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Disposal of dredged sediments in tropical soils: ecotoxicological evaluation based on bioassays with springtails and enchytraeids

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Abstract Metal reference values established in Brazilian legislation for terrestrial disposal of dredged sediments and soil quality were derived for temperate regions. To evaluate the adequacy of such metal reference values to tropical soils, the ecotoxicity of a dredged sediment (from the Guanabara bay, Rio de Janeiro, Brazil) was investigated in two local soils (ferralsol and chernosol) by performing avoidance and reproduction tests using *Folsomia candida* and *Enchytraeus crypticus*. Test doses consisted of 0 %, 1.25 %, 2.5 %, 5, 10 %, and 20 %. Total and potentially bioavailable metal concentrations were determined in the test mixtures. Although the chernosol mixtures had the highest total metal concentrations, the influence of the expansive clay minerals (with high ability to adsorb metals) and the high contents of nutrients typical from this type of soils, seem to reduce the ecotoxicity. Collembolan avoidance behavior was the least sensitive endpoint. The lowest sediment doses increased the reproduction of *F. candida* in ferralsol mixtures. *E. crypticus* reproduction in the ferralsol mixtures were more pronounced

at lower concentrations than in chernosol mixtures. Possibly the low nutrient content of the ferralsols, in connection with the addition of small amounts of sediment, created particular conditions that promoted reproduction of the test species. Data obtained in the ecotoxicological tests may support the establishment of a “safe” ecological dose of dredged sediments to be applied in tropical soils, supporting decision-makers in programs of environmental management.

Keywords Guanabara bay · Metal contamination · Ferralsol · Chernosol · Ecotoxicity · Soil

Introduction

The growing industrial activity and the consequent development of urban areas in tropical countries have generated high amounts of contaminated wastes. Most of such wastes have been discharged into aquatic systems over the last decades. Because of this, nowadays, Brazil contains many aquatic systems contaminated with metals, petroleum hydrocarbons, and pathogenic microorganisms (Lutterbach et al. 2001; Lacerda and Gonçalves 2001; Hortellani et al. 2005; Covelli et al. 2012). The regular dredging of waterways and rivers is usually disposed in soils from adjacent areas as organic amendment. This practice contributes to contaminate local soils, reducing biodiversity of soil dwelling organisms, and consequently, compromising soil productivity. This reality has been demonstrated under standard conditions with key-soil species in laboratory tests (Cesar et al. 2014a).

It is well-known that physical and pedogeochemical parameters (e.g., texture, pH, organic matter, clay minerals, iron and aluminum oxi-hydroxides, sulfides and chlorides) can have a strong influence on the mobility, bioavailability and ecotoxicity of metals for soil biota (Van Gestel and Hoogerwerf 2001; Van Gestel and Mol 2003; Natal-da-Luz

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et al. 2008; Chelinho et al. 2011). Different sources of pollution, associated with distinct chemical forms in the environment, can also play an important role in the toxicity of contaminants (Katz and Salem 1993; Yallouz et al. 2008). In this respect, there is a lack of studies regarding the role of tropical soil properties in the availability of toxicants for edaphic fauna. Moreover, in Brazil, the legislation for terrestrial disposal of dredged sediments are based on Canadian protocols (CONAMA 2004), whose concentrations for metals were estimated for temperate regions, and consequently, do not reflect the dynamic and the particular characteristics of tropical ecosystems. The characterization of the ecotoxicity associated with its disposal onto soils from surrounding areas became a necessity in order to decrease costs of the dredging activities. The establishment of “safe” ecological doses of sediment to be applied in soil and the identification of the most adequate areas to receive those sediments are often required to support decision-makers in activities of environmental management and public health (Munns et al. 2002).

The bioavailability of metals in soils have been investigated through extractions using diluted salts or diluted acids (e.g., CaCl_2 and HCl ; DePaula and Mozeto 2001; Kobeticova et al. 2011). Sequential extractions methods have been also applied to describe the mobility of potentially bioavailable geochemical fractions (Tessier et al. 1979; Gleyzes et al. 2002; Cesar et al. 2011). Even though such analytical perspectives are extremely relevant, they do not take into account the effect of multi-contaminant interactions, the presence of contaminants that are not included in the chemical analyses and the effect of other factors, like the influence of soil properties, on the contaminants toxicity. Chemical analyses should be complemented with data describing the effects of contaminants as a whole to soil organisms. In this respect, ecotoxicological assays are promising since they may provide data taking into account the effect of the whole sediment within local soils, allowing the identification of potentially toxic doses to soil biota.

