

# Growth and Nutrient Distribution in Young Plants of *Virola Surinamensis* Exposed to Cadmium

**WV. ANDRADE JÚNIOR**

UFRA: Universidade Federal Rural da Amazonia

**Cândido Oliveira Neto**

UFRA: Universidade Federal Rural da Amazonia

**Benedito Filho**

UFRA: Universidade Federal Rural da Amazonia

**Ricardo Okumura**

UFRA: Universidade Federal Rural da Amazonia

**Eniel Cruz**

Embrapa: Empresa Brasileira de Pesquisa Agropecuaria

**Cristine Amarante**

Museu Paraense Emilio Goeldi

**Vinicius Barbosa**

UFRA: Universidade Federal Rural da Amazonia

**Glauco Nogueira** (✉ [glauand@yahoo.com.br](mailto:glauand@yahoo.com.br))

UFRA: Universidade Federal Rural da Amazonia <https://orcid.org/0000-0003-3229-5694>

**Vitor Nascimento**

UFRA: Universidade Federal Rural da Amazonia

**Diana Sousa**

UFRA: Universidade Federal Rural da Amazonia

**Jessica Teixeira**

UFRA: Universidade Federal Rural da Amazonia

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# Abstract

The contamination of soils and water as a result of human actions has been increasingly frequent in the world, the cadmium element the as a promising contaminant of these environments. This element affects the growth and development of vegetables. The objective of the study was to evaluate the growth and concentration of macro and micronutrients in the different organs young plants of *Virola surinamensis* exposed to Cd. The Cd significantly affected the growth of *V. surinamensis* reducing the height, stem diameter and biomass production. The Cd influenced negatively Fe, Mg, Ca, N, P and K, especially in the root. The Zn increased in the roots and leaves, while Mn reduced in the root and increased in the leaves of the plants on exposure to Cd. The increase of Zn and Mn in the leaves may have been a strategy to maintain the stability and protection of the photosynthetic apparatus of the plant. the research concluded that cadmium affects the nutritional relationship of this vegetable, however, we could observe that the influence of metal depends on the species being studied, the time of exposure to the metal and the amount of this metal.

## 1. Introduction

The increase in cadmium (Cd) levels and their persistence in ecosystems has aroused great concern (Liu et al. 2015). This is because, even at low concentrations, Cd can have toxic effects on aquatic and soil organisms, including plants and animals, and ultimately on human health (Khan et al. 2017).

Cd is a non-essential heavy metal that is readily absorbed by plant roots through essential nutrient carriers (Bashir et al. 2015), and depending on concentration may interfere with the uptake, transport and use of macro and micronutrients plants (Di Baccio et al. 2014). Reduction of magnesium ( $Mg^{2+}$ ) (Di Baccio et al. 2014; Liu et al. 2015; Wang et al. 2016; Zouari et al. 2016), calcium ( $Ca^{2+}$ ), iron ( $Fe^{2+}$ ), manganese ( $Mn^{2+}$ ), zinc ( $Zn^{2+}$ ) (He et al. 2013), nitrogen (N), phosphorus (P) ( He et al. 2013) and potassium (K) (Gomes et al. 2013) in plants treated with Cd may cause reduction of photosynthesis and result in symptoms of phytotoxicity, such as leaf chlorosis, especially in young leaves (Fernández et al. 2013, Elloumi *et al.*2014, Yan et al. 2015), low biomass production, lower root growth, stem diameter and height, reduction of leaf numbers and, eventually, plant death (Yan et al. 2015; Solti et al. 2016; Nikolić et al. 2017).

However, tolerance to Cd has been observed in several species of plants due to the development of different mechanisms to restrict or neutralize the inhibitory effect of the excess metal (Gallego et al. 2012, Hernández et al. 2015; Wang et al. 2016). Some plants, including *Virola surinamensis*, have greater capacity to extract and accumulate the metal in the root and restricting its transport to the aerial parts (Andrade Júnior et al. 2019). While others, called hyperaccumulators, accumulate large amounts of metals, especially in the leaves, without presenting symptoms of phytotoxicity (Singh et al. 2016).

Cd, being one of the most toxic heavy metals with a high bioaccumulation capacity, has stimulated the search for solutions to remediate environments contaminated by it (Andrade Júnior et al. 2019). In this

sense, phytoremediation has been proposed as one of the main promising techniques for decontamination of soils and water (Zhao et al. 2015). Identification of forest species with long life cycle, large biomass production (Caires *et al.* 2011), tolerant to and with phytoextraction capacity of Cd can serve the preservation of natural areas and the recomposition of environments contaminated by this metal.

In the Amazon, studies involving the effect of Cd on woody species are scarce (Fan et al. 2011; Silva et al. 2017, Pereira et al. 2017, Andrade Júnior et al. 2019). *V. surinamensis* (Ucuúba), a widely distributed forest species adapted to the Amazonian floodplain and igapó ecosystems, especially in the estuaries, which are environments potentially susceptible to heavy metals, had high root and medium Cd accumulation and high metal tolerance (Andrade Júnior et al. 2019). Species with these characteristics are important for phytoremediation of metals. Therefore, it is important to analyze plant growth, biomass and nutritional changes of *V. surinamensis* when exposed to high concentrations of Cd.

In this study, we tested the hypothesis that young plants of *V. surinamensis* trigger different mechanisms to tolerate environments contaminated by Cd. Thus, the objective was to evaluate (1) the concentration of Cd in different tissues, (2) the growth of the plant in different concentrations of Cd and (3) the concentration of micro and macronutrients in roots and leaves of young plants of *V. surinamensis* under different concentrations of Cd.

## 2. Material And Methods

### 2.1 *Location of the experiment*

The experiment was conducted in a greenhouse at the Federal Rural University of Amazonia (UFRA) in Belém, State of Para, Brazil (01° 27' 21" S, 48° 30' 16" W) during September 15th to November 14th of 2017. According to the climatic classification of Köppen, the climate is type Af (Tropical rainforest), with an annual average rainfall of 2921.7 mm, average temperature of 25.9°C, average relative humidity of 86.8% and wind speed of 1.35 m s<sup>-1</sup>. (Ramos et al. 2009).

