Review

Toxicity of Cadmium in plants and strategies to reduce its effects. Case study: The tomato

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ABSTRACT

Cadmium (Cd) is one of the most toxic heavy metals. Its high mobility and bioaccumulative power differentiate it from the rest of its group and motivate the interest of scientists to know its effects and interaction with plants. In the present work, a bibliographic review of the main mechanisms of entry and transport of the Cd in the plants and their toxic effects in them was carried out. Also, issues such as the defense mechanisms of plants against Cd stress and existing strategies to reduce their toxicity are addressed. Within the different crops, tomato is of special interest, because it is the most widespread vegetable in the world and has been shown to be a plant tolerant to Cd and with potential for its accumulation.

Key words: heavy metals, relationship, vegetables
INTRODUCTION

Cadmium is a highly toxic transition metal at very low exposure levels and has acute and chronic effects on the health of plants, animals, humans and all living things in general. Because of industrial activity and anthropization, it is estimated that 30,000 tons of Cd are released into the environment every year \(^{(1)}\). Therefore, in different parts of the planet and in our country, Cd levels have been in water, soil and plants, detected that exceed the permissible limits established for different uses \(^{(2-7)}\).

Cadmium is not degradable in nature, so once released to the environment it will remain in circulation. This property together with its high mobility, bioaccumulative power and toxicity at very low concentrations make it one of the most important heavy metals. In the 1960s, environmental contamination with this metal became apparent when in Japan more than 100 people died from a disease named Itai-Itai, which was caused by high concentrations of Cd in the Jinzu River, in rice \((4.2 \text{ mg L}^{-1})\) and consequently in the human body \(^{(8)}\). These facts motivated the interest of soil and plant science to know and control the effects that metal produced in different crops.

This metal is as one of the most toxic and inhibitory of the physiological processes of plants recognized. Studies in several crops have shown that it reduces growth, photosynthetic activity, perspiration and chlorophyll content \(^{(9-12)}\). In addition, it causes chlorosis, oxidative stress, and nutritional imbalances and modifies the activity of enzymes, involved in the metabolism of organic acids and in the Krebs cycle \(^{(13-16)}\). In general, the effects caused in some physiological processes can be so marked that plants are not able to evade them and manifest themselves in other processes. The toxicity of Cd can lead to the death of the plant and this depends, among other factors, on the exposure time, the metal content and the specific adaptations they develop.

The specific adaptations of plants to Cd stress are two main mechanisms based on. Some prevent or regulate the entry and transport of the same \(^{(17)}\) and others tolerate certain contents of Cd, through its detoxification, by chelation in intracellular organelles \(^{(18)}\). Based on these tolerance mechanisms, several research groups have proposed different strategies to lessen the effects of Cd on plants. Most strategies include making changes in nutrition management \(^{(19)}\). Nevertheless, other practices have also shown favorable results, such as inoculation with beneficial bacteria \(^{(20)}\), grafts on resistant patterns \(^{(21, 22)}\), addition of different growth regulators \(^{(23, 24)}\) and application of amendment in the soil.
Knowing the interaction of Cd with plants, as well as the search for alternatives to minimize its effects have caught the interest of the scientific community, product of the accelerated growth of contamination with this metal and its high toxicity. The objective of this study is to make an updated review of research results related to these aspects.

Within the different crops, tomato is of special interest, because it is the most widespread vegetable in the world and of greater economic value. It has been not only as food used, but also as a model plant in dissimilar research. The tomato plant has many interesting features, such as fleshy fruit, a sympodial bud and compound leaves, which other model plants (for example, rice and Arabidopsis) do not have \(^{(25)}\). In addition, some of its varieties have shown to be a Cd tolerant plant, with potential for accumulation \(^{(26)}\).

**Cadmium absorption and transportation in plants**

Cd enters the plant mainly in the form of Cd\(^{2+}\), since its chelated ions are generally not available for root absorption. The epidermal cell layer is the first tissue for ion uptake and within it; radical hairs are the most active area to absorb ions from the soil and is the structure that facilitates the absorption of Cd\(^{2+}\) \(^{(27)}\).

Three different routes of Cd entry into the root have been proposed \(^{(28)}\):

*First way:* in the plasma membrane of the epidermal cells of the root, CO\(_2\) (ac) dissociates into H\(^+\) and HCO\(_3^-\), through plant respiration. The H\(^+\) is with the Cd\(^{2+}\) exchanged, of the soil and the metal adsorbs on the surface of the epidermal cells of the root. This adsorption process is rapid and does not require energy and is the stage preceding the subsequent absorption of Cd\(^{2+}\) in the epidermis through the apoplast pathway.