Several studies have been conducted to evaluate the potential harmful effects of organic wastes to soil fauna. Bioassays using representatives of soil organisms, such as collembolans (*Folsomia candida*), enchytraeids (*Enchytraeus crypticus/albidus*), earthworms (*Eisenia foetida/andrei*), nematodes (*Caenorhabditis elegans*), and predatory mites (*Hypoaspis aculeifer*), have been performed to evaluate the ecotoxicity associated to the disposal of domestic wastes to soils (Crouau et al. 1999; Peredney and Williams 2000; Bakker et al. 2003; Fountain and Hopkin 2004; Natal-da-Luz et al. 2008; Chelinho et al. 2011; Van Gestel et al. 2011). However, the establishment of the most adequate bioassays to characterize the ecotoxicity of dredged sediments is still needed, especially taking into consideration the particular properties of soils from tropical regions. Two recent studies evaluated the ecotoxicity of a metal-contaminated dredged sediment obtained at the

Guanabara Bay (Rio de Janeiro Municipality, Brazil). This evaluation comprised a preliminary assessment composed by earthworm acute and avoidance tests using two natural tropical soils (Cesar et al. 2014a) followed by a deeper ecotoxicological evaluation comprising collembola avoidance and feeding inhibition tests and chronic tests with earthworms, collembolan, and potworms using artificial soil. The preliminary evaluation (Cesar et al. 2014a) highlighted the importance of natural properties of local soils to the toxicity of dredged sediments and suggested that the threshold limits established by Brazilian law are not adequate to prevent noxious effects on local soil systems. The assessment with artificial soil reinforced the need for an ecotoxicological evaluation of dredged sediments to complement chemical analyses required by Brazilian law.

The present work aims to evaluate the ecotoxicity of the same dredged sediment using two tropical soils (representative of two soil classes) that were used in the previous preliminary ecotoxicological evaluation (Cesar et al. 2014a) to prepare gradients of sediment doses to be used in bioassays with *E. crypticus* and *F. candida*. Our working hypothesis are as follows: (a) the ecotoxicity of dredged sediments is reduced in soils containing expansive clay minerals and (b) the dredged sediment increase the reproduction of soil fauna at the lowest doses due to its high organic matter content, independently of the test soil.

Materials and methods

Sample processing

Samples of dredged sediment were collected at three points (station 1: 22° 51' 46.77"S, 43° 14' 0.92"E; station 2: 22° 51' 41.63"S, 43° 14' 10.90"E; station 3: 22° 50' 48.28"S, 43° 14' 29.48"E) at the Cunha estuary from the Guanabara bay basin (Rio de Janeiro Municipality, RJ, Brazil). Sediment samples were dried at 20 °C and then mixed, homogenized, and grounded, in order to generate a composite sample of sediment. Cesar et al. (2014a) measured total metal concentrations in the same composite sample and found metal concentrations generally higher than those established in the level three (the limit of low probability of adverse effects on biota) of Brazilian legislation for terrestrial disposal of dredged sediments (Table 1). Organic matter content was 19.1 % (Cesar et al. 2014a).

Ferralsol and chernosol were collected at the B horizon of a field in the Rio de Janeiro State (*chernosol*: 22° 51' 22.5" S, 43° 30' 0.7" W; *ferralsol*: 22° 41' 34.2" S, 43° 17' 14.5" W). Samples were taken using a soil auger. Such soil classes (ferralsol and chernosol) were selected due to their antagonistic physical, chemical and mineralogical properties, which allow evaluating the role of distinct soil properties in the ecotoxicity and metal availability of the test sediment to soil biota. Soils

Table 1 Total metal concentrations of the dredged sediment studied and the upper limit values of metals allowed for dredged sediments to be disposed in soil according to CONAMA (2004)

Metal	Dredged sediment (mg/kg)	Limit values (mg/kg)	
		Level three ^a	Level four ^b
Hg	1.08	0.15	0.71
Cu	92.0	34	270
Zn	329	150	410
Pb	124	46.7	218
Ni	20.3	20.9	51.6
Cr	94.5	81	370

Table taken from Cesar et al. (2014a)