### 2.2 *Plant material and growth conditions*

Seeds of *V. surinamensis* were collected in the area of the Brazilian Agricultural Research Corporation (Embrapa Amazônia Oriental), located in Belém, State of Pará, Brazil (01° 26' 44.2" S, 48 ° 25' 03.8" W). These seeds were seeded in 5-L polyethylene trays containing sand and sterilized sawdust (1:1, v/v), and maintained at medium air temperature ( $T_{air}$ ) and relative humidity (RH) of 28 °C and 90%. After emergence, seedlings containing the first pair of eofilos were transplanted into 10-L polyethylene pots containing yellow latosol and avian bed (3:1, v/v). The seedlings were grown in a greenhouse for 180 days, and irrigated daily to replace the water lost by evapotranspiration.

Subsequently, the young plants were removed and their roots washed with deionized water and transferred to 5-L Leonard pots containing sterilized and washed sand and 800 mL of nutrient solution of

Sarruge (1975), replaced weekly and constituted of ( $\mu\text{M}$ ):  $\text{KH}_2\text{PO}_4$ , 400;  $\text{KNO}_3$ , 2000;  $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$ , 2000;  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ , 800;  $\text{FeEDTA}$ , 400;  $\text{H}_3\text{BO}_3$ , 400;  $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$ , 400;  $\text{ZnCl}_2$ , 400;  $\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$ , 400 and  $\text{H}_2\text{MoO}_4 \cdot \text{H}_2\text{O}$ , 400. The pH was maintained at  $5.9 \pm 0.2$ , using HCl and NaOH. The ionic strength was initiated in 25% (10 days) and increased to 50% (35 days), remaining for a period of acclimatization of 45 days.

## ***2.3 Experimental design and treatments evaluation***

After 45 days of cultivation, we selected the most uniform seedlings considering height, stem diameter and number of leaves and subjected to five Cd concentrations (treatments) as follows:  $0 \text{ mg L}^{-1}$  of  $\text{CdCl}_2$  (control), 15, 30, 45, and  $60 \text{ mg L}^{-1}$  of  $\text{CdCl}_2$ . The doses of Cd were determined based on the Resolution 420 of the National Council of the Environment, CONAMA (Brazil, 2009), which establishes criteria and guiding values of soil quality regarding the presence of chemical substances. The experimental design was completely randomized, with seven replicates, totaling 35 experimental units. A single plant per pot was considered a replicate. All variables for treatment comparisons were evaluated 60 days after differentiation of treatment with Cd.

## ***2.4 Growth parameters***

The height of the plant was measured, from the base of the collection to the apical bud of the plant (cm), stem diameter, measured at 4 cm in relation to the neck, using a ZAAS precision digital caliper (cm), the number of leaves was obtained by counting; for the determination of the dry matter, the plants of each treatment were taken to the Laboratory of Estudo da Biodiversidade em Plantas Superiores (EBPS), located in UFRA, where they were separated into root and aerial parts and packed in paper bags of known mass for later drying in a forced ventilation oven at  $65 \text{ }^\circ\text{C}$  until constant mass was obtained.

Each part of the plant was weighed in an analytical balance to determine root dry mass (RDM), dry mass of the stem (DMS), dry mass of leaves (DML), dry mass of aerial parts (DMAP) and total dry mass (TDM), determined by the sum of RDM and DMAP. With the values of RDM and DMAP the relation between RDM and DMAP (R/AP) was calculated. After weighing, the dry matter was milled and stored in Falcon tubes and later used in biochemical analysis. Some of the dried material was taken to the Museu Paraense Emílio Goeldi (MPEG) to analyze the concentration of micro and macronutrients in the roots and leaves of the plants.

## ***2.5 Macro and micronutrients analysis***

Macro and micronutrient analysis were processed in triplicate. Magnesium (Mg), Calcium (Ca), Iron (Fe), Zinc (Zn) and Manganese (Mn) were determined according to the methodology described by Miyazawa et al. (2009). The dry matter (0.5 g) of each sample was digested in a digester tube with 8 mL of nitric acid solution ( $\text{HNO}_3$ ) + perchloric acid ( $\text{HClO}_4$ ) (3:1). After cooling, the solution in the tube was filtered and diluted with deionized water to a final volume of 50 mL. The Mg, Ca, Fe, Zn and Mn content were determined in this solution by atomic absorption spectrometry (Thermo Scientific ICE 3000). Nitrogen (N), Phosphorus (P) and Potassium (K) were analyzed according to the methodology proposed by Tedesco et

al. (1995). Samples of 0.2 g of dry matter were submitted to digestion with sulfuric acid ( $\text{H}_2\text{SO}_4$ ). The Nitrogen (N) was determined for titulation with  $\text{H}_2\text{SO}_4$  0,0025 M, after distillation. The P was determined by spectrophotometry and K by flame photometry.

## **2.6 Data analysis**

The experimental data were evaluated for the normality and homogeneity of variances by Shapiro-Wilk and Bartlett tests, respectively. For the parametric variables, the means of the treatments were submitted to the PROC GLM, HSH test post hoc of Tukey utilizing the software SAS 9.1.3 (SAS, 2007). For the non-parametric variables, data were evaluated by the Kruskal-Wallis test with Bonferroni correction by the software RStudio version 1.1.383. The experimental data of all analysis were evaluated at 5% of significance.

## **3. Results**

### **3.1 Cd effect on growth parameters**

During the experiment, symptoms of metal phytotoxicity was observed as minor and necrotic roots regardless of metal concentration (Fig. 1A, B). In addition, they exhibited smaller leaves and symptoms of intercostal chlorosis (Fig. 1A, B).

The height, diameter, number of leaves and biomass of all plants exposed to Cd for 60 days were significantly lower than those of the control group (Fig. 2A). The growth in height of the plants treated with Cd (60 mg Cd) was 56.8% smaller in relation to the control (Fig. 2A). In the same treatment (60 mg Cd), stem diameter and number of leaves were 53.6 and 62.6% smaller in relation to the control, respectively (Fig. 2B, C). In comparison to the control treatment (0 mg Cd) the RDM, SDM, DML and TDM decreased by 60.4, 57.5, 48.0 and 62.5%, respectively, in concentrations of 60 mg Cd (Fig. 3).

The DMAP and R/AP were reduced by 53.1 and 15.4%, respectively (Fig. 3) in the highest Cd concentration (60 mg) compared to the control (0 mg Cd). All plants survived until the end of the experiment.