*Second way:* the Cd is a non-essential element and, therefore, it is assumed that the plants do not have specific input mechanisms for it. It enters the plant cells through the Fe\(^{2+}\), Zn\(^{2+}\) and Ca\(^{2+}\) essential metal transporters, as is the case with IRT1 and LCT\(_1\) proteins. After combined with the transporter proteins, the Cd enters the epidermis layer of the root, through the path of the simplast.

*Third way:* to increase the availability of ions in the rhizosphere soil, the roots of plants secrete compounds of low molecular mass, such as mugineic acids (MA), which form complexes with Cd\(^{2+}\). Therefore, Cd\(^{2+}\) enters the root epidermis layer through YSL-like proteins in the form of chelates.
The movement of Cd from the root to the stem is through three processes controlled: the sequestration of metals within the root cells; the transport to the wake and the release of the metal to the xylem (29). Retention is the product of apoplastic barriers and chelation in vacuoles; it demonstrated that phytokelatins and other thiols are the main chelators in the kidnapping of Cd at the root (30). Another of the proposed mechanisms of Cd retention in roots is through the impregnation of the suberine in the cell wall during the maturation of exodermis and endodermis, which affects the plasticity and restricts its movement to the wake (31).

The transfer and remobilization of the Cd from the xylem to the phloem is another crucial process in the transport of this ion. Other authors identified high concentrations of phytokelatins, glutathione and Cd in the sap of the Brassicanapus phloem and suggested that the phloem is also a conduit for the transport of the Cd-phytokeratin and Cd-glutathione complexes (32).

**Effects of toxicity per cadmium on plants**

Numerous authors have studied the toxic effects of Cd on plants. The main visible symptoms caused by Cd toxicity are chlorosis and leaf curl. Chlorosis may appear by exchanging Cd with Fe or Mg (13), in the latter case affecting the stability and biosynthesis of chlorophylls. Damages also associated with chlorosis are P deficiency and reduction in the transport of Mn (33).

The reduction in photosynthetic growth and activity, nutritional imbalance, oxidative stress and effects on enzymatic activities are the most pronounced damages that are in the different studies of toxicity with Cd frequently expressed (Table 1).
Table 1. Effects of Cd on the physiological processes of various crops

<table>
<thead>
<tr>
<th>Physiological effects</th>
<th>Plants</th>
<th>Reference</th>
</tr>
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<tbody>
<tr>
<td>Reduction in growth and development</td>
<td>Reduction of root length, leaf area and fresh root, stem and leaf biomass</td>
<td>Pepper</td>
</tr>
<tr>
<td>Root and leaf biomass reduction</td>
<td>Lettuce Radish Soy</td>
<td>(35,36)</td>
</tr>
<tr>
<td>It showed no fruit production in 90 days.</td>
<td>Tomato</td>
<td>(11)</td>
</tr>
<tr>
<td>Photosynthetic activity reduction</td>
<td>Reduction in chlorophyll content</td>
<td>Pea Potato Soy</td>
</tr>
<tr>
<td>Decrease Net Photosynthetic Rate</td>
<td>Soy</td>
<td>(39)</td>
</tr>
<tr>
<td>Reduction in the content of chlorophylls and carotenoids</td>
<td>Tomato</td>
<td>(11)</td>
</tr>
<tr>
<td>Interferes in the entry and transport of nutrients</td>
<td>Reduction of N fixation and assimilation of NH$_4^+$ in nodules</td>
<td>Soy</td>
</tr>
<tr>
<td>Reduction of total concentrations of K, P, Ca, Mn, Zn, S and B</td>
<td>Pea</td>
<td>(41)</td>
</tr>
<tr>
<td>Reduction of Mn and increase of Fe and Zn</td>
<td>Soy</td>
<td>(42)</td>
</tr>
<tr>
<td>Reduction of Zn, Mn, Ca and K in the leaf tissue</td>
<td>Tomato</td>
<td>(43)</td>
</tr>
<tr>
<td>Mn reduction in root and leaves</td>
<td>Tomato</td>
<td>(44)</td>
</tr>
<tr>
<td>Reduction of Mn, Zn, Cu, Fe and Ca in root, stem and leaves</td>
<td>Potato Lettuce Tomato</td>
<td>(45)</td>
</tr>
<tr>
<td>Increase in P, K, Ca Mg, Fe and Zn in stems and leaves (Cd-1; 2.5; and 5 mg kg$^{-1}$)</td>
<td>Onion</td>
<td>(15)</td>
</tr>
<tr>
<td>Reduction in more severe stress conditions</td>
<td>Tomato</td>
<td>(46,47)</td>
</tr>
<tr>
<td>Provokes oxidative stress</td>
<td>Increased concentrations of Malondialdehyde (MDA) and H$_2$O$_2$</td>
<td>Tomato</td>
</tr>
<tr>
<td>Increase in MDA concentrations</td>
<td>Lettuce Pea</td>
<td>(35,37)</td>
</tr>
</tbody>
</table>