^a Limit of low probability of adverse effects on biota^b Limit of high probability of adverse effects on biota (CONAMA 2004)

were dried at 20 °C and then sieved at 1.7 mm to remove large particles. Physical, chemical, and mineralogical properties of these soils were measured by Cesar et al. (2012) and Alamino et al. (2007) and are shown in Table 2. Ferralsols are typically acidic soils, with high contents of iron and aluminum, low fertility, and its clay mineralogy is mainly composed by kaolinite, gibbsite, and goethite. This type of soils is spatially well-distributed in tropical regions, and it is the most abundant soil type in Brazil. Chernosols are usually less acidic soils, with high content of nutrients (high natural fertility) and its mineralogy is essentially composed by expansive clay minerals (illite, interstratified illite-smectite, and vermiculite) and kaolinite. These soils are widely distributed in tropical and sub-tropical regions (e.g., southern Brazil). Both sites with ferralsols and chernosols are extensively used for agriculture.

Table 2 Physical and chemical properties of ferralsol and chernosol used in bioassays

Soil parameters	Ferralsol	Chernosol	
Texture (% , n=3)	Clay	58	24
	Silt	6	35
	Sand	36	41
pH (n=3)	H ₂ O	4.2	6.2
	KCl	3.6	4.1
Sortion complex (cmol _c /dm ³ , n=3)	Mg ⁺² +Ca ⁺²	0.2	40.5
	K ⁺	0.02	0.03
	Na ⁺	0.03	1.44
	CEC	2.3	48.3
Total concentrations (% , n=3)	Al	26.4	18.9
	Fe	11.3	7.9
Organic matter (% , n=3)		0.22	0.34

Data are taken from Alamino et al. (2007) and Cesar et al. (2012)

CEC cation exchange capacity

Ecotoxicological tests

The definition of the sediment doses prepared to the bioassays was based on results obtained in previous experiments (Cesar et al. 2014a). The sediment doses tested were: 0 %, 2.5 %, 5 %, 10 %, and 20 %. Both springtails and enchytraeids used in the experiments were originated from laboratory cultures and maintained as described by Natal-da-Luz et al. (2008) and Natal-da-Luz et al. (2009), respectively. Bioassays were performed at 20±2 °C under a photoperiod of 16:8 h light: dark. The moisture of the test mixtures was adjusted to 50±10 % of the water-holding capacity, before being used in the experiments. The pH and the moisture of the substrates were determined at the beginning and at the end of the tests.

Bioassays with *F. candida*

Only springtails 10–12 days old taken from synchronized cultures were used in the tests. Missing adult organisms at the end of the tests were considered dead.

Two-chamber avoidance tests were performed following the ISO 17512-2 (ISO 2007). Each replicate consisted of cylindrical plastic vessels (11 cm diameter and 4 cm height) divided into two equal sections, using a plastic card introduced transversally. One section was filled with 30 g (fresh weight equivalent; FW) of uncontaminated natural soil and the other was filled with uncontaminated soil (dual-control test) or dredged sediment-amended soils (avoidance tests). The uncontaminated soils were tested against each sediment dilution with the same soil, and five replicates were prepared per each combination. After filling both sections, the plastic card was removed and 20 organisms were placed on the middle line between the soils. After 48 h, the card divider was reintroduced in the same position and the content of each section was emptied into different vessels. These vessels were filled with water and a few drops of blue ink were added. After gently stirring, the individuals floating on the water surface were counted. An additional container without individuals was prepared for each combination, for pH and moisture determination at the end of test. Validity criteria assumed a mortality lower than 20 % and a distribution of 40–60 % in dual-control tests (uncontaminated soil in both sections of the replicates).

Collembola reproduction tests were performed according to the ISO 11267 (ISO 1999). Replicates consisted of cylindrical glass containers (4 cm diameter and 7 cm height) filled with 30 g of test mixture (FW) and 10 organisms. An additional test mixture of 1.25 % of dredged sediment in ferralsol and chernosol was included in the gradients used in the tests and pure artificial soil (composed by 70 % of quartz sand, 20 % of kaolin, 10 % of ground Sphagnum peat and 0.5 % to 1 % of calcium carbonate) was tested to check the good condition of the test organisms. Five replicates were prepared

per treatment and about 2 mg of dry granulated yeast was added in each as food at the beginning and after 14 days of test. The test containers were opened once a week to allow aeration and to adjust moisture content (compensating the weight losses of test vessels by adding distilled water). After 28 days of exposure, each test container was emptied into a small vessel that was filled with water. After the addition of a few drops of blue ink and gently stirring, the animals floating on the water surface were photographed and counted using the Image Tool software (<http://ddsdx.uthscsa.edu/dig/itdesc.html>). An additional replicate per test treatment, but without organisms, was prepared and subjected to the same conditions to allow pH and moisture measurements at the end of the test. Validity criteria assumed mortality lower than 20 %, a coefficient of variance of reproduction no higher than 30 % and more than 100 juveniles per test container in replicates with artificial soil.

