



Changes on microbial C and enzyme activities in soil with amendment of composted tannery sludge after 9 years

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Abstract

Purpose The consecutive amendment of composted tannery sludge (CTS) could promote changes in the status of soil microorganisms. Thus, this study evaluated the changes on microbial C and enzyme activities in soil after 9 years of CTS amendment.

Methods CTS was amended from 2009 to 2018 at five rates: 0, 2.5, 5, 10, and 20 ton ha⁻¹. In 2018, the soil chemical properties (pH, electric conductivity, P, K, Cr, and total organic C), microbial C and enzyme activities were evaluated after 30 days from the amendment.

Results The values of chemical properties increased after nine years of CTS application. The content of microbial C and the enzyme activities increased with the amendment of 2.5 and 5 ton ha⁻¹, and decreased with the amendment of 10 and 20 ton ha⁻¹.

Conclusion This study showed that the amendment of 10 and 20 ton ha⁻¹ of CTS increased soil pH and Cr concentration and promoted a decreasing on soil microbial C and enzyme activities.

Keywords Industrial waste · Microbial biomass · Waste management · Enzymes activities

Introduction

The use of municipal and industrial wastes in agricultural soils has increased in the world, since these wastes are accumulating in the environment (Boechat et al. 2017). Among them, tannery sludge presents organic and inorganic substances, such as salts and carbonates, but mainly chromium (Cr) (Araújo et al. 2016) and, thus, the amendment this waste should be monitored over time to prevent soil pollution. However, some alternative methods for treating the tannery sludge, before its application in soil, could be applied, such as composting (Sousa et al. 2018), although this process does not degrade metals. Although the composting of this waste could improve its quality and release nutrients

available to plants (Liu et al. 2018), the presence of salts, carbonates and Cr could increase the salinity, alkalinity and Cr pollution over time. Therefore, changes in soil chemical properties after consecutive amendment of this compost could change the status of microorganisms and their activities in the soil.

Soil microorganisms present high sensitivity to the soil conditions and also to the environmental impact of municipal and industrial wastes (Boechat et al. 2012; Sousa et al. 2017; Miranda et al. 2018). In these conditions, the evaluation of microbial C is considered as a suitable and robust method for measuring the effect on soil microorganisms (Lopes et al. 2013). In addition, microorganisms produce and release enzymes that act and degrade on different substances being considered a suitable measure of soil microbial activity (Cenciani et al. 2008). However, changes in the soil chemical properties, influenced by amendment of wastes, can influence the soil microorganisms and their activities (Margon et al. 2014; Malik et al. 2018). Indeed, Araujo et al. (2013) reported that the amendment of composted tannery sludge (CTS) increased soil pH and the content of Na and

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Cr in soil. Thus, we hypothesized that these shifts in soil chemical properties, after consecutive amendment of this composted waste, could change the status of microbial C and enzymes in soil. Therefore, the aim of this study was to evaluate the changes on microbial C and enzymes after 9 years consecutive amendment of composted tannery sludge.

Materials and methods

This experiment is located at the Department of Soil Science and Agricultural Engineering from Federal University of Piauí, Brazil. The soil presents 10% clay, 28% silt, and 62% sand. The compost was obtained through the mixture of tannery sludge, sugarcane straw and cattle manure (1:3:1; v:v:v), after composting for 3 months. Three subsamples of CTS (100 g) were collected and mixed to form a composite sample that was analyzed according to USEPA (1996) (Table 1).

The amendment of CTS began in 2009 at five rates: 0 (without CTS), 2.5, 5, 10, and 20 ton ha⁻¹. The experiment is designed as a randomized block design with four replicates. This compost was amended, along 8 years 2009–2017 (a frequency of one application per year), in experimental plots with 20 m² each one. In 2018 (9th year), CTS was amended on the soil surface and incorporated into the 20-cm layer. Soil sampling was done after 7 days of the amendment of CTS. In each plot, five soil subsamples were collected at 0–20-cm depth, mixed to form a composite sample, sieved (2 mm) and stored at 4 °C.