### **3.2 Cd effect on macro and micronutrient absorption**

The concentrations of Fe and Mg in plants submitted to Cd stress were significantly lower than those in the control group (Fig. 4), except for Fe in the leaves at the dose of 45 mg Cd (Fig. 4B). The lowest Fe values were 35.6 and 5.6% in the roots ( $15 \text{ mg L}^{-1} \text{ Cd}$ ) and leaves ( $30 \text{ mg L}^{-1} \text{ Cd}$ ), respectively, in relation to the control treatment. The Mg reduced to 38.1 and 21% in the roots ( $45 \text{ mg L}^{-1} \text{ Cd}$ ) and leaves ( $60 \text{ mg L}^{-1} \text{ Cd}$ ), respectively, in comparison to the control. Cd significantly affected Ca concentrations in plants (Fig. 4). The highest Ca reductions were 36.3 and 20.7% in the roots ( $60 \text{ mg L}^{-1}$ ) and leaves ( $45 \text{ mg L}^{-1} \text{ Cd}$ ), respectively, in comparison to the control (Fig. 4).

Compared with the treatment without Cd, Zn increased significantly in the roots (Fig. 5D), reaching values of 42.1% in the 30 mg L<sup>-1</sup> Cd. In the leaves, Zn increased significantly at the dosages of 45 and 60 mg L<sup>-1</sup> Cd (Fig. 5B), reaching the value of 17.5% (60 mg L<sup>-1</sup> Cd) compared to the control. The Mn significantly reduced in the root and increased significantly in the leaves, except for the dosage of 30 mg L<sup>-1</sup> Cd (Fig. 5). In the roots Mn reduced to 43.5% (60 mg L<sup>-1</sup> Cd) in comparison to control. In leaves, the largest reduction in Mn was 33% (40 mg L<sup>-1</sup> Cd) compared to control.

Cd significantly affected N, P and K, both in roots and leaves (Fig. 6). In the roots, the lowest values of N, P and K were respectively, 29.4 (60 mg L<sup>-1</sup> Cd), 46.1 (15 mg L<sup>-1</sup> Cd) and 7.8% (45 mg L<sup>-1</sup> Cd) in relation to control. In leaves, the biggest reduction of N, P and K was respectively of 39.6 (45 mg L<sup>-1</sup> Cd), 63.8 (60 mg L<sup>-1</sup> Cd) and 38% (45 mg L<sup>-1</sup> Cd) in relation to control.

## 4. Discussion

The negative effect of Cd on growth parameters in *V. surinamensis* (Figs. 1 and 2) are in agreement with those obtained in other tree species (Zouari et al. 2016; Pereira et al. 2017; Nikolić et al. 2017).

The toxicity of Cd in *V. surinamensis*, more pronounced in the roots, evidenced by inhibition of its growth, especially at higher concentrations of the metal (Fig. 1A), can be explained by the direct effect of Cd and its greater accumulation in this organ of the plant (Andrade Júnior et al. 2019). In fact, in many species of plants, including the *V. Surinamensis* Cd is mainly accumulated in the roots and, to a lesser extent, in the aerial part (Pereira et al. 2017; Andrade Júnior et al. 2019). This has caused root growth retardation, suberization, damage to internal and external root structures (Dai et al. 2013). The highest negative effect of Cd on the root can also be attributed to changes in cellular redox balance by the increase of reactive oxygen species (EROS) (Yan et al. 2015, Singh et al. 2016; Zouari et al. 2016) which resulted in cell death and defects in root growth and development in the zone of elongation and meristem (Abozeid et al. 2017). However, the reduction of the root system of this species was not limiting for its survival during the period of exposure to Cd.

On the other hand, the lower growth of roots, stem and leaves (Figs. 1 and 2) in *V. surinamensis* under Cd resulted in reduction of biomass production (Fig. 3) due to changes in photosynthesis and reduction in nutrient absorption that resulted in the low synthesis of photoassimilates. Reductions in the biomass related to the lower photosynthetic activity under Cd effect were observed by Chaves & Souza (2014). Depending on concentration, Cd may interfere with the uptake, transport and use of mineral ions by plants (Di Baccio et al. 2014). Therefore, it is suggested that Cd possibly competed with the ions by the same capture system, affecting the absorption and assimilation of the macro and micronutrients resulting in the loss of photosynthetic activity and, consequently, the reduction of dry mass (Fig. 3) in *V. surinamensis*. Otherwise, Cd may have affected the meristematic cells of the root and shoot, causing a decrease in the dry mass of these organs (Abdul Qados 2015). Biomass is the main indicator of energy accumulation in plants (Zang *et al.* 2014). Therefore, the survival of the plant under conditions of stress by Cd is dependent on a balance in the distribution of photoassimilates among its various parts. The

lower shoot root ratio in *V. surinamensis* under the effect of Cd (Fig. 3) indicates that the growth of the root system was more strongly reduced than the aerial part. This may be explained by the higher accumulation of Cd in *V. surinamensis* root (Andrade Júnior et al. 2019), which may have contributed to the lower absorption and transport of macro and micronutrients (Figs. 4, 5 and 6). On the other hand, the lower ratio of root biomass to shoot can be a strategy of tolerance to the metal, reserving less energy to the roots to reduce the absorption of Cd and the greater energy investment in the leaves for the maintenance of the vital functions. Similar results were observed in other tree species exposed to Cd (Abdul Qados 2015, Silva et al. 2017, Nikolić et al. 2017).

