# Effects Of Heavy Metals (Cd & Cr) On Physiological And Morphological Parameters In *Zea Mays* L.: An overview

*Syed Ahtisham Masood[1], Hafiz Ghanzafar Abbas[2], Haseeb Ur Rehman[3], Said Salman[4], Qurban Ali[5] and Arif Malik[5]

[1]Cotton Research institute, Khanpur, Rahimyar Khan, Pakistan
[2]Cotton Research Institute, Ayub Agriculture Research Institute, Faisalabad Pakistan
[3]Department of Agronomy, Faculty of Agricultural Science and Technology, Bahauddin Zakariya University Multan, Pakistan
[4]Department of Plant Breeding & genetics, Ghazi University, Dera Ghazi Khan, Pakistan
[5]Institute of Molecular Biology and Biotechnology, University of Lahore, Lahore Pakistan
**Corresponding Author:** s.ahtisham01@gmail.com

**Abstract:** Existence of heavy metals [Cadmium (Cd) & Chromium (Cr)] in *Zea mays* L. (Maize) influence production of crop plants by inhibiting their physiological processes, such as water balance, ions equilibrium, protein metabolism and photosynthesis. Cadmium and Chromium are present in soil as $CdCl_2$ and $CrCl_3$, respectively**.** The accumulation of Cd in stem causes reduction in the absorption, transport of water, minerals uptake, organic and inorganic solutes which lead towards the abnormal growth of leaves and unusual accumulation of reserved food materials in plant. The toxic effect of Cd causes leaf chlorosis and reduces the stomatal conductance by the production of reactive oxygen species (ROS) in leaves and other parts. While, buildup of Cr in the Maize plant, reduces quantity of photosynthetic organic compounds that results in decrease of stem diameter. Both Cd and Cr also decrease the plant height. Present study is to pin point the differences in heavy metals uptake in grain and their impact on different morphological parameters in Maize (*Zea mays*).

[Masood SA, Abbas HG, Rehman HU and Ali Q. **Effects Of Heavy Metals (Cd & Cr) On Physiological And Morphological Parameters In *Zea Mays* L.: An overview.** *Life Sci J* 2018;15(5):38-43]. ISSN: 1097-8135 (Print) / ISSN: 2372-613X (Online). http://www.lifesciencesite.com. 7. doi:10.7537/marslsj150518.07.

**Keywords:** Effect; Heavy Metal; Cd; Cr; Physiological; Morphological; Parameter; *Zea Mays* L

## Introduction

Maize is the third leading field crop in Pakistan after wheat and rice. Maize (Zea Mays) is also known as corn that was originated in central Mexico about 5,000 BC ago. Later, it was introduced to Europe in the sixteenth century. It spread to Africa and Asia afterwards. Now, Maize is among the most widely-grown cereal crops around the world in both tropical and temperate climates. Approximately eighty percent of world's maize is produced in the Americas (53%) and Asia (28%) and Europe (15%) (Ranum *et al*., 2014). *Zea mays* (maize/corn) is cultivated as both sweet corn for human eating and as a field crop for livestock and biofuels. The crop is rich in minerals, vitamins (especially vitamin B-6), dietary fiber and carbohydrates. It is a predominantly important source of nutrition and supplies a high energy density of 365 Kcal/100g (Nuss *et al*, 2010). Grain of maize is enriched with different nutritive components as compared to the other food grains. Wet milling of the maize grain produces an array of products, by products and value additions. Maize contributes 2.7 % to the value added in agriculture and 0.5 % to the GDP. Cultivated area of Maize is 1334 thousand hectares showing an increase of 12.0 %. Maize has shown a record production of 6.130 million tonnes in 2016-17 against 5271 million tonnes in 2015-16 (Govt. of Pakistan, 2015-16 & 2016-17). Yield is increased due to the contribution of hybrid seed with better inputs of crop management (Govt. of Pakistan 2012-13).

Maize is the short duration cereal crop as compared to the other cereals and it has ability to use solar energy more efficiently. In high altitude areas where the snow falling and chilling conditions limits the growing period of other cereals maize attains the great priority for its growing period Ali *et al*. 20160. Maize kernel contains 72 % starch, 10 % protein, 9.5% fiber, 4.80% oil, 3.0 % sugar and 1.7% ash content (Ali *et al*. 2013; Ali *et al*. 2014ab). Starch of maize is used to manufacture the cotton yarn and corrugated board in winding process. Due to low cholesterol in Corn oil it is useful for human diet and it is very good for heart patients. Developed countries are using maize to produce bio-fuel for vehicles. Corn alcohol is converted into bio-fuel which is an alternative source of energy instead of petrol.

