

# Chromium accumulation in maize and cowpea after successive applications of composted tannery sludge

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**ABSTRACT.** Chromium (Cr) accumulation in soil and plants has been reported after successive applications of tannery sludge, which is a matter of concern because Cr can promote environmental contamination and affect the food chain. In this study, we evaluated the growth of and Cr accumulation in maize and cowpea after seven years of consecutive applications of composted tannery sludge (CTS) under field conditions. The experiment consisted of application of CTS at five levels: 0 (control), 2.5, 5, 10, and 20 Mg ha<sup>-1</sup> (dry basis). The growth of and Cr accumulation in maize and cowpea were evaluated at 75 and 65 days after plant emergence, respectively. CTS application increased the shoot biomass of maize and cowpea. Accumulation of Cr was similar for both plant species, with higher accumulation in roots. However, Cr accumulation in grains differed as the CTS doses increased; cowpea accumulated more Cr while maize did not accumulate more Cr in grains with higher CTS doses. In conclusion, application of CTS led to higher growth of maize and cowpea plants, and both species exhibited similar Cr accumulation in roots and shoots.

Keywords: waste management; tannery sludge; plant and soil interaction; metals.

# Acumulação de cromo em milho e feijão-caupi após aplicações sucessivas de lodo de curtume compostado

**RESUMO.** A acumulação de cromo (Cr) no solo e nas plantas tem sido relatada após sucessivas aplicações de lodo de curtume, o que é motivo de preocupação tendo em vista que este elemento pode promover a contaminação ambiental e afetar a cadeia alimentar. Neste estudo, avaliou-se o crescimento e a acumulação de Cr no milho e caupi após sete anos de aplicações consecutivas de lodo de curtume compostado (LCC) em condições de campo. O experimento consistiu na aplicação de LCC em cinco doses: 0 (controle); 2,5; 5; 10 e 20 Mg ha<sup>-1</sup> (base seca). O crescimento e o acúmulo de Cr no milho e no caupi foram avaliados aos 75 e 65 dias após a emergência das plantas, respectivamente. A aplicação de LCC aumentou a biomassa do milho e caupi. A acumulação de Cr foi semelhante para ambas as espécies; oc maior acumulação relatada nas raízes. No entanto, a acumulação de Cr nos grãos diferiu entre as espécies: o caupi acumulou Cr, enquanto o milho não acumulou esse elemento nos grãos. Em conclusão, a aplicação de LCC proporcionou maior crescimento de plantas de milho e feijão-caupi, e ambas as espécies apresentaram respostas semelhantes à acumulação de Cr nas raízes e parte aérea.

Palavras-chave: gestão de resíduos; lodo de curtume; interação planta e solo; metais.

# Introduction

Tannery industries generate an enormous amount of solid waste, commonly known as tannery sludge. In Brazil, the volume of tannery sludge generated by the industry is approximately 150 - 200 kg per ton of leather (dry basis). This waste usually contains a high amount of chromium (Cr) that is deposited in the environment and can result in environmental contamination and, consequently, affect the food chain (Singh et al., 2015; Silva et al., 2010). Some studies have proposed the agricultural use of composted tannery sludge as an alternative treatment method like that of other similar solid waste (Singh et al., 2015; Singh, Singh, Ibrahim, & Hashim, 2011; Silva et al., 2010). Previous studies have shown that the use of composted tannery sludge has improved the soil fertility and crop yield (Miranda, Nunes, Oliveira, Melo, & Araújo, 2014; Araújo, Lima, Santos, & Schmidt, 2016). On the other hand, the application of composted tannery sludge has been reported to increase the Cr concentrations in the soil (Santos, Araújo, Nunes, Oliveira, & Melo, 2014), which could limit its use for agricultural purposes, since this element can be easily absorbed by the plants (Gondek, 2008).

However, different plant species have distinct responses to the absorption and translocation of Cr after the land application of tannery sludge (Tudunwada, Essiet, & Mohammed, 2007; Cavallet, Selbach, & Gianello, 2007). Tudunwada et al. (2007) evaluated the effect of repeated applications of tannery sludge and found that Cr accumulated more in maize than in sorghum. Similarly, Cavallet et al. (2007) studied the effect of tannery sludge on plant Cr concentrations and observed that Cr was concentrated in the shoots of radish, while in sorghum, the element was accumulated in the roots without translocation to the shoots and grains.