In general, the presence of Cd causes varied effects on enzyme activities. Enzymes containing sulfhydryl groups are the most prone to oxidation caused by Cd, it destroys the disulfide bridges, causing protein denaturation and its consequent enzymatic activities (12,13). The Cd also causes inhibition in the activities of the metalloenzymes, due to the substitution of it with metals with similar load or size such as Zn and Mg, the latter present in the enzyme Rubisco and its exchange with the Cd results in dissociation of the enzyme in subunits (13). Several authors have suggested that Cd toxicity causes oxidative stress in plants, either by an exaggerated increase in the production of reactive oxygen species, or by a deficit in the antioxidant response (48). However, other authors believe that Cd does not act directly in the production of reactive oxygen species (49).
Stress tolerance mechanisms for Cd

The specific adaptations of plants to Cd stress are two main strategies based on; some prevent or regulate the entry and transport of the same and others tolerate certain contents of Cd, through its detoxification, by chelation in intracellular organelles \(^{(17,18)}\). Other tolerance mechanisms are, the increase of the antioxidant defense system, cell homeostasis \(^{(50)}\), the increase in endogenous production of plant growth regulators and the modification of metabolism in function of repairing the damaged cell structure \(^{(51,52)}\).

The plants prevent the entry of Cd by immobilizing it in the cell wall of the roots through links with extracellular exudates, such as polygalacturonide acids and this limits their transport to the aerial part \(^{(53)}\). Other plants have developed tolerance to stress, accumulating metals in the leaves, in the form of stable non-toxic metal complexes, with different chelants: organic acids, amino acids, ferritins, phytokelatins and metallothioneins. Studies have shown that vacuoles are the site of heavy metal accumulation including Zn and Cd \(^{(54)}\).

Within the different chelants in plants, phytokelatins have shown greater capacity to form complexes with the Cd, hence they have been subject to analysis in several tolerance studies. Plants that overexpress the enzyme phytokelatin synthase showed a greater tolerance against Cd \(^{(55)}\).

It has also been shown that plant exposure to Cd results in an increase in sulfate assimilation \(^{(56)}\) and in the activity of enzymes involved in the biosynthesis of GSH, the starting substrate in the synthesis of phytokelatins \(^{(57)}\). Two cell lines of tomato plants tolerant to Cd have been identified and their tolerance capacity depends on the potential of cells to synthesize phytokelatins, and complex with Cd \(^{(58)}\).

However, other evidence indicates that the increase in the production of phytokelatins is not responsible for the high tolerance to Cd in some plants, since both sensitive and tolerant populations produce equivalent amounts of phytokelatins when exposed to equal concentrations of Cd \(^{(59)}\). In addition to phytokelatins, other amino acids and vitamins have also shown alterations against Cd, an increase in the contents of \(\alpha\)-tocopherol, asparagine, tyrosine and proline was observed in different tomato cultivars exposed to stress by this metal \(^{(49,60)}\).

An extreme case of the accumulation strategy is that of hyperaccumulating plants, which can exceed 100 or more times the normal values of metals found in the aerial part. Plants mostly contain measurable levels of Cd mainly in the roots, but when they exceed the established
threshold of 10 µg g\(^{-1}\) (0.01 % dry mass) they are considered hyperaccumulators of this metal \(^{(61)}\). Hyperaccumulation occurs in unrelated families. Most species and those with greater absorption capacity have been found in naturally occurring sites rich in metals, among them Arabidosishalleri, Thlaspirotundifolium and praecox, Sedumalfredii, Salsolakali and Viola baoshanensis are identified, but also a large number of them grow in clean soils such as Solanumnigrum and Rorippa globosa \(^{(62)}\).

It has been shown that in tomato cultivation the accumulation and tolerance of stress due to Cd depends on the variety. The most tolerant varieties, Río Grande and other non-referred were evaluated, respectively, in other investigations for their possible use in the phytoremediation of contaminated soils \(^{(63,64)}\). In both studies, the tomato behaved like an exclusive plant, with a higher concentration of Cd in the roots (13.4 µg g\(^{-1}\) and 4.3 mg kg\(^{-1}\)).