Reduction of minerals like  $Mg^{2+}$  (Di Baccio et al. 2014, Liu et al. 2015, Wang et al. 2016, Zouari et al. 2016),  $Ca^{2+}$ ,  $Fe^{2+}$ ,  $Mn^{2+}$  e  $Zn^{2+}$  in plants treated with Cd, probably occurs because  $Cd^{2+}$  competes with the membrane transporters of these minerals (He et al. 2013). In this study, the lower concentration of Mg and Fe in plants exposed to Cd (Fig. 4) indicates that the heavy metal interfered in the absorption of these nutrients in the root and in the transport to the aerial part of the plant. Probably, Cd competed with these minerals via membrane carriers (He et al. 2013), limiting the availability of Mg and Fe in the plant. In addition, Cd may have inhibited iron chelate reductase and interfered with Fe uptake (Parmar et al. 2013). Thus, in *V. surinamensis* exposed to Cd, the reduction of Mg and Fe may have negatively affected chlorophyll molecules, resulting in the symptoms of interveinal chlorosis (Fig. 1) and, possibly, the decrease of photosynthesis. This is because Mg and Fe, essential nutrients for chlorophyll biosynthesis (Di Baccio et al. 2014, He et al. 2013, Huang et al. 2015) may have been replaced by Cd. (Baxter *et al.* 2006). In the present work, it is possible to identify the presence of the chloroform molecule (Bashir *et al.* 2006). This may lead to an adverse effect on chlorophyll metabolism and, subsequently, the reduction of photoassimilates and the lower growth of the plant. Furthermore, Fe deficiency in plants treated with Cd negatively affects the multiprotein complex (MPCs), including photosystem II and I, LHC, Cytb6f and ATPase, (Basa et al. 2014, Bashir et al. 2015), which holds on to many electron carriers. This occurs, at least in part, because Cd replaces Iron (Fe) from its interaction with S (S) (Fe-S) sulfur in MPCs proteins (Bashir et al. 2013), possibly altering biological activity of proteins and affecting the transport of electrons to ferredoxin, resulting in the reduction of photosynthesis. On the other hand, the lower accumulation of Fe in the plants with Cd may have been a strategy of protection of the biomembranes against EROs. This is because it has been reported that the increase of Fe in plants exposed to Cd leads to the destabilization of membranes by the synthesis of lipoxygenase enzyme, since it is directly involved in the production of ROS through the reaction of Fenton and Haber-Weiss (Kumar et al. 2018). Di Baccio et al. (2014) studying tree species submitted to Cd, observed increase in Fe and Mg reduction.

Ca acts as a secondary messenger that modulates the activity of a variety of proteins (Eller and Brix 2016). Therefore, the Ca dislocation of the calmodulin protein by Cd may interfere with its ability to function correctly in signal transduction and transcriptional regulation (Dal Corso, Manara, and Furini 2013). Ca is also an essential cofactor of the inorganic catalytic core ( $Mn_4CaOxCl_y$ ) in photosystem II (PSII) and plays an important role in the stability of chlorophyll (Huang *et al.* 2017), in the electron flux of photosystems and light dependent metabolism reactions (Hochmal et al. 2015). In addition, Ca is an

essential element for the growth and development of plants (Huang et al. 2017). Therefore, it is suggested that the Cd may have substituted Ca during catalytic core formation and affected the photochemical efficiency of PSII or by competition, reducing Ca uptake by the roots, resulting in the decrease of the chlorophyll molecule and affected the photosynthetic activity of the plant, which negatively influenced the height, root growth and biomass production of *V. surinamensis* (Figs. 2 and 3). Reduction of  $\text{Ca}^{2+}$  was evidenced in other tree species exposed to Cd (Di Baccio et al. 2014).

It has been reported that Cd can damage plant DNA through the activation of restriction enzymes and / or due to the production of oxidants such as hydroxyl (OH) radical (Paunov et al. 2018). In addition, Cd can displace essential cofactors, such as Mn and Zn, and bind to functional groups (sulfhydryl, -SH) of proteins and enzymes and cause inactivation or denaturation of these organic compounds (Dal Corso et al. 2013) and may lead to various metabolic disorders (Yan et al. 2015). Thus, it is suggested that the increase of Zn (Fig. 5) in roots and leaves of *V. surinamensis* subjected to Cd stress would be a plant response to DNA protection by inhibition of endonucleases and the OH radical, or for protection of the -SH group, possibly to minimize oxidative damage caused by Cd to the genetic material and plant proteins. Cd stress induces changes in cellular redox balance resulting in increased ROS, such as the superoxide anion ( $\text{O}_2 \cdot^-$ ) (Zouari et al. 2016) that can cause oxidation of membrane lipids, proteins and nucleic acids, changes in structure (Wierke *et al.* 1995) and electrolyte leakage (ZOUARI et al. 2016). Thus, the increase of Zn in *V. surinamensis* may have played an important role in the synthesis of antioxidant enzyme, since it constitutes the cofactor of Zn-SOD associated with chloroplast (Nagajyoti et al. 2010). In a way, it could, at least in part, regulate the cellular redox potential and sustain or restore the PSII reaction center and the photosynthetic activity of the plants (Solti et al. 2016). Contrasting Zn ratio was observed in other tree species submitted to Cd (Di Baccio et al. 2014). Transport of mineral elements from soil to different tissues of plants requires different types of membrane transporters (Sasaki et al. 2016). Membrane transport proteins of the ZIP and Nramp family are involved in the uptake and translocation of Zn and Mn from the root to the aerial part of the plant at different levels (present patterns of different expressions depending on the tissues), and the Cd can inhibit these transporters (Wu et al. 2016; Akhtar et al. 2017) or Cd can compete with Zn and Mn via the cell membrane through the same uptake sites (Printz et al. 2013). In this study, the increase in Zn concentration in the root and leaves (Fig. 5) suggests that Cd did not interfere with the membrane transporters of this mineral. On the other hand, the reduction of the root Mn and the increase of this mineral in the leaves of the plants exposed to the Cd (Fig. 5), may be due to different expressions of the Mn transporters. Thus, Cd possibly may have affected the expression of the membrane transporters for Mn in the root, but no effect on the air tissue transporters of *V. surinamensis*. The lower concentration of Mn in the root of *V. surinamensis* exposed to Cd may have affected the growth and functionality of the root system. However, Mn is required in the water oxidation reaction in PSII (Schmidt et al. 2016) and its increase in Cd plant leaves (Fig. 5B) may have been a strategy to maintain stability and activity photosynthesis of PSII, although affected by Cd (Andrade Júnior et al. 2019). In addition, Mn for its role as a cofactor was possibly essential in the production of the antioxidant enzyme Mn-SOD associated with glyoxysomes (Nagajyoti et al. 2010). Mn reduction was evidenced in other tree species exposed to Cd (Printz et al. 2013).

A reduction of nitrogen (N), phosphorus (P) (He et al. 2013) and potassium (K) has been observed in plants exposed to Cd (Gomes et al. 2013). Nitrogen (N) is an essential macronutrient because it is the main constituent of many structural, genetic and metabolic compounds in plants (Kulcheski et al. 2015), such as amino acids, proteins, nucleic acids, vitamins and hormones, which play an important role in general plant growth (Singh et al. 2016). Research has shown that Cd negatively affects nitrogen metabolism due to activation or inactivation of proteins and enzymes involved in the uptake, transport and assimilation of N, resulting in reductions of N in plant tissue (Nikolić et al. 2017). Therefore, it is suggested that the reduction of N (Fig. 6) in *V. surinamensis* subjected to Cd doses may have caused changes in nitrogen metabolism, with a negative effect on growth (Fig. 2) and on the production of plant biomass (Fig. 4). On the other hand, N-reduction may have occurred because of its use in amino acid synthesis, such as proline, to form non-toxic Cd-proline complex in plant tissues to reduce metal phytotoxicity (Chen *et al.* 2001, Aslam et al. 2014).

