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Ecotoxicological assessment of the potential impact on soil porewater, surface and groundwater from the use of organic wastes as soil amendments

Paula Alvarenga [a,b,*], Clarisse Mourinha [a], Márcia Farto [a], Patrícia Palma [a,c], Joana Sengo [d], Marie-Christine Morais [d], Cristina Cunha-Queda [b,d]

[a] Departamento de Tecnologias e Ciências Aplicadas, Escola Superior Agrária, Instituto Politécnico de Beja, Rua Pedro Soares S/N, Apartado 6155, 7800-295 Beja, Portugal
[b] LEAF – Centro de Investigação em Agronomia, Alimentos, Ambiente e Paisagem, Instituto Superior de Agronomia, Universidade de Lisboa, Tapada da Ajuda, 1349-017 Lisboa, Portugal
[c] CIMA-Centro de Investigação Marinha e Ambiental, CIMA, FCT, Edifício 7, Piso 1, Universidade do Algarve, Campus Universitário de Gambelas, 8005-139 Faro, Portugal
[d] Instituto Superior de Agronomia, Universidade de Lisboa, Tapada da Ajuda, 1349-017 Lisboa, Portugal

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ABSTRACT

This study aimed to assess the potential impact on soil porewater, surface and groundwater from the beneficial application of organic wastes to soil, using their eluates and acute bioassays with aquatic organisms and plants: luminescence inhibition of *Vibrio fischeri* (15 and 30 min), *Daphnia magna* immobilization (48 h), *Thamnocephalus platyurus* survival (24 h), and seed germination of *Lolium perenne* (7 d) and *Lactuca sativa* (5 d). Some organic wastes' eluates promoted high toxic responses, but that toxicity could not be predicted by their chemical characterization, which is compulsory by regulatory documents. In fact, when organisms were exposed to the water-extractable chemical compounds of the organic wastes, the toxic responses were more connected to the degree of stabilization of the organic wastes, or to the treatment used to achieve that stabilization, than to their contaminant load. That is why the environmental risk assessment of the use of organic wastes as soil amendments should integrate bioassays with eluates, in order to correctly evaluate the effects of the most bioavailable fraction of all the chemical compounds, which can be difficult to predict from the characterization required in regulatory documents. According to our results, some rapid and standardized acute bioassays can be suggested to integrate a Tier 1 ecotoxicological evaluation of organic wastes with potential to be land applied, namely luminescence inhibition of *V. fischeri*, *D. magna* immobilization, and the germination of *L. perenne* and *L. sativa*.

1. Introduction

The beneficial use in agricultural soils of organic wastes produced in different human activities is very important, not only because it can reduce their landfill disposal, contributing to the end-of-waste policy and recycling targets in Europe (Council Directive 1999/31/EC, 1999; Saveyn and Eder, 2014), but also because it can enhance soil fertility. In fact, in countries from the European semi-arid Mediterranean region, soils have very low topsoil organic matter content and are very prone to erosion (European Commission, 2012). To overcome this problem, the use of organic wastes as soil amendments seems an attractive option, because it would: augment soil organic C storage, recycle valuable nutrients (e.g. N, P, K), promote stable soil aggregation, improve soil aeration, and augment cation exchange and water holding capacities (Larchevêque et al., 2006; Diacono and Montemuro, 2010; Mattana et al., 2014). However, these organic wastes are potentially contaminated with undesired substances (e.g. heavy metals, organic pollutants and pathogens), which, by this practice, can reach different environmental compartments, like soil, surface and groundwaters, and, ultimately, have major impacts on human health, climate change, biodiversity, and food safety (COM, 2006).

Through the Sewage Sludge Directive 86/278/EEC, which

* Corresponding author at: Department of Applied Sciences and Technologies, Escola Superior Agrária-Instituto Politécnico de Beja, Rua Pedro Soares S/N, Apartado 6155, 7800-295 Beja, Portugal.
E-mail address: paula.alvarenga@ipbeja.pt (P. Alvarenga).

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primary aim was to protect the environment from the sewage sludge heavy metal load, European countries have acknowledged that concern and, nowadays, several of them, like Portugal, have evolved to more updated legislation, which includes organic contaminants and pathogenic microorganisms (Kapanen et al., 2013; Decree-Law No. 276/2009, 2009). However, apart from the legislated contaminants that are identified and frequently analyzed, sewage sludges, and other organic wastes, may also contain other toxic compounds, which are not identified and, most certainly, not measured in a routine basis (Malara and Oleszczuk, 2013; Kapanen et al., 2013). The identification and measurement of these chemicals often requires costly equipment, available only at a limited number of laboratories, and the exhaustive identification and quantification of all potentially toxic compounds is impractical (Wilke et al., 2008; Malara and Oleszczuk, 2013). Still, even if those chemical compounds were measured, chemical analysis does not provide direct information of their biological effects (Ramírez et al., 2008). So, despite the importance of chemical parameters to assess the risk of the application of organic wastes to soil, chemical information must be complemented with ecotoxicological tests, since bioassays integrate the effects of all contaminants, including additive, antagonistic and synergistic, and are sensitive to the bioavailable fraction of contaminants (Wilke et al., 2008). In fact, often there is no correlation between the contamination of organic wastes and their effective ecotoxicological effects on the environment. Therefore, ecotoxicological testing may be a pragmatic approach for assessing the intrinsic toxicity of wastes as a complement to chemical analysis. For instance, Roig et al. (2012) concluded, as other authors in previous studies (Domene et al., 2007; 2008a; Ramírez et al., 2008), that organic waste stability, conditioned by their treatment, might have a greater influence on ecotoxicity than its pollutant load. That is why some of these studies have emphasized the need for the stabilization of organic wastes prior to their application to soil, which could lower their toxicity (Domene et al., 2007; Roig et al., 2012; Kapanen et al., 2013; Chen et al., 2014). So it is important to gather knowledge in this area, in order to sensitize and pressure the stakeholders to the beneficial recycling of organic wastes to agricultural soils, which can only be achieved with uncontaminated and properly stabilized materials.