Therefore, these previous studies confirm that different plant species respond differently to tannery sludge amendments in soil. However, the effect of repeated application of composted tannery sludge on the growth and Cr accumulation in different plant species is unclear. The main hypothesis is that grasses and legumes present different behavior to Cr accumulation after repeated applications of composted tannery sludge. In this way, this study evaluated the growth, Cr accumulation and yield response of maize and cowpea after seven years of consecutive applications of composted tannery sludge (CTS) in agricultural soil.

# Material and methods

The experiment was carried out under field conditions at the "Long-Term Experimental Field" of the Agricultural Science Center, Teresina, Piauí State (05°05 S; 42° 48 W, 75 m). The regional climate is dry tropical (Köppen), and it is characterized by two distinct seasons: a rainy summer and a dry winter, with annual average temperatures of 30°C and rainfall of 1,200 mm. The soil of the area is classified as a Fluvisol (10% clay, 28% silt, and 62% sand). Composted tannery sludge (CTS) was produced by mixing tannery sludge with sugarcane straw and cattle manure (ratio 1:3:1; v:v:v), and the composting was performed using the aerated-pile method for 90 days. The physicochemical characteristics of the CTS were evaluated at the end of the composting process through the method proposed by USEPA (1986), presenting: pH - 7.5; total organic C - 201 g kg<sup>-1</sup>; N - 15 g kg<sup>-1</sup>; P -4.9 g kg<sup>-1</sup>; K - 2.9 g kg<sup>-1</sup>; Ca - 121 g kg<sup>-1</sup>; Mg - 7.2 g kg<sup>-1</sup>; Na - 49.1 g kg<sup>-1</sup>; and Cr - 1,941 mg kg<sup>-1</sup>.

CTS has been applied annually since 2009 at five rates: 0 (without CTS application), 2.5, 5, 10, and 20

Mg ha<sup>-1</sup> of CTS (dry basis). The experimental site is arranged in a completely randomized design with four replicates. Plots are 20 m<sup>2</sup> each, with 12 m<sup>2</sup> of usable area for soil and plant sampling, and rows are spaced 1.0 m apart. In the seventh year (2015), CTS was applied 10 days before maize (Zea mays L.) AG 1051 was sown. The CTS was spread on the soil surface and incorporated into the 20-cm layer with a harrow. Maize was grown at a density of 5 plants m<sup>-1</sup> (approximately 62,000 plants ha<sup>-1</sup>) for 75 days. After this period, cowpea [Vigna unguiculata (L.) Walp.], cv. BRS Tumucumaque, was sown at the density of 6 plants m<sup>-1</sup> (approximately 120,000 plants ha<sup>-1</sup>) for 68 days. During the experiment, the plants were irrigated using conventional irrigation according to the water requirements of the plants. Thus, a total of 656 mm and 670 mm of water were applied for maize and cowpea, respectively (Figure 1).



**Figure 1.** Climatic data during the experiment with maize and cowpea cultivation (CTS application, soil sampling, input of water). PET - potential evapotranspiration.

Soil and plant samples were collected during the CTS application (0 days) and during the harvesting of maize (75 days) and cowpea (180 days). For soil sampling, five subsamples were collected (0-20 cm depth) per plot to form a composite sample. The samples were air-dried and sieved (2 mm), and the pН  $(CaCl_2),$ electric conductivity (EC), exchangeable cations (Ca<sup>2+</sup>, Mg<sup>2+</sup>, K<sup>+</sup>, and Na<sup>+</sup>) and available phosphorus (P) were analyzed according to Donagema, Campos, Calderano, Teixeira, and Viana (2011). Total organic C (TOC) was determined by the wet method using a mixture of 5 mL of potassium dichromate (0.167 mol L<sup>-1</sup>) and 7.5 mL of concentrated sulfuric acid, with the application of heat (170°C for 30 min.). Soil Cr was extracted by the DTPA-TEA method and measured using the USEPA-3050 method (USEPA, 1986).