P is a constituent of nucleic acids and phospholipids of the cell membrane and is indispensable for the phosphorylation reaction (Singh et al. 2016). P is important in several metabolic and physiological processes, such as the synthesis of pigments, proteins and enzymes, energetic and photosynthetic metabolism, plant growth and development (Kumar et al. 2018). Studies point to an antagonistic effect between Cd and phosphate in the solution (Cui et al. 2016). One possible explanation is that the Cd associates with the phosphate ions ( $\text{H}_2\text{PO}_4^-$ ) forming insoluble complexes  $\text{Cd-H}_2\text{PO}_4$ , which would limit the mobilization and bioavailability of  $\text{H}_2\text{PO}_4^-$  (Siebers et al. 2013). Therefore, it is suggested that reduction of P in Cd-treated *V. surinamensis* occurred due to the sorption of Cd to P in the solution or in the cellular and subcellular compartments of the plant, which limited the availability of P (Fig. 6), as reported in other studies (Degola et al. 2014). This could possibly have interfered in the synthesis of ATP and consequently in the intracellular energy pathways (Dal Corso et al. 2013) or the formation of RubisCo, affecting the photosynthesis by limitation of the carboxylation capacity of the enzyme, resulting in the lower production of photoassimilates (Singh et al. 2016), thus causing less plant growth (Fig. 2). On the other hand, P reduction may be related to sugar synthesis through the pentoses-phosphate cycle to eliminate or keep ROS levels under control and thus repair the toxic effects of oxidants (El-Beltagi and Mohamed 2013).

The K is an essential macronutrient involved in several signaling pathways (Kulcheski et al. 2015), as in cell elongation, in osmoregulation. It activates a complex of several enzymes that aid in stomatal movement, photosynthesis, synthesis of soluble carbohydrates, protein and compounds containing soluble nitrogen (Ahmad *et al.* 2016, Singh et al. 2016). It is suggested that the reduction of K in *V. surinamensis* on Cd effect may have caused changes in the osmotic potential and interfered in water potential, in the translocation of mineral ions and amino acids or inactivating enzymes involved in photosynthesis resulting in reduction in vegetative growth (Fig. 2). The results of the present study are in agreement with those obtained by Kapoor et al. (2013).

## 5. Conclusion

Cadmium affected the uptake and translocation of Fe, Mg, Ca, N, P and K, and negatively interfered with the growth and biomass production of *V. surinamensis*.

The increase in Zn concentration in the root and leaves suggests that Cd did not interfere with the membrane transporters of this mineral.

The lower concentration of Mn in the root of *V. surinamensis* exposed to Cd may have affected the growth and functionality of the root system.

## **Declarations**

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### **Ethics approval and consent to participate**

not applicable

### **Consent for publication**

we consent to the publication of the manuscript.

### **Availability of data and materials**

not applicable

### **Competing Interest**

The authors declare that there is no conflict of interest publishing of the paper, that the paper has been not published elsewhere, and not include any form of plagiarism. All the authors mentioned above have approved the manuscript and have agreed with the submission of the manuscript.

On behalf of all authors, the corresponding author states that there is no conflict of interest.

### **Availability of data and material (data transparency)**

Under the domain of the corresponding author

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### **Authors' Contributions**

A.J. : Installation and application of experiment in a greenhouse;