Bioassays with *E. crypticus*

The procedures adopted in the enchytraeids reproduction tests were based on the ISO 16387 (ISO 2004). Cylindrical glass containers (5 cm diameter and 9 cm height) with twenty grams of test mixture (dry weight equivalent; DW) and 10 adult organisms (with a visible well-developed clitellum) were used per replicate. As for collembola reproduction tests, replicates with artificial soil were prepared in addition to the gradient of sediment dilutions with natural soil to attest the good condition of the test organisms. About 15 mg of grounded rolled oats were added in each replicate as food at the beginning of the test. The test containers were opened once a week to allow aeration, adjust moisture content (compensating the weight losses of the replicates by adding distilled water) and add more food when necessary. After 28 days, each replicate was filled with ethanol (at 70 %) up to a depth of 4 cm plus 200–300 µL of Bengal red (1 % solution in ethanol), gently stirred, left to rest overnight, washed through a sieve (0.25 mm) with tap water, and transferred to a Petri dish to count juveniles under a binocular microscope. Validity criteria assumed a number of juveniles higher than 25 per test vessel and a coefficient of variation no higher than 50 % in replicates with artificial soil.

Metal determination

Total concentrations of zinc (Zn), copper (Cu), chromium (Cr), nickel (Ni), and lead (Pb) were quantified in triplicate using 80–100 mg of pre-homogenized test mixture. The acid extraction of the samples was performed using 2 ml of 69 % HNO₃ solution in a PDS-6 pressure digestion systems at 150 °C for 10 h, as described by Natal-da-Luz et al. (2011). The quality of this analysis was checked using SRM 2709 (San Joaquin Soil—Standard Reference Material) certified by the National Institute of Standards and Technology

(Department of Commerce, USA) as reference material. The potentially bioavailable concentration of the same metals was performed in triplicate using one gram of sample and 25 mL of a 0.1-mol/L HCl solution according to the methodology proposed by DePaula and Mozeto (2001). Metal concentrations in the analytical solutions were determined by flame AAS (2380 Absorption Atomic Spectrometer, Perkin-Elmer). The detection limits were 0.04, 0.1, 0.0012, 0.06, and 0.05 mg/kg, for Cu, Pb, Zn, Ni, and Cr, respectively. For both total and potentially bioavailable metal concentrations replicate blanks were prepared.

Statistical analysis

The Fisher's exact test was used to evaluate significant differences in collembolan avoidance responses as described by Natal-da-Luz et al. (2009). Limited habitat function was attributed to the test mixtures that provoked an avoidance behavior higher than 70 %. The sediment doses that provoked 50 % of mortality (in reproduction tests only) and avoidance behavior (LC50 and EC50, respectively) and the respective 95 % confidence limits (CL) were computed using the Priprobit 1.63 software (Sakuma 1998; <http://bru.gmpcr.ksu.edu/proj/priprobit/download.asp>). Non-linear regression models (exponential, Gompertz and Homersis models) were used to estimate the reproduction EC50 values and respective 95 % CL. Differences in reproduction were evaluated by one-way analysis of variance (ANOVA) followed by Dunnett's tests, to test for significant differences between the control (uncontaminated soils) and the test mixtures. The treatments where the reproductive output was lower than 1 % of the control replicates were not considered in one-way ANOVAs. The collembolan reproductive output in uncontaminated natural soils was compared by *t* test independent samples. For non-linear regressions, one-way ANOVAs and *t* test, normality and homogeneity of data were checked before performing statistical analysis using Kolmogorov–Smirnov and Levene's tests. These statistical analyses were performed using STATISTICA version 7.