For the estimation of Cr accumulation in maize and cowpea, ten whole plants of each crop were

#### Cr accumulation in maize and cowpea

randomly collected per plot and were divided into different parts; cowpea was divided into roots, leaves, pods and grains, while maize was divided into roots, stems, leaves and grains. The plants were dried at 65°C until constant weight and weighed. Plant organs were ground, and the Cr concentration was determined using the methodology proposed by USEPA (1986). The translocation factor (TF) (Patel, Pandey, & Patra, 2016), which describes the ability of the plant to translocate Cr from roots to shoots in different concentrations, was calculated as:

TF = Cr content in the shoot (mg kg<sup>-1</sup>) / Cr content in the roots (mg kg<sup>-1</sup>)

The bioaccumulation factors (BAF) were calculated as the ratio between the Cr concentration in each partition and the total Cr concentration in the soil (Carbonell, Imperial, Torrijos, Delgado, & Rodriguez, 2011). Data were statistically analyzed using an analysis of variance (ANOVA), and means were compared by the Tukey test. All data were analyzed using the R software (R Core Team, 2016).

# **Result and discussion**

Composted tannery sludge (CTS) presents high levels of organic matter and chemical elements that can improve soil fertility and, consequently, the availability of nutrients for plants (Table 1). Similarly, several previous studies have also shown increases in organic C, total N, available P and total Fe concentrations in the soil due to sludge amendments (Singh & Agrawal, 2007; 2008; 2009). Silva et al. (2010) also reported that the nutrient contents and heavy metal contents in soil increased with increasing compost rates.

Previous studies have shown that the application of organic sludge to agricultural soil not only facilitates nutrient cycling, but may be helpful in eliminating the need for the commercial fertilization of cropland (Sommers, 1977; Singh & Agrawal 2007; 2009; Singh et al., 2011). This finding agrees with previous studies that applied tannery sludge to soil and found significant increases in plant biomass (Araujo, Tiritan, Pereira, & Caetano Júnior, 2008; Carbonell et al., 2011). On the other hand, the application of CTS did not influence the root dry weight (Figure 2B) suggesting that CTS does not present toxicity for root growth. This behavior may be related to the low mobility of Cr through the cell membrane resulting in the rare occurrence of toxicity in plants (Panda & Choudhury, 2005). Silva et al. (2010) reported significant increases in root biomass of *Capsicum* due to different CTS amendments compared to unamended soil.

The application of CTS significantly increased the shoot dry weight of maize and cowpea (Figure 2A) by 107% and 76% at the highest CTS rate (20 Mg ha<sup>-1</sup>) compared with the control, respectively. This increase is related to the greater availability of nutrients in the soil after the application of CTS (Table 1). Chromium (Cr) concentrations varied according to different parts of plants. For maize, the Cr concentration in the roots increased significantly with the application of CTS, where the values ranged from 47.17 to 152.07 mg kg<sup>-1</sup> for the control and 10 Mg ha<sup>-1</sup>, respectively. However, there was no significant increase in Cr concentrations in the stem, with a mean Cr value of 2.44 mg kg<sup>-1</sup>. Similarly with the roots, there was increased Cr concentrations in maize leaves with the application of CTS, where the values ranged from 4.39 to 10.77 in the control and 20 Mg ha<sup>-1</sup>, respectively. Interestingly, the concentration of Cr in the grains decreased with the addition of CTS (Figure 3A). For cowpea, Cr concentrations increased significantly with the application of CTS (Figure 3B). The concentration of Cr in the roots increased from 31.17 mg kg<sup>-1</sup> to 93.26 mg kg<sup>-1</sup> for the control and 20 Mg ha<sup>-1</sup>, respectively. No significant differences were found for Cr concentrations in the pod, with a mean Cr value of 4.99 mg kg<sup>-1</sup>. However, significant differences were observed in leaves, with values ranging from 11.31 to 29.92 for the control and 20 Mg ha<sup>-1</sup>, respectively. In contrast with maize, the concentrations of Cr in the grain increased with the application of CTS.

Table 1. Soil chemical properties under application of different CTS rates (0, 2.5, 5, 10, and 20 Mg ha<sup>-1</sup>).