According to research, the variety under study can only accumulate Cd in soils with low metal levels because its accumulation capacity decreases with increasing pollution. In addition, cultivating Shenbaofen-2 is tolerant and identified as a Cd hyperaccumulator, accumulated amounts of 144 and 130 mg kg\(^{-1}\) in roots and aerial tissues respectively \(^{(26)}\).

However, Tres Cantos tomato variety, originally from Tenerife, was not identified as a hyperaccumulator or with potential for phytoextraction, but the results indicated that it developed tolerance mechanisms in the processes of nutrient absorption and distribution, and preserved its growth without affect biomass production \(^{(65)}\). Other varieties showed lower tolerance than the previous ones, since they did not maintain their biomass production without affectation, but they were not as sensitive varieties identified. Within this group are varieties 4641 and Yufen 109 that accumulated respectively 2,316 and 2,237 mg kg\(^{-1}\) of Cd in fruits \(^{(66)}\). Another study in these cultivars confirmed their translocation capabilities, since Cd accumulated mainly in leaves and stems, and the 4641 variety showed greater resistance to stress than Yufen 109 \(^{(67)}\). The Ibiza F1 variety is also included within this group, with the difference that developed the exclusion as a tolerance mechanism. The highest concentration of Cd was found in the roots, but its accumulation in this organ and in leaves increased with increasing levels of Cd in solution \(^{(68)}\).

On the other hand, the Rutgers tomato variety was as a non-tolerant species of Cd and Zn identified \(^{(69)}\). In addition, the Navodaya cultivar was as sensitive to high doses of Cd identified and the flowering phase showed greater sensitivity than fruiting \(^{(70)}\).
Other authors evaluated the degree of tolerance to Cd of 10 tomato varieties and in all cases observed reductions in growth and development indicators, but each cultivar showed an inherent ability to tolerate Cd stress. Varieties K-25, K-21 and NTS-9 showed maximum resistance, the Kaveri, NbR-Uday and Swarnodya varieties were moderately affected, the Sarvodya, NBR-Uttam and Maltti varieties experienced severe damage and the S-22 variety did not survive in the presence of Cd \(^{(71)}\).

A study similar to the previous one was in 100 tomato genotypes conducted. Genotypes 9086, Roma, Sitara TS-01, pak0010990, CLN-2123A, PICDENEATO, 0.006231 and 7035 showed the best yields while genotypes 42-07, 17883, BL-1176-Riostone-1-1, MARMANDE and 17882 showed low yields in both groups the highest concentration of Cd was in the aerial tissues with respect to the root, the tolerant genotypes accumulated mainly in the buds and the sensitive ones in the fruit. Both results confirm that Cd stress tolerance has a varietal response in crops such as tomato \(^{(72)}\).

### Strategies to mitigate stress by cadmium

Due to the damage caused by Cd toxicity in plants and the risk caused by their accumulation in them, several research groups have proposed different strategies to reduce their effects. Most strategies include making changes in nutrition management. However, other practices have also shown favorable results, such as inoculation with beneficial bacteria \(^{(20)}\), grafts on resistant patterns \(^{(21,22)}\), the addition of different growth regulators \(^{(23,24)}\) and the application of amendment in soil.

Many authors suggest the optimization of nutrient management as a useful strategy to mitigate Cd toxicity. A group of authors carried out a review on the subject in 2012 \(^{(19)}\). Subsequent research in other crops and other elements continued to promote adequate nutrition as a way to mitigate Cd stress. Among the different nutrients P, K, S, Fe and Zn showed significant favorable effects. The application of P in wheat plants increased the biomass of the shoots, the leaf area, and the content of photosynthetic pigments and, in turn, favored the assimilation of other nutrients, such as K, Ca, Mg and Mn. It also increased the activity of antioxidant enzymes and decreased the content of Cd and H\(_2\)O\(_2\) in the outbreaks \(^{(73)}\).

On the other hand, the addition of K reduced the absorption and translocation of Cd in sunflower plants and inhibited the increase in membrane permeability caused by stress. However, in this study no effects were observed on organ biomass, nor on the content of
photosynthetic pigments \(^{(74)}\), although in another investigation it was proposed that K participates in the formation of photosynthetic pigments and prevents the decomposition of chlorophylls \(^{(75)}\).

Similarly, the KCl supplement in rice plants grown with high concentrations of CdCl\(_2\) increased its growth and decreased the activity of the enzyme NADPH oxidase \(^{(76)}\). However, other results showed that K deficiency protects rice plants from subsequent oxidative stress caused by Cd, as it increases the activities of antioxidant enzymes (superoxide dismutase, ascorbate peroxidase, glutathione reductase and catalase).