Results

Bioassays with *F. candida*

The validity criteria of collembolan avoidance tests were fulfilled. Dual-control tests of ferralsol and chernosol showed distributions of 48.5–51.5 % and 56.5–43.5 %, respectively, and no mortality was observed in both tests. No mortality was found at the end of the avoidance tests in all combinations tested. A significantly higher number of organisms were found in the 2.5 % sediment dose of ferralsol mixtures. Significant avoidance behavior towards the sediment mixtures

occurred since the 5 % sediment dose in both ferralsol and chernosol mixtures ($EC_{50}=4.09\%$ and 16.71% , respectively; Table 3). In both ferralsol and chernosol treatments, the habit function was limited (avoidance $>70\%$) in the highest sediment dose (20 %; Fig. 1).

In the reproduction tests, the validity criteria were fulfilled (data from artificial soil not shown). The percentage of surviving adults in the chernosol mixtures was higher than 80 % up to the 2.5 % sediment dose, above which the surviving percentage decreased to about 70 %. These data did not allow the estimation of the LC_{50} value. Collembola reproduction significantly decreased (one-way ANOVA, $F_{(5,24)}=36.47$; $p<0.001$) since the 2.5 % chernosol mixture and a reproduction EC_{50} value of 9.52 % was estimated (Fig. 2, left graph; Table 3). In the ferralsol treatments, the surviving percentage was about 60 % until the highest concentration (20 %) where the surviving decreased to 20 % ($LC_{50}=8.73\%$; Table 3). Collembola reproduction was significantly higher than control at the 1.25 and 2.5 % doses (one-way ANOVA, $F_{(5,24)}=35.42$; $p<0.001$) and significantly lower at the 20 % dose (one-way ANOVA, $F_{(5,24)}=42.36$; $p<0.001$). These data did not allow the estimation of the reproduction EC_{50} value (Fig. 2, right graph). The reproductive outcome in the replicates with uncontaminated soils (chernosol versus ferralsol) was significantly higher in the chernosol rather than in the ferralsol (t test, $p<0.001$).

Bioassays with *E. crypticus*

The validity criteria were fulfilled (data from artificial soil not shown). In chernosol mixtures the reproduction of echytraeids was significantly higher than the control (0 % dose) at the 2.5 and 5 % doses and significantly lower at the 20 % dose (one-way ANOVA, $F_{(5, 24)}=35.32$; $p<0.001$; Fig. 3, left graph). The reproduction EC_{50} value estimated for these mixtures was 11.96 % (Table 3). The mixtures with ferralsol significantly increased enchytraeids reproduction at the 2.5 % dose

Table 3 Avoidance EC_{50} and reproduction LC_{50} and EC_{50} values (with corresponding 95 % confidence intervals) of *Folsomia candida* and *Enchytraeus crypticus* exposed to mixtures of chernosol or ferralsol with a dredged sediment

		Chernosol	Ferralsol
<i>Folsomia candida</i>			
Avoidance tests	EC_{50}	16.71 (–) ^a	4.09 (2.18–5.73)
Reproduction tests	LC_{50}	– ^b	8.73 (–) ^a
	EC_{50}	9.52 (6.08–12.97)	– ^b
<i>Enchytraeus crypticus</i>			
Reproduction tests	EC_{50}	11.96 (8.68–15.21)	– ^b

Toxic values are expressed in percentage

^a Data does not allow estimation of a 95 % confidence interval

^b Data does not allow the estimation of the toxic value

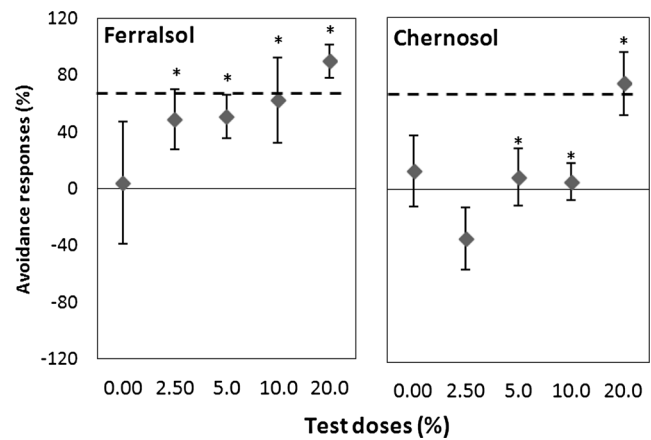


Fig. 1 Percentage of avoidance behavior $([No\ control - No\ test]/[No\ control + No\ tests] \times 100)$ of *Folsomia candida* (average \pm standard deviation, $n=5$) on a ferralsol or a chernosol against mixtures of these soils with a dredged sediment in different doses. Percentages of the test section below the dashed arrow means limited habitat function of the test mixture. Asterisk indicates avoidance behavior significantly different from control ($p \leq 0.05$)

and significantly decreased at the 10 and 20 % doses (one-way ANOVA, $F_{(5, 24)}=42.24$; $p<0.001$; Fig. 3, right graph). The reproductive outcome in the replicates with uncontaminated soils was significantly higher in chernosol rather than in ferralsol (t test, $p<0.001$).