Sampling time	CTS	pН	TOC	EC	Р	К	Ca	Mg	Na	
	Mg ha <sup>-1</sup>	CaCl <sub>2</sub>	g kg <sup>-1</sup>	dS m <sup>-1</sup>	mg dm-3	mmol, dm <sup>-3</sup>				
	0	5.6 d	5.5 b	0.7 b	6.0 c	2.2 a	13.3 b	6.5 a	4.6 a	
Maize (75 days)	2.5	5.8 c	7.0ab	0.7 b	6.8bc	2.2 a	20.0ab	7.8 a	5.0 a	
	5	6.0 c	7.3 a	0.7 b	7.3bc	2.2 a	26.0 a	8.0 a	4.9 a	
	10	6.6 b	7.7 a	0.7 b	10.3ab	2.2 a	24.3 a	8.5 a	4.9 a	
	20	6.9 a	8.5 a	0.9 a	12.5 a	2.2 a	27.3 a	8.8 a	4.9 a	
Cowpea (180 days)	0	5.1 d	4.9 b	0.5 a	4.3 c	1.9 a	12.5 с	5.0 b	4.4 a	
	2.5	5.4 d	5.7ab	0.5 a	5.0bc	1.8 a	17.0bc	5.8ab	4.9 a	
	5	5.8 c	6.8 a	0.5 a	6.0ab	1.9 a	22.8ab	7.0 a	4.9 a	
	10	6.2 b	6.6 a	0.6 a	7.8ab	1.9 a	23.5ab	7.5 a	4.6 a	
	20	6.6 a	7.1 a	0.6 a	9.5 a	1.8 a	25.8 a	7.0 a	4.9 a	

Values followed by the same letter in the column are not significantly different (p < 0.05) by Tukey test, between treatments for maize and cowpea.



**Figure 2.** Dry matter of shoots - DMS (A) and roots – DMR (B) of maize and cowpea plants after application of different of CTS doses. Different lowercase letters indicate a significant difference (p < 0.01) between treatments for maize. Different upper case letters indicate a significant difference (p < 0.01 by Tukey test) between treatments for cowpea. The bars represent the standard error of the mean (n = 4).



**Figure 3.** Total concentration of Cr (mg kg<sup>-1</sup>) in different parts of the maize and cowpea plants grown in soil submitted to different doses of composted tannery sludge (0, 2.5, 5, 10, and 20 Mg ha<sup>-1</sup>). Letters indicate a significant difference at p < 0.05 by Tukey test.

Independent of plant species, the concentrations of Cr were higher in the roots than in the shoots. The highest Cr accumulation in the roots occurs due to their direct contact with Cr in the soil solution (Sampanpanish, Tippayasak, & Chairat-Utai, 2010). This Cr accumulation in the roots may be explained by two possible mechanisms: a) a fraction of Cr ions would be physically adsorbed to the negative charges on the root cell walls (Gomes, Marques, Nogueira, Castro, & Soares, 2011); b) another fraction would be absorbed by the cells, being immobilized in the root vacuoles (Nematshahi, Lahouti, & Ganjeali, 2012). In addition, we could consider that the retention of Cr in the roots would represent a natural protection strategy against Cr toxicity (Sinha, Gupta, & Bhatt, 2007), since there was an increase in the shoot biomass and no negative effect in the roots after CTS application.

The results showed similar behavior of maize and cowpea to the accumulation of Cr in their organs (Table 2). The values of bioaccumulation factors (BAF) confirm that the roots are the main bioaccumulator of Cr. Similar results were reported by Cavallet et al. (2007) and Patel et al. (2016), who evaluated the uptake of Cr by Sorghum vulgare and Mentha spicata, respectively, grown under different levels of tannery sludge, where the highest accumulation of Cr occurred in the roots. Our results also showed a reduction in the bioaccumulation with the application of the highest rate of CTS. Interestingly, the grains showed bioaccumulation factors lower than 1, which indicates low Cr uptake (Carbonell et al., 2011). The reduction of Cr uptake with the application of CTS may be related to the increased soil organic matter content, which can regulate the availability of metals in soil (Singh & Agrawal, 2007).