O. N. : and G.N: Biochemical analysis of the samples in a Plant Physiology laboratory;

S. F. : production of data figures

E. C. : Collection of plant material (seeds) and storage;

C. A. : Biochemical Analysis;

S. V. : Statistical analysis of the data;

V. B. : Translation of the scientific text;

D. S and J. T. : Withdrawal of the greenhouse experiment

## statement

all authors have read and approved the manuscript

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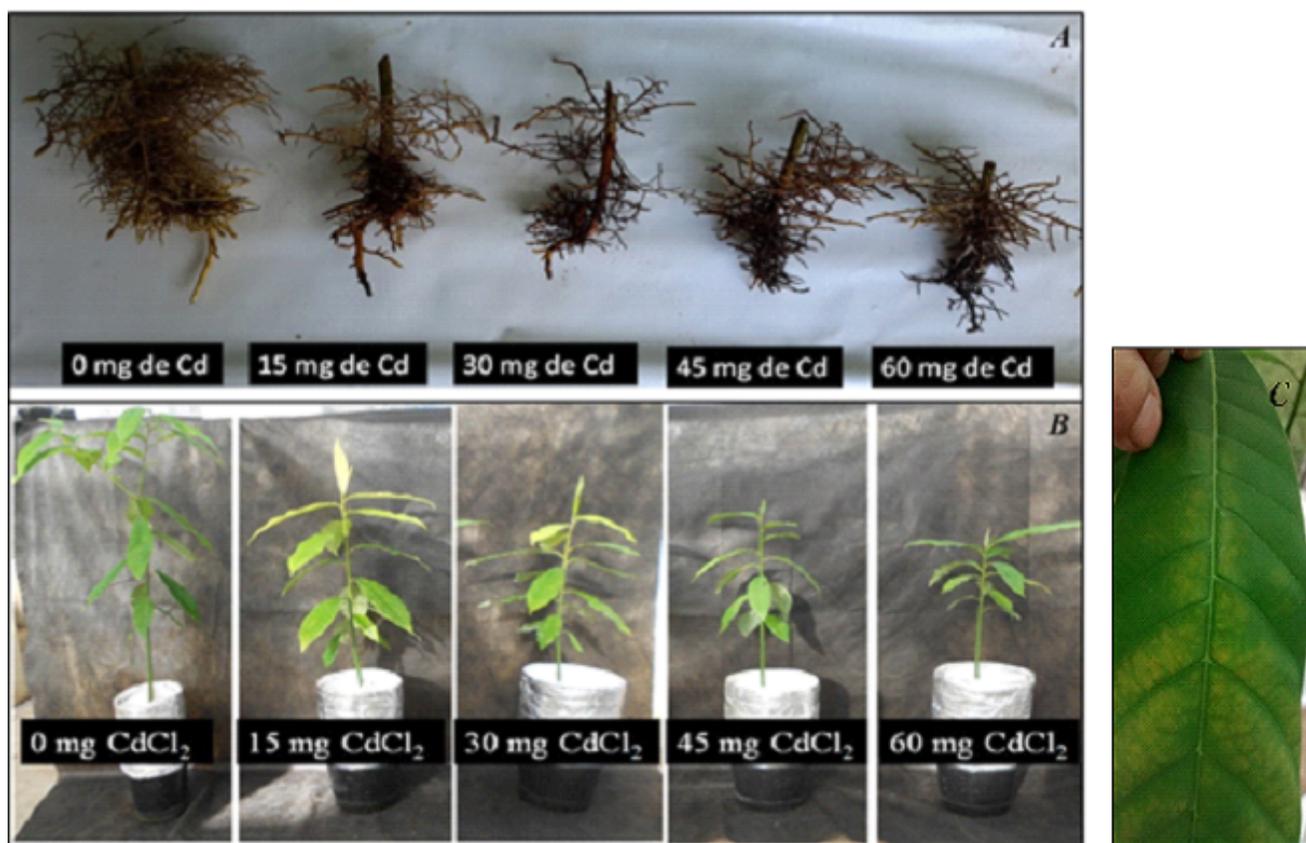
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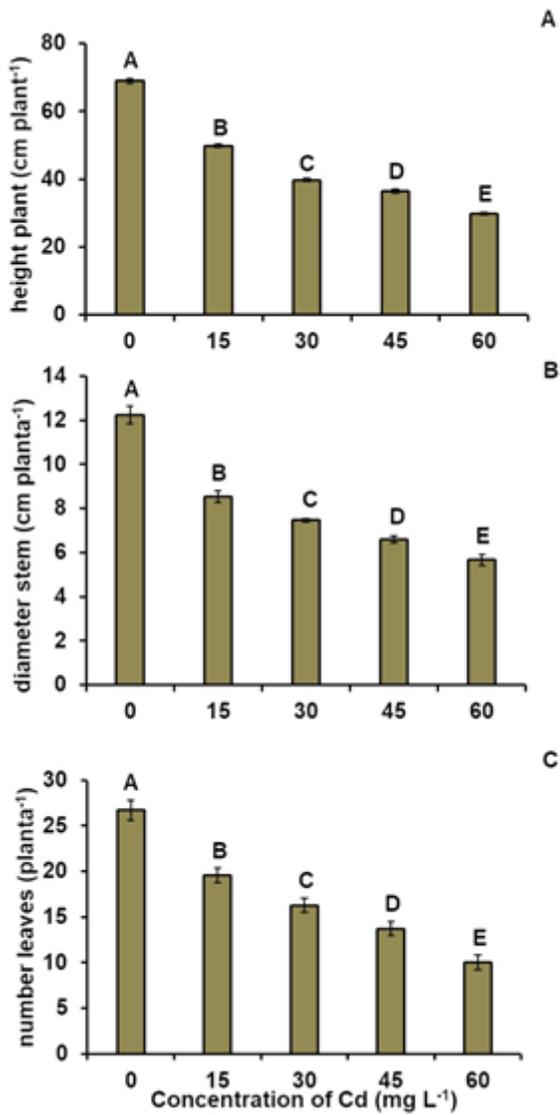
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## Figures