Metal concentrations

Total metal concentrations measured in the mixtures of chernosol were generally higher than those measured in the mixtures of ferralsol within the same sediment dose. These values were generally between the prevention and the intervention values established by Brazilian legislation for soil quality (CETESB 2005; Table 4). The exceptions were in the ferralsol mixtures for Cu where concentrations were below the reference values for all doses and for Zn with concentrations higher than the intervention values for the 20 % dose. In chernosol mixtures, the exceptions were for Cu, where the concentrations were between the reference and the prevention values for concentrations above 2.5 %, and for Pb and Zn with concentrations higher than the intervention values only for the 20 % dose (Table 4).

Potentially bioavailable metal concentrations were generally lower in chernosol mixtures comparing to ferralsol mixtures. Zinc was the metal with the highest percentage of bioavailable concentrations in mixtures of both natural soils rounding 50 % of the total concentration in few doses (Table 5).

Discussion

Although total metal concentrations measured in the chernosol mixtures were generally higher than those

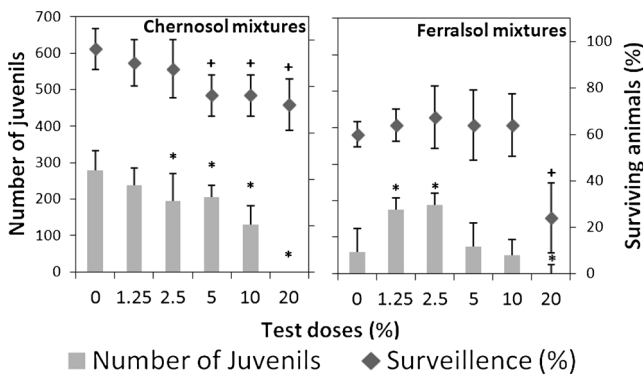


Fig. 2 Reproduction tests with *Folsomia candida*. Reproduction (bars, left scale) and number of surviving *F. candida* (squares, right scale; average±standard deviation, $n=5$) when exposed to mixtures of a dredged sediment in ferralsol (right graph) or chernosol (left graph). Asterisk indicates reproduction significantly different from control ($p \leq 0.05$). Plus sign indicates surviving organisms significantly different from control ($p \leq 0.05$)

determined in ferralsol treatments, all the endpoints measured indicate the highest levels of ecotoxicity for the ferralsol mixtures. Potentially bioavailable metal concentrations were lower in chernosol treatments compared to those of ferralsol, which is in agreement with the lower levels of toxicity observed in chernosol mixtures. Apparently, soil properties played a crucial role in the toxicity of the dredged sediment to the test organisms. Most probably this occurred due to the presence of expansive clay minerals in the chernosol (as indicated in the section 2.1), which confirms the first working hypothesis. Expansive clay minerals are well known by its capacity to adsorb metallic cations from the soil solution due to its high cation exchange capacity, which may explain the low metal concentrations in soil solution of chernosol mixtures and, consequently, the low toxicity observed. In fact, some authors have reported the capacity of expansive clay minerals of decreasing the ecotoxicity of metals and organic substances to microorganisms (Babich and Stotzky 1977), earthworms (*Eisenia andrei*; Cesar et al. 2012) and plants (*Triticum aestivum* and *Lepidium sativum*; Matzke et al.