Soil chemical properties (pH, electric conductivity, P and K content) were estimated according to Embrapa (1997). Total organic C (TOC) was estimated by wet digestion with a mixture of potassium dichromate and sulfuric acid using

an external heating (Yeomans and Bremner 1988). Cr content was analyzed according to the method 3050B (USEPA 1996). The soil extract was analyzed for Cr by atomic absorption spectrophotometry.

The microbial C was estimated by the microwave irradiation extraction method (Islam and Weil 1998) using 0.5 mol L⁻¹ sulfuric acid as extractant and extraction efficiency coefficient of 0.21. Soil respiration was estimated by CO₂ evolution for 7 days (Alef and Nannipieri 1995). Briefly, soil samples (50 g) were sealed in a 1-L pot with 10 mL of 1 mol L⁻¹ KOH and titrated with 0.1 mol L⁻¹ HCl. The respiratory quotient (qCO₂) was calculated from the ratio between soil respiration and microbial C (Anderson and Domsch 1993). The microbial quotient was estimated using the ratio of microbial C to TOC. Fluorescein diacetate (FDA) hydrolysis and dehydrogenase activity were estimated according to Schnurer and Rosswall (1982), and the amount of produced fluorescein was spectroscopically measured at 494 nm. The dehydrogenase (DHA) activity was estimated by the spectrophotometric determination of the triphenyltetrazolium formazan released by 5 g of soil after 24-h incubation at 35 °C (Casida et al. 1964).

The data were submitted to the analysis of variance (ANOVA) and the means were compared by the Student's test (5% level). All the statistical analyses were performed with the SPSS (version 15.0) software package.

Results and discussion

The annual amendment of CTS promoted changes in soil chemical properties. The results showed significant increases in TOC, pH, P, K, EC and Cr after 9 years of amendment of CTS, mainly at the highest rates (Table 2).

These shifting on chemical properties are directly related with the composition of CTS and its permanent amendment over 9 years. On one hand, the increase in the content of TOC, P and K in the soil is important for improving its fertility and also the status of nutrients available to plants. Some previous studies have found the increase of organic C and nutrients, such as P, K and Ca, after application of municipal (Khaliq et al. 2017) and industrial (Bose and Bhattacharyya 2012) wastes. On the other hand, the high values of pH and EC, mainly using the highest CTS rates, should be monitored to avoid their negative effect to plant growth. For example, high soil pH influences the availability some micronutrients, such as Zn, Cu, Mn, and Fe, to plants and, therefore, this unavailability can decrease plant growth (Mickelbart et al. 2012); while that EC can influence the germination, growth and the physiology of plants (Ambede et al. 2012). The results also showed that the Cr content strongly increased from the lowest to the highest CTS rate and it can be explained by the high concentration of Cr in

Table 1 Chemical properties of CTS

Variable	CTS
pH	7.4
C (g kg ⁻¹)	205.1
N (g kg ⁻¹)	1.59
P (g kg ⁻¹)	4.19
K (g kg ⁻¹)	3.90
Ca (g kg ⁻¹)	103.2
Mg (g kg ⁻¹)	7.6
S (g kg ⁻¹)	9.20
Cu (mg kg ⁻¹)	17.08
Zn (mg kg ⁻¹)	119.6
Mo (mg kg ⁻¹)	9.42
Ni (mg kg ⁻¹)	25.06
Cd (mg kg ⁻¹)	2.47
Cr (mg kg ⁻¹)	1891
Pb (mg kg ⁻¹)	41.30



