminants is simply not feasible, as there are some 80,000 different compounds being traded in the marketplace (Rockström et al., 2009) and, besides, the possibilities for synergistic, additive or antagonistic effects are countless.

3. Impact: consider biological interactions

Leonardo Da Vinci stated that “we know more about the movement of celestial bodies than about the soil underfoot”. Centuries later, the soil remains a “black box” (European Commission, 2012). Then, the difficulties of unlocking the secrets of the functioning of the soil ecosystem hinder assessing the impact of soil contaminants.

It is widely accepted that it is not possible to assess the impact of soil contaminants by simply measuring the levels of those contaminants (Ludwig and Iannuzzi, 2005). Such measurements provide information about “contamination” (presence of a substance where it should not be or at concentrations above the natural background level for the area), but they do not provide

information about “pollution” (contamination that causes adverse biological effects on resident organisms) (Chapman, 2007). Then, it is not surprising that some Environmental Risk Assessments include the incorporation of three lines of evidence (TRIAD methodology): chemical, toxicological and ecological (Mesman et al., 2006). After all, for the determination of adverse biological effects, biological measurements (toxicological, ecological) are needed, as chemical measurements of bioavailability (frequently considered as proxies for contaminant toxicity) are nothing but questionable estimations. Therefore, biological tools (bioindicators and biomarkers) are currently being used to directly determine adverse biological effects of soil contaminants (Bartell, 2006). But, during their interpretation, the responses provided by bioindicators and/or biomarkers are traditionally compared to total or bioavailable concentrations of soil contaminants in an attempt to establish “cause-effect” relationships (cause: contaminant concentration; effect: biological response). Here, we want to emphasize that we should also try to interpret biological responses in the light of Biology itself, leaving aside that habit of many of us (i.e., soil biologists) who only evaluate toxicological and ecological data in the light of Chemistry. What is more, we should consider the possibility of setting regulatory limits for contaminated (or remediated) soils from biological data, together with or without chemical data (i.e., contaminant concentrations).

In any event, we must always define which and how many bioindicators/biomarkers need to be measured in order to obtain an appropriate diagnosis of impact. Standardized ecotoxicological assays based on model organisms such as, for instance, *Vibrio fischeri*, *Daphnia magna*, *Eisenia fetida*, *Lactuca sativa*, etc. are frequently performed in contaminated soils. Although they are intended to provide information regarding the impact of contaminants on the soil ecosystem, in most cases, model organisms do not have any ecological relevance and, therefore, their biological responses are not easily extrapolable to resident organisms. In addition, selection pressures at contaminated soils promote the development of tolerance and resistance mechanisms in resident organisms, further complicating the validity of ecotoxicological studies carried out with model organisms. We suggest the combined use of ecotoxicological assays with model organisms and ecological assays with resident organisms, but in both cases covering at least three different trophic levels of the soil foodweb. The soil foodweb includes more than five trophic levels: first level = photosynthesizers; second level = decomposing, mutualists, pathogens, parasites, root-feeders; third level = shredders, predators, grazers; fourth level = higher order predators; fifth and higher level = higher order predators (Rhodes, 2014). When possible, within the same trophic level, it is also desirable to use organisms from different taxa (bacteria, fungi, protozoa, nematodes, mites, etc.) to complete a more ecologically relevant impact assessment. Finally, to obtain an even more ecologically relevant assessment, and since biological interactions are fundamental for the functioning of the soil ecosystem (in real soils, no organism exists in absolute isolation), we propose to study the impact of contaminants on specific biological interactions (e.g., competition, amensalism, antagonism, symbiosis, commensalism, predation, parasitism), overcoming the reductionist approach based on the study of isolated components.

4. Remediation: consider contaminated soil as a valuable resource

Conventional physicochemical methods of soil remediation are usually expensive and frequently have a negative impact on the soil ecosystem (Ali et al., 2013). In particular, the most common remediation method used so far, i.e. excavation and dumping in a controlled landfill, is currently not considered an optimal solution

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