The ecotoxicological tests can be used to assess the effects of the organic wastes application on soil “habitat function” or “retention function”. The effects on soil “habitat function” are usually evaluated using “the whole material”, in bioassays with terrestrial organisms (Domene et al., 2007; Moreira et al., 2008; Natal-da-Luz et al., 2009a; 2009b). On the other hand, if we want to assess the effects on the quality of porewater, surface and/or groundwater, which can be affected through leaching or runoff of contaminants, *i.e.* the effects on soil “retention function”, organic wastes' eluates and bioassays with aquatic organisms should be used (Roig et al., 2012, Malara and Oleszczuk, 2013).

There are several published works on the selection of bioassays, using the organic waste or its extracts, for the ecotoxicological characterization of wastes (Rojícková-Padrtová et al., 1998; Pandard et al., 2006; Wilke et al., 2008; Pablos et al., 2009; Huguier et al., 2015). Some of these studies were developed under the framework of the Hazardous Waste Council Directive 91/689/EEC, 1991 which defined a set of 14 properties for the hazardous classification of a waste, being Hazard 14 (H14), or “ecotoxic” property, created without referencing specific methods (Pandard et al., 2006; Wilke et al., 2008; Pablos et al., 2009). Other studies were more specifically devoted to the evaluation of organic wastes, intending their beneficial use in agricultural soils (Alvarenga et al., 2007; Domene et al., 2007, 2008a, b; Moreira et al., 2008; Ramírez et al., 2008; Natal-da-Luz et al., 2009a; 2009b; Roig et al., 2012; Kapanen et al., 2013; Huguier et al., 2015). From all these studies, one conclusion is consensual, a battery of bioassays, representative of different trophic levels, should be considered. It is also quite consensual that solid-phase bioassays, using the material as a whole, provide a more relevant evaluation of its toxicity, as it relates to the potential exposure conditions and can mimic the “in situ” interactions between the material and the exposed organisms (Alvarenga et al., 2007; Domene et al., 2008a; Huguier et al., 2015). However, Kapanen et al. (2013) have alerted on one important problem, biotests have limitations in assessing the ecotoxicity of test media rich in organic matter, such as sewage sludge and compost. This fact makes the bioassays using the waste water-extract, which are more suitable to assess the toxicity of the contaminants bioavailable fraction, important in a complete ecotoxicity assessment of an organic waste (Loureiro et al., 2005). In fact, when an organic waste is intended to be land applied, it is important to consider not only the interactions between the contaminant and the soil matrix, which can affect soil organisms, but also the contaminants that may be leached, and could affect surface and groundwater systems (Alvarenga et al., 2007).

Taking all this into account, the aims of this study were: (i) to assess the potential impact on porewater, surface and groundwater from the use of organic wastes as soil improvers, using a battery of bioassays, with organisms representative of different taxonomic groups, and the water-extracts of nine different organic wastes, representative of a broad range of organic wastes, and (ii) to contribute to the selection of the most appropriate bioassays to be used in the Tier 1 ecotoxicological evaluation of organic wastes with potential to be land applied.

The bioassays selected are quite well standardized and commonly used: inhibition of light emission of *Vibrio fischeri* (15 and 30 min), *Daphnia magna* immobilization (48 h), and *Thamnocephalus platyurus* survival (24 h). A monocotyledonous, ryegrass (*Lolium perenne* L.), and a dicotyledonous, lettuce (*Lactuca sativa* L.), common as forage crops and recommended by OECD guideline 208A for ecotoxicological testing (OECD, 1984), were used to assess phytotoxic effects of the water-extracts on seed germination and root elongation.

The results reported here are intended to contribute to the risk evaluation of organic wastes application to soil, in an ongoing project entitled: “ResOrgRisk – Environmental risk assessment of the use of organic residues as soil amendments” PTDC/AAC-AMB/119273/2010, financed by “Fundação para a Ciência e Tecnologia” (FCT).

2. Materials and methods

2.1. Organic wastes

Nine different organic wastes were collected at different locations in Portugal: two samples of untreated dewatered municipal sewage sludge (SS1 and SS2); an agro industrial sludge from the wastewater treatment plant of a big winery (AIS); a municipal slaughterhouse sludge (MSS); a mixed municipal solid waste compost (MMSWC); an agricultural wastes compost (AWC), produced from 21% olive mill waste, 61% sheep manure, 10% olive leaves, and 8% meat flour, fresh mass ratio; a compost produced from agricultural wastes and sewage sludges (AWSSC), with 30% sewage sludge, 25% agricultural wastes and 45% woody materials, fresh mass ratio; a pig slurry digestate (PSD); and wastes from pulp and paper mill (PMW), with cellulosic sludges and ashes (primary sludge, biological sludge, and ash from the biomass combustion, 1:1:1 fresh mass ratio).

The organic wastes thorough description, characteristics and the possibility of their use in agricultural soils, taking in account their chemical composition, total heavy metal concentrations,