There was variation between maize and cowpea regarding Cr translocation from roots to shoots. Although higher translocation factors were observed in the leaves for both species, for maize, the Cr translocation in the grain was reduced with the application of CTS (Figure 4A), while for cowpea, there was greater translocation in the grain with the application of CTS (Figure 4B). Therefore, the different responses of each plant species to Cr translocation from leaves to grains after applications of composted tannery sludge may indicate the different influence of the physiological characteristics of these species to the presence of Cr.

CTS (Mg							Maize						
ha <sup>-1</sup> ) Roots					Stems			Leaves			Grains		
0	12.26	±	3.64	0.46	±	0.26	1.	14	±	0.30	1.08	±	0.23
2.5	2.98	±	0.53	0.07	±	0.03	0.	16	±	0.05	0.08	±	0.04
5	2.89	±	0.44	0.05	±	0.03	0.	13	±	0.04	0.02	±	0.02
10	1.51	±	0.42	0.03	±	0.00	0.	06	±	0.02		< 0.01	
20	0.98	±	0.36	0.01	±	0.01	0.	07	±	0.02		< 0.01	
CTS (Mg							Cowpea						
ha <sup>-1</sup> )	Roots			Pods			Leaves				Grains		
0	5.53	±	1.59	1.02	±	0.26	2.	00	±	0.70	0.12	±	0.03
2.5	1.31	±	0.16	0.17	±	0.03	0.	34	±	0.04	0.03	±	0.00
5	0.88	±	0.29	0.09	±	0.05	0.	23	±	0.07	0.07	±	0.01
10	0.60	±	0.28	0.07	±	0.05	0.	26	±	0.06	0.07	±	0.03
20	0.56	±	0.05	0.03	±	0.00	0.	18	±	0.04	0.03	±	0.01

Table 2. Bioaccumulation factor of Cr in different parts of maize and cowpea plant cultivated in soil submitted to consecutive applications of composted tannery sludge.

 $\pm$  standard error of the mean (n = 4).

This may be explained through the modification in plant metabolism for adapting to an excess of Cr (Wang et al., 2013). Interestingly, maize plants accumulated a relatively higher Cr content in the roots than cowpea plants. This may explain the low translocation to the shoots, considering that the Cr fraction bound to root cells cannot be translocated to the shoots and, therefore, could not be removed in the shoot biomass harvest (Terfie & Asfaw, 2015). Hossner, Loeppert, Newton, and Szaniszlo (1998) reported that dicotyledon species, such as cowpea, uptake and transport more Cr to shoots than monocotyledons, such as maize. This result may be due to differences in the rooting patterns, transpiration rates and metabolism between these two groups of plants (Hossner et al., 1998). Nonetheless, it was observed that for both crops, the translocation factors of all organs were smaller than 1, suggesting a low translocation of Cr from roots to shoots and, thus, indicating efficient immobilization of this element in the root system (Carbonell et al., 2011).



**Figure 4.** Cr translocation factor in shoots of maize (A) and cowpea (B) after application of different doses of composted tannery sludge (Mg  $ha^{-1}$ ) in the soil. The bars represent the standard error of the mean (n = 4).

#### Conclusion

The application of CTS improved the soil fertility and increased the concentration of Cr in the soil. Therefore, the application of CTS provided better growth of maize and cowpea plants. Both plant species exhibited similar responses to the accumulation of Cr in the roots and shoots. However, they showed different behaviors to the accumulation of Cr in the grains, i.e., cowpea accumulated Cr as CTS levels increased, while maize did not accumulate this element in the grains. This finding indicates that further studies should be performed to understand the plant mechanisms involved in these different responses of maize and cowpea to the accumulation of Cr in CTStreated soil.

# Acknowledgements

The authors thank "Coordenação de Aperfeiçoamento de Pessoal de Nivel Superior – CAPES" (grant 23038.007660/2011-51) and "Conselho Nacional de Desenvolvimento Científico e Tecnológico - CNPq" (grants 471347/2013-2 and 305102/2014-1) for financial support to this project.

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Received on February 13, 2017. Accepted on August 28, 2017.

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