Unlike K sufficiency, its deficiency does not inhibit the entry of Cd into the plant \(^{(77)}\). The results showed that both K deficiency and deficiency have positive effects to mitigate Cd stress, but with different consequences each. Similar to K, the deficiency of Mg, Ca and N allows the previous activation of the antioxidant defense, but does not prevent the absorption of Cd by the roots \(^{(78-80)}\).

In the case of S, some authors suggest that it is involved in the biosynthesis of heavy metal detoxifying agents \(^{(81)}\). In mustard (Brassica juncea) the application of 30 µM and 300 µM of S reduced the impact on chlorophyll content and increased the activities of antioxidant enzymes, ascorbate peroxidase, glutathione reductase and catalase \(^{(82)}\).

Numerous studies have shown that adequate nutrition of Fe can mitigate Cd toxicity. Exogenous application of Fe decreased the concentration of Cd in tomato and rice plants \(^{(67,76)}\) and there are criteria that an adequate concentration of Fe can promote photosynthesis and perspiration, as well as increasing the dry mass of each organ in tomato plants \(^{(67)}\). On the other hand, the foliar application of the Zn-lys complex in wheat, increased photosynthesis, grain yield, enzymatic activities and reduced oxidative stress and Cd content in the different organs of the plant \(^{(83)}\).

In addition to the macro and micronutrients analyzed, there are other elements such as La, Se and Si, which have shown favorable effects in the mitigation of Cd stress. A study in two tomato varieties (var 4641 and Yufen109) enriched with Cd, showed that the application of 10 and 20 mg L\(^{-1}\) of LaCl\(_3\) reduces the concentration of Cd in leaves, stem, roots and fruits. Consequently it reduces the damage caused by Cd in growth and yield \(^{(66)}\).

Another study, which is based on pretreatment with different forms of selenium (selenocysteine, selenite and sodium selenate), induced the biosynthesis of melatonin. In
turn, the presence of Se and melatonin increased tolerance to Cd, with reductions in decreased growth, photoinhibition and loss of electrolytes (84).

The Si is not an essential nutrient considered, but it is a beneficial element to maintain growth in stressful environments. Facing the toxicity of Cd has also increased the resistance of various crops. In peanut, tomato and cucumber plants, the concentration of Cd in the shoots and leaves decreased, but the mechanisms involved in it were different for each species. In tomato and peanut the transport of Cd from the root to the leaves was reduced and in cucumber the absorption of Cd by the roots decreased (85,86).

However, other authors demonstrated that in tomato the presence of Si in the soil (25, 50 and 75 mg kg\(^{-1}\)) caused the precipitation of Cd in it and consequently the reduction of its bioavailability for root uptake (87). Applications of CaSiO\(_3\) in ornamental amaranth plants (Amaranthus hypochondriacus L) reduced the concentration of Cd in the root, stem, leaves, and increased the dry mass and the content of photosynthetic pigments. The increase in the content of Cd in the chloroplasts and the change of free ions to inactive forms sequestered in cell compartments were other advantages also associated with CaSiO\(_3\) (88).

Another study showed that the application of Si in Arabidopsis thaliana reduced the content of Cd absorbed by the plant by 50 % and increased the antioxidant enzyme activity. The proteomic analysis of this study allowed us to conclude that Si has an active participation in the mechanisms involved in tolerance to Cd (89). On the other hand, the pretreatment of wheat plants with 50-µM salicylic acid contributed to maintain plant growth at levels close to the control. This pretreatment also regulated the concentration of abscisic acid and idolacetic acid which are substances affected by Cd (90).

**CONCLUSIONS**

- Cd toxicity reduces growth, photosynthetic activity, and chlorophyll content and causes chlorosis mainly in young leaves. In addition, it interferes with the entry and transport of nutrients and causes oxidative stress and effects on enzymatic activities.
- The formation of complexes between Cd and generally sulfur proteins are the main mechanism of tolerance of plants to toxicity by this metal. Through the formation of these complexes, plants prevent or regulate the entry and transport of the Cd or detoxify the metal and tolerate certain contents in intracellular organelles.
● The response to toxicity by Cd in tomato plants depends on the variety, some varieties do not survive in the presence of Cd and others, however, are tolerant and even classify as metal hyper-accumulators.

● Nutrient management is one of the most efficient strategies to reduce the effects of Cd on plants. The studies carried out on this topic only evaluate the effects of certain independent nutrients and the simultaneous effects of the combination of several of them are not evaluated.

BIBLIOGRAPHY