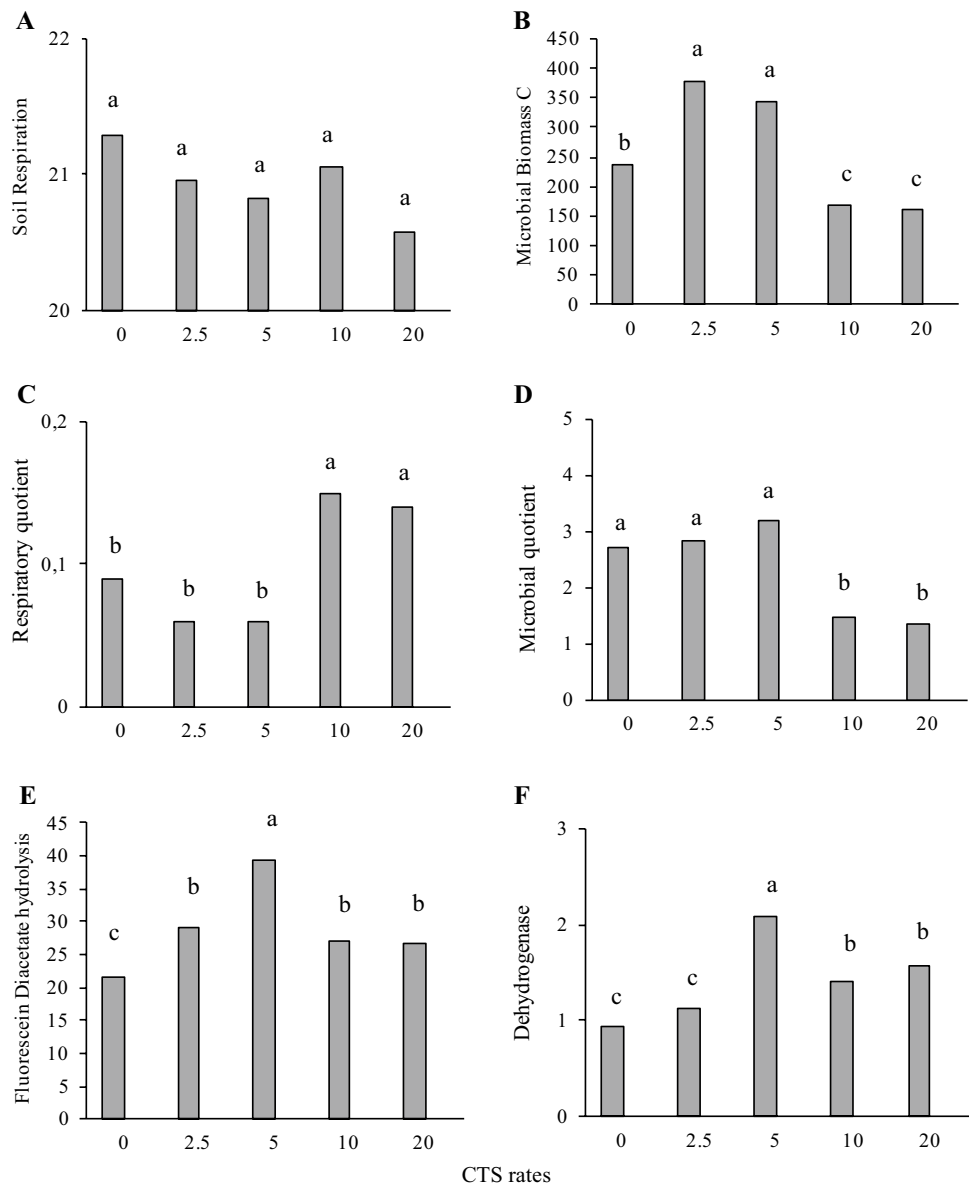
Table 2 Soil chemical properties after CTS amendment

	TOC g kg ⁻¹	pH	P mg kg ⁻¹	K mg kg ⁻¹	EC dS m ⁻¹	Cr mg kg ⁻¹
0	7.3 d	6.9 c	2.0 c	23.1 c	0.12 b	4.5 e
2.5	9.5 c	7.2 b	2.4 b	25.3 c	0.13 b	17.6 d
5	10.2 c	7.3 b	2.7 b	32.5 b	0.15 b	32.5 c
10	11.4 b	7.7 a	3.6 a	37.4 a	0.25 a	85.6 b
20	12.2 a	8.1 a	3.4 a	35.0 a	0.26 a	216.4 a