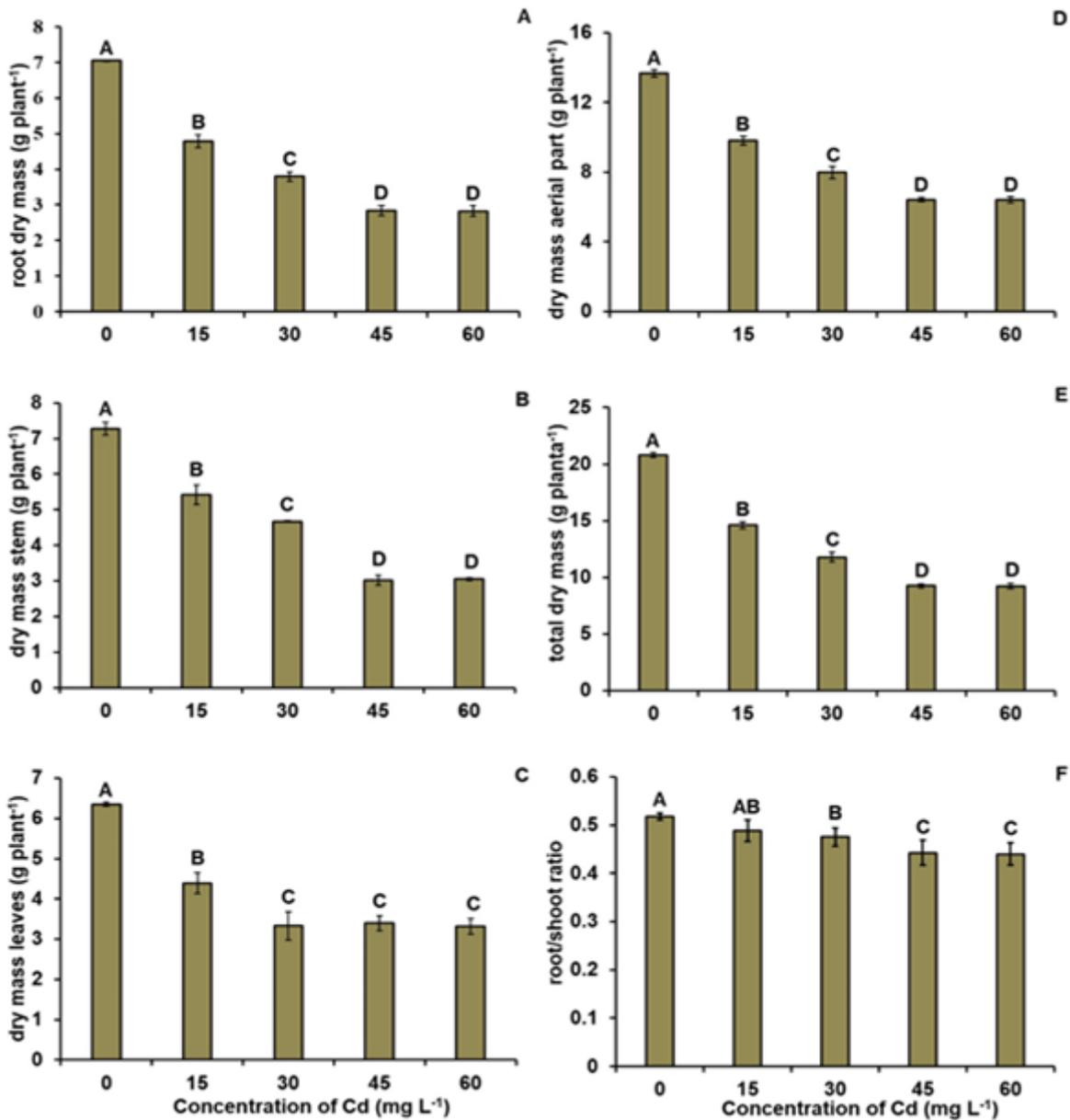
**Figure 1**

A: Minor and necrotic roots, B: minor and chlorotic leaves, C: detail of intercostal chlorosis in *V. surinamensis* after 60 days of exposure to cadmium.



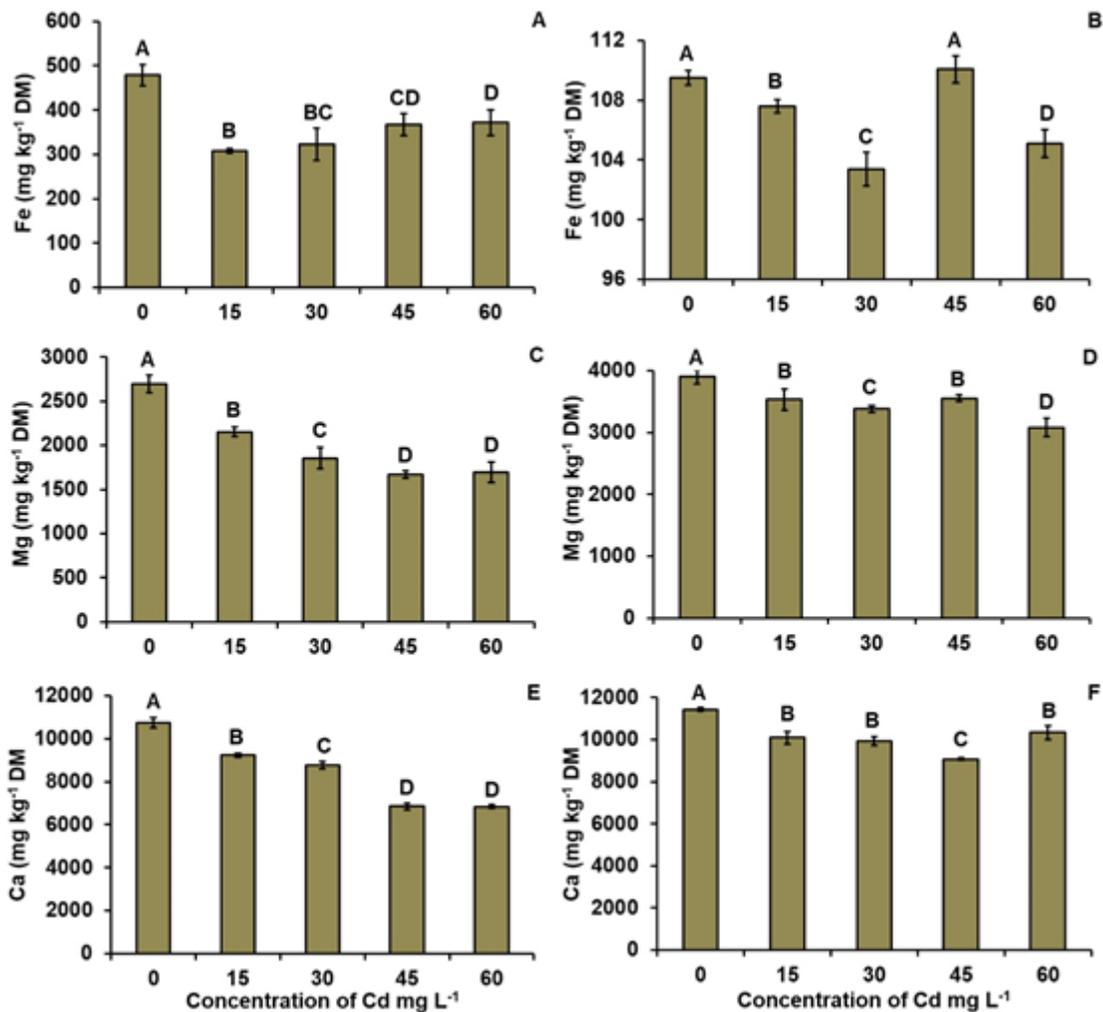
**Figure 2**

A: Height, B: diameter, C: number leaves in young plants of *V. surinamensis* exposed to five concentrations of cadmium (0, 15, 30, 45, and 60 mg). Different letters for concentrations of cadmium in solution indicate significant differences in the Tukey's test ( $P < 0.05$ ). Mean  $\pm$  SD,  $n = 7$ .