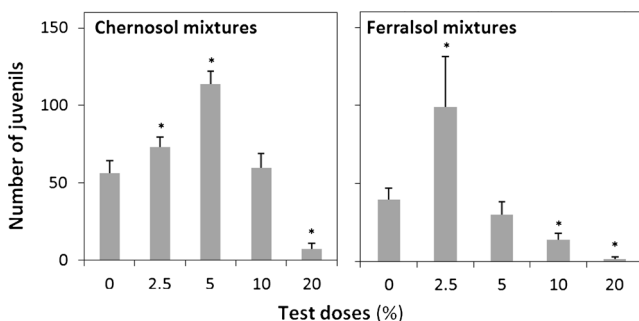


Fig. 3 Reproduction tests with *Enchytraeus crypticus*. Reproduction of *E. crypticus* (average±standard deviation, $n=5$) when exposed to mixtures of a dredged sediment in ferralsol or chernosol. Asterisk indicates reproduction significantly different from control ($p \leq 0.05$)

2008). An ecotoxicological evaluation of the same dredged sediment used in the present study was recently performed using standard artificial soil to perform a battery of ecotoxicological tests including collembolan avoidance tests and collembolan and enchytraeids reproduction tests. The toxic values estimated from the data obtained in those tests (*F. candida*: avoidance $EC_{50}=20.28$ %, reproduction $EC_{50}=7.72$ %; *E. crypticus*: reproduction $EC_{50}=10.10$ %) were generally in the same order of magnitude of the toxic values obtained in the present study for the same ecotoxicological tests (exception only for collembolan avoidance tests; Table 3). Given the higher organic matter content of the artificial soil (about 10 %) compared to that of local soils (0.22 and 0.34 % for ferralsol and chernosol, respectively), data obtained in the present study lead to suppose that the organic matter content of soils is not determinant to predict toxicity of dredged sediments. This fact highlights the need of using local soils in the ecotoxicological evaluation of organic wastes to be applied in soils.

Collembolan avoidance tests showed lower sensitivity compared to the reproduction tests in ferralsol mixtures. This agrees with the results obtained in a recently study performed with artificial soils and the same dredged sediment, which also reported a lower sensitivity of avoidance tests compared to collembolan and enchytraeids reproduction tests. As hypothesized by the authors, it is probable that the high organic matter content of the test sediment had mitigated the avoidance behavior towards the test mixtures. The influence of organic matter of test mixtures in the avoidance response was clearly observed in the combination of the lowest sediment dose of chernosol mixtures (2.5 %), where the highest number of organisms was found in the section with the sediment mixture. Apparently, particularly in this combination, the higher organic matter content of the 2.5 % sediment dose (originated from the sediment addition), compared to the pure chernosol, had predominance in relation to the repellent effect of the sediment contamination towards the collembolans. This behavior has been reported in other studies when testing soil dilutions of organic wastes (Natal-da-Luz et al. 2009).

Collembolan reproduction significantly increased in the lowest sediment doses of ferralsol mixtures (1.25 % and 2.5 %). Therefore, the second hypothesis was confirmed by collembolan reproduction but only for ferralsol mixtures. The ferralsols are characterized by low concentrations of organic matter, low contents of nutrients (low natural fertility) and high concentrations of total iron and aluminum. These properties originated a low reproductive output in the 0 % dose of ferralsol and a mortality higher than 20 %. Consequently, the addition of low amounts of organic matter (originated from the sediment addition) stimulated the reproduction of *F. candida*. This agrees with Natal-da-Luz et al. (2009) who also reported the increase of collembolan reproduction in

Table 4 Total metal concentrations in the test mixtures of chernosol or ferralsol with a dredged sediment and upper limit values of metals established by Brazilian legislation for soil quality (CETESB 2005; CONAMA 2009)

Test mixtures (%)	Cr (mg/kg)	Cu (mg/kg)	Ni (mg/kg)	Pb (mg/kg)	Zn (mg/kg)
Chernosol					
0.00	8.61	2.21	5.97	4.13	22.29
1.25	73.83	59.03	26.63	51.94	111.81
2.5	82.46	47.32	38.34	43.71	119.12
5.0	85.12	62.57	33.96	24.77	134.98
10.0	94.77	91.15	34.91	48.03	146.02
20.0	118.15	114.93	56.90	97.41	216.42
Ferralsol					
0.00	36.08	< 0.04	7.97	6.67	7.06
1.25	63.67	< 0.04	25.70	8.40	9.11
2.5	54.79	5.74	30.42	27.00	35.53
5.0	73.04	7.29	27.70	32.58	49.38
10.0	75.60	29.06	34.98	44.70	119.81
20.0	81.45	54.36	29.60	63.42	207.52
Reference ^a	35	60	17	13	40
Prevention ^b	60	300	72	30	75
Intervention ^c	200	400	180	70	150

^a Values representing pedogeochemical background of the São Paulo State, Brazil

^b Values estimated with tests using ecological receptors

^c Values based on potential risks on human health

natural soils treated with different doses of an organic sewage sludge. On the other hand, although the organic content in the pure chernosol is also low, this type of soils has high nutrient

contents, fact that may have favored the reproduction of the test organisms.