Means with different letters vary significantly (Tukey's HSD test)

TOC total organic C, EC electrical conductivity

Fig. 1 Soil microbial properties after amendment of CTS rates (0, 2.5, 5, 10 and 20 ton ha⁻¹) at the 9th year. Soil respiration (**a**; mg CO₂-C kg⁻¹ day⁻¹), microbial biomass C (**b**; mg kg⁻¹), respiratory (**c**; g CO₂-C day⁻¹ g⁻¹ MBC) and microbial quotients (**d**; %), fluorescein diacetate hydrolysis (**e**; µg FDA g⁻¹) and dehydrogenase (**f**; µg triphenyl tetrazolium chloride g⁻¹)



CTS. However, the increase in soil pH associated with the highest organic C, after the amendment of CTS, may control the availability of metals, such as Cr, and it could be a strategy for moving pollutants with environmental risk (Navarro-Pedreño et al. 2018).

Although CTS has promoted an increase in the values of chemical properties, soil microbial properties behave differently to the amendment of CTS over nine years (Fig. 1).

Soil respiration did not change (Fig. 1a), while microbial biomass C (Fig. 1b) presented the highest values with the



amendment of CTS at 2.5 and 5 ton ha⁻¹, and decreased in the treatments with 10 and 20 ton ha⁻¹. The respiratory and microbial quotients were differently influenced by CTS; respiratory quotient increased while microbial quotient decreased in the treatments with 10 and 20 ton ha⁻¹ CTS (Fig. 1c, d). FDA hydrolysis and dehydrogenase activity presented the highest values in the treatment with of 5 ton ha⁻¹ CTS and decreased in 2.5, 10 and 20 ton ha⁻¹ CTS.

The results showed that soil respiration was not a sensitive indicator for measuring the effect of CTS on soil. It can be explained by the time that this variable was measured, i.e., 30 days after the amendment of the compost. Thus, the absence of significant differences in soil respiration between treatments could be related with the consumption of C from soil during the period between the amendment and measurement of the carbon dioxide released by the soil. On the other hand, the effect of CTS was significant on the content of microbial C and enzymes. Therefore, the use of 2.5 and 5 ton ha⁻¹ of this compost promoted an increase in the size of microbial pool and the catabolic activity. It can be related with the C and nutrient sources released by CTS and also the better soil conditions found with the amendment of these rates. This finding agrees with Boechat et al. (2012) who evaluated the effect of industrial and organic waste on soil and found an increase in soil microbial biomass with the application of the wastes. Previously, Bouzaiane et al. (2007) evaluated the effect of municipal solid waste compost on soil microbial biomass C and N, and found an improvement in the size of soil microbial biomass with the application of 40 t ha⁻¹ of compost. On the other hand, the decrease in the content of microbial C and enzyme activity with the use of 10 and 20 ton ha⁻¹ could be related with the high values of soil pH and Cr found after 9 years of amendment of CTS. It corroborates with Malik et al. (2018) which found that the increase in soil pH decreased soil microbial biomass and reduced its efficiency. Interestingly, it could explain the lowest microbial quotient found in this study with the amendment of 10 and 20 ton ha⁻¹ of CTS. For Cr, Margon et al. (2014) reported lowest microbial biomass and enzyme activities with the presence of this metal in soil. Chromium can act as an oxidizing agent and oxidative damage on microbial cells (Ackerley et al. 2006).

Conclusions

In conclusion, this study showed that the amendment of 10 and 20 ton ha⁻¹ of the compost increases soil pH and Cr concentration, and it contributes for decreasing the content of microbial C and enzymes activity. Further studies should be done evaluating methods for restoring this area which present high soil pH and Cr concentration. Also, the prospection of microbes with tolerance to Cr can be important for

selecting strains with potential for detoxifying contaminated soils with Cr.

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