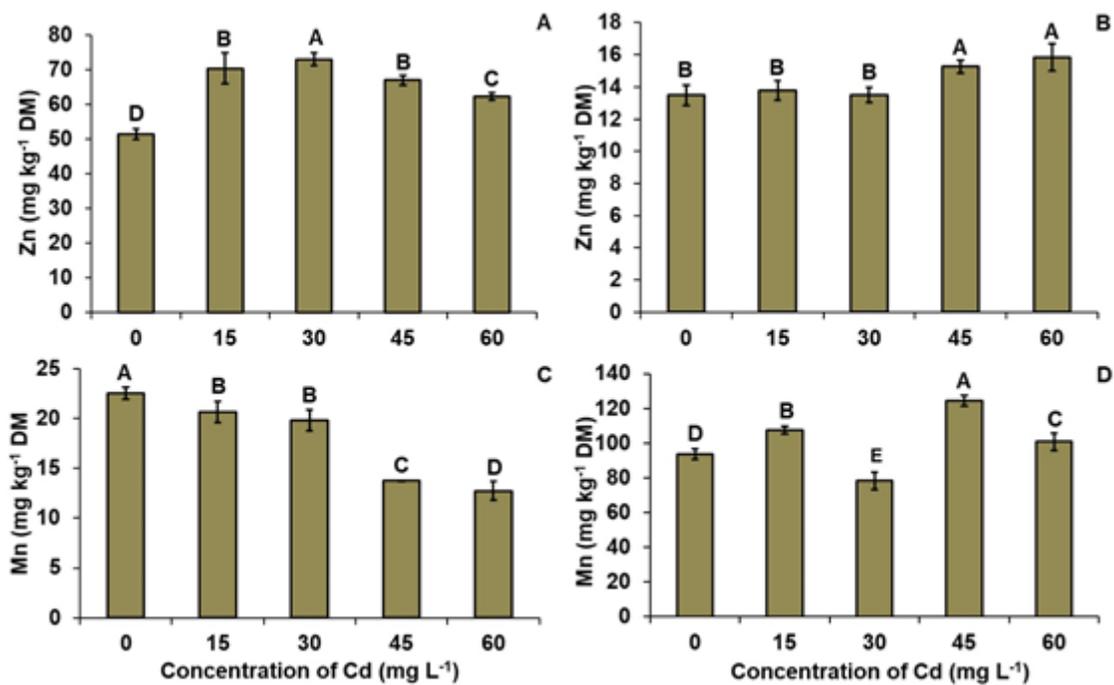
**Figure 3**

A: root dry mass, B: dry mass stem, C: dry mass leaves, D: dry mass aerial part, E: total dry mass, F: root/shoot ratio in young plants of *V. surinamensis* exposed to five concentrations of cadmium (0, 15, 30, 45, and 60 mg). Different letters for concentrations of cadmium in solution indicate significant differences in the Tukey's test ( $P < 0.05$ ). Mean  $\pm$  SD,  $n = 7$ .



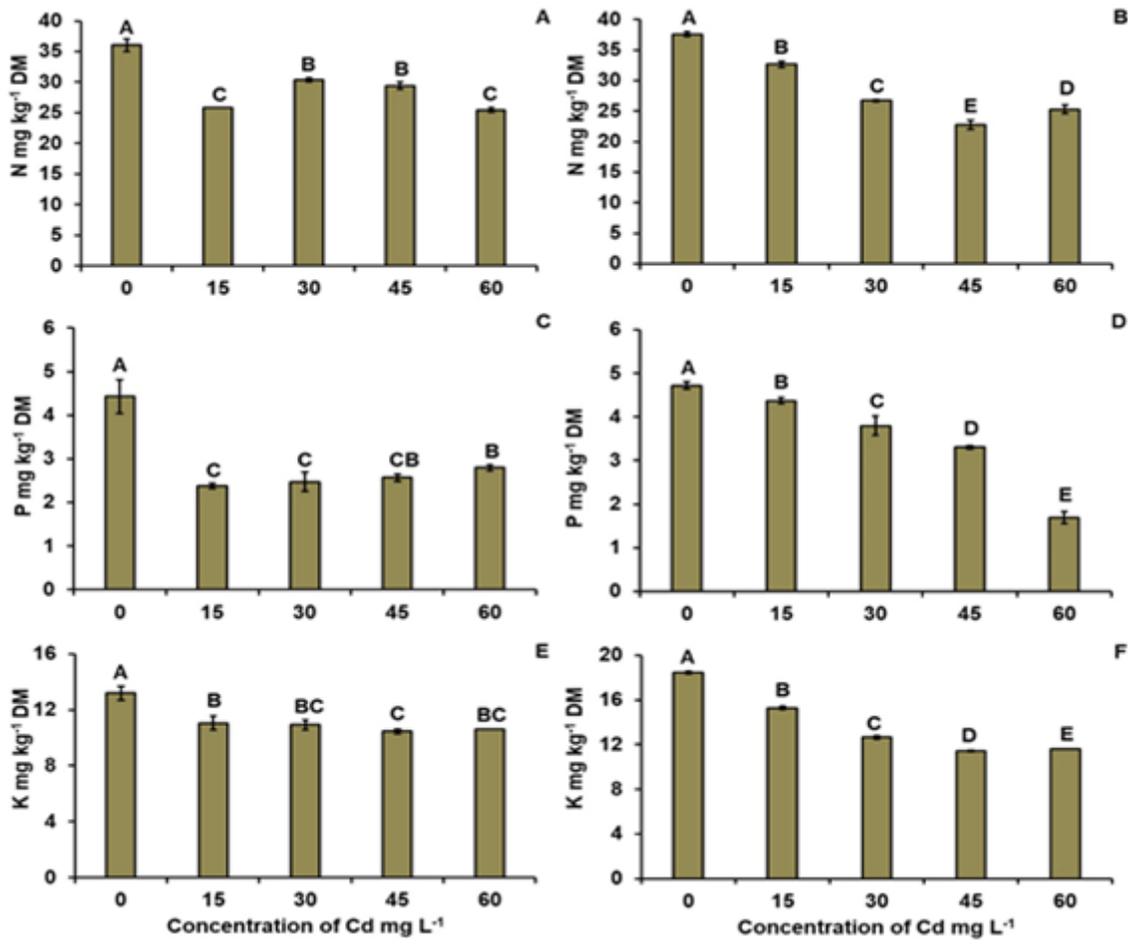
**Figure 4**

A: Fe concentration in the root, B: Fe concentration in the leaves, C: Mg concentration in the root, D: Mg concentration in the leaves, E: Ca concentration in the root, F: Ca concentration in the leaves of young plants of *V. surinamensis* exposed to five concentrations of cadmium (0, 15, 30, 45, and 60 mg). Different letters for concentrations of cadmium in solution indicate significant differences in the Kruskal-Wallis ( $P < 0.05$ ). Média  $\pm$  DP,  $n = 7$ .



**Figure 5**

A: Zn concentration in the root, B: Zn concentration in the leaves, C: Mn concentration in the root, D: Mn concentration in the leaves of young plants of *V. surinamensis* exposed to five concentrations of cadmium (0, 15, 30, 45, and 60 mg). Different letters for concentrations of cadmium in solution indicate significant differences in the Kruskal-Wallis ( $P < 0.05$ ). Média  $\pm$  DP,  $n = 7$ .



**Figure 6**

A: N concentration in the root, B: N concentration in the leaves, C: P concentration in the root, D: P concentration in the leaves, E: K concentration in the root, F: K concentration in the leaves of young plants of *V. surinamensis* exposed to five concentrations of cadmium (0, 15, 30, 45, and 60 mg). Different letters for concentrations of cadmium in solution indicate significant differences in the Tukey (P < 0.05). Média ± DP, n = 7.