Similar to that observed with collembolans in ferralsol mixtures, the lowest sediment doses increase the reproduction of enchytraeids in mixtures of both chernosol and ferralsol, which also confirms the second working hypothesis. This increase was more pronounced at the lowest sediment dose of ferralsol treatments (2.5 %) compared to the same dose of chernosol mixtures. Once more such fact may be associated with the low nutrient concentrations of the ferralsols, compared to that of the chernosol.

The reproduction EC50 value estimated for collembolans in the chernosol mixtures was in the same order of magnitude of that estimated for enchytraeids. This does not agree with Gejlsbjerg et al. (2001) who supported that *F. candida* reproduction is more sensitive than *Enchytraeus albidus* when evaluating the ecotoxicity of sewage sludge-amended soils. The authors suggested that enchytraeids are more vulnerable to toxicants from pore water, while collembolans seek for dried zones into the soil, feed on relative higher amounts of sludge and, consequently, are more often exposed to sludge-bound contaminants.

Concerning the outcome observed in the replicates with pure test soils, reproduction in the chernosol was higher compared to that observed in ferralsol, for both test species. Since the chernosol contains higher natural levels of nutrients and much lower contents of total iron compared to ferralsol (Table 2), most probably the availability of food for the organisms was higher in the chernosol, fact that might have favored the reproductive performance of the test organisms in that soil.

Table 5 Potentially bioavailable (Bio) metal concentrations in the test mixtures of chernosol or ferralsol with a dredged sediment

Test mixtures (%)	Cr		Cu		Ni		Pb		Zn	
	Bio (mg/kg)	Bio (%)	Bio (mg/kg)	Bio (%)	Bio (mg/kg)	Bio (%)	Bio (mg/kg)	Bio (%)	Bio (mg/kg)	Bio (%)
Chernosol										
0.00	9.74	9.90	< 0.04	—	5.60	8.53	2.18	11.73	3.25	46.01
1.25	7.39	11.61	< 0.04	—	2.79	10.87	4.86	12.92	4.81	52.84
2.5	7.94	14.48	0.04	0.08	3.89	12.79	3.15	11.65	9.31	26.20
5.0	9.48	12.98	0.11	0.18	3.49	12.58	3.36	9.50	21.85	44.25
10.0	12.79	16.92	0.13	0.14	4.77	13.63	6.38	14.27	22.55	18.82
20.0	8.07	26.99	0.25	0.22	2.52	31.16	7.58	11.95	45.01	21.69
Ferralsol										
0.00	nd	nd	0.04	0.48	3.86	64.69	1.97	47.67	5.27	23.63
1.25	11.26	3.27	0.10	0.13	3.99	12.37	4.96	9.54	33.77	18.28
2.5	13.94	47.42	0.15	0.18	4.74	14.97	4.78	10.94	35.11	30.20
5.0	8.74	16.91	0.33	0.39	7.19	21.17	9.09	36.71	66.93	29.47
10.0	3.10	10.27	0.07	0.08	8.54	24.45	12.20	25.39	26.69	49.58
20.0	19.72	16.69	0.25	0.21	8.64	15.18	12.50	12.83	33.72	6.53

Conclusions

The results indicated that soil properties played an important role in the ecotoxicity of the dredged sediment for soil fauna and that chemical analyses are not sufficient to predict toxic effects to soil fauna (the concentration gradient with the highest total metal concentrations was not the most toxic one). In this case, the presence of expansive clay minerals and variations of fertility seem to be the properties which most effectively influence the ecotoxicity of sediment-amended soils. The present study highlights that the actual Brazilian legislation for soil quality and terrestrial disposal of dredged sediments should be reviewed according to the geo-diversity of tropical soils from Brazil and the influence of their properties on the bioavailability and ecotoxicity of contaminants for edaphic fauna should be taken into account. In this context, combinations of other types of tropical soils and sediments from different sources (e.g., port sediments) should also be tested, in order to generate data which can reflect distinct and spatially representative Brazilian biogeographical, pedological and geological attributes. Such information is necessary to support decision-makers for public health and environmental management programs, including remediation practices, hydrographical basin management, ecological zoning, agriculture planning and urbanization of areas treated with dredged sediments.

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