Being a C4 plant maize plant is able to thrive well under the conditions of soil moisture deficit coupled with high oxidative stress. Crop productivity is limited due to biotic and abiotic stresses like temperature extremes, disease attack, salinity, water scarcity, water logging and heavy metal toxicity *etc*. (Boyer, 1970). Its plants have been reported to have ability to hyperaccumulate a number of heavy metals (Shanker *et al.,* 2005). Total 35 metals are present in nature out of which 23 are considered as heavy metals. Arsenic (As), Antimony (Sb), Cesium (Cs), Cadmium (Cd), Bismuth (Bi), Cobalt (Co), Cromium (Cr), Galilium (Ga), Copper (Cu), Manganese (Mn), Lead (Pb), Iron (Fe), Gold (Au), Nickel (Ni), Silver (Ag), Mercury (Hg), Platinum (Pt), Thallium (Ta), Tellurium (Te), Uranium (Ur), Tin (Sn), Vanadium (v) and Zinc (Zn) (Glanze *et al.,* 1996, Mahrukh *et al*., 2016). Presence of any of these heavy metals in the human diet causes serious health problems.

Cadmium (Cd) inhibits the seed germination in maize which disturbs the transpiration rate, mineral nutrition, induce the reactive oxygen species and most importantly reduce the biomass production. Its presence results in chlorosis, root and leaf necrosis which is a form of stunted growth. While, Chromium (Cr) restricts different metabolic processes and heavy metal toxicity inhibits the seed germination, proper accumulation of phytomass that causes stunted growth of seedlings and finally plant death *(*Zou *et al. 2009*).

**Heavy metals and maize/corn**

Agreger and Lindberg (1987) claimed that the heavy metals could compete with Ca for entry into plant cells and affect accumulation and distribution of Ca in plants. Corradi *et al*. (1999) conducted an experiment in which they grew *Zea mays* (maize) in a nutrient solution of $KCr_2O$. Results suggested that the level of Cromium (Cr) was approximately the same in the root tissues after 6 days of treatment. Though Cr altered the nuclear structure and also modified the cell cycle in $G_1$ phase.

Mantosiewicz (1993) reported that Lead (Pb) and Cadmium (Cd) uptake affected accumulation of Calcium (Ca) in organs of various plant species. Rodriguez *et al*. (1997) conducted an experiment in which they treated the maize plants with 0.0 (control), 0.01 and 0.05 mm Cd in the growing medium for 11 days. The plants exhibited severe toxicity symptoms. While, fresh weight, percentage of water content of root and shoot decreased alongside the Cd supply. High levels of cadmium were found in the cell-wall fraction (Fraction I) and in Fraction IV (soluble) of maize plants.

Majer *et al.* (2002) suggested that Lead (Pb) is potentially a noxious contaminating heavy metal which has no noticeable role in biological functioning of living organisms; causing the deteriorating effects on health, normal growth and development of plant as well as humans. Toppi *et al,* (2002) evaluated the tomato (*Lycopersicon esculentum*), Corn (*Zea mays*), and Cabbage (*Brassica oleracea*) for the effect of hexavalent chromium Cr (VI) on their different plant parts (seeds, roots and leaves). Prominently, chromium changed internal structures and metabolic activities of plants with ultimate plants death.

Sumitra and Parmar (2003) concluded that under normal condition hypocotyls of germinating seeds of *Phaseolus vulgaris* (common beans) seedlings grew properly but when seeds were germinated under $K_2Cr_2O_7$ (Cr source), it reduced the length of hypocotyl. So, it was established that hypocotyl elongation got reduced by Cr application. Liu *et al*. (2003) reported that lead (Pb) and chromium (Cr) are efficient and serious potential environmental pollutants. The toxicity produced by Cr in the soil and water caused different effects in plants. Cr (6) is usually more toxic and more movable as compared to Cr (3) that is less toxic and less moveable.

Pinto *et al*. (2004) while studying the impact of increasing level of toxicity of Cd on shoot, root growth and their proper functioning concluded that the Cd affects a wide range of physiological attributes in *Zea mays* L. (maize). Azevedo *et al,* (2005) studied the growth traits by exposing Sunflower (*Helianthus annuus* L.) to four levels of Cd (0, 5, 50, and 500 μM) for 21 days. As a result, the toxic effect of Cd reduced the Chlorophyll content, flowering, and the growth rate of both root and shoot. Panda *et al*. (2005) suggested that the chromium (Cr) induced the oxidative stress by causing lipid peroxidation, degradation of photosynthetic pigments, decline in growth and severe damages to cell membrane. Even, higher amount damaged the ultrastructure of chloroplast and affected the efficiency of antioxidant metabolism of the plants.

Shanker *et al*. (2005) evaluated the toxic effects of Cr on maize plants growth and development. They reported alterations in the germination process, roots, stems and leaves growth, which triggered the reduction in the total dry matter and final yield. Tsadilasa *et al*. (2005) concluded that the uptake of Cd in tobacco was greatly affected by soil PH and form of Nitrogen fertilizers. And, the unpredictability of Cd uptake by tobacco plant was credited to the difference in soil PH.

Yang *et al.* (2005) reported that the exchangeable fractions of Cd, Cu and Cr were all higher in the rhizosphere of maize than in bulk soil. Their results show that addition of Cd at low levels stimulates the ammonification and nitrification in soil, while inhibitory influences are shown at its higher concentrations. Nitrifying bacteria were proved to be the most sensitive to heavy metals presence, whilst the effect on denitrifying bacteria was very limited. Cd was the most effective inhibitor of ammonification and de-nitrification, while Cr (VI) had the strongest inhibitory influence on nitrification at concentration of 20 mg/kg. Cui *et al*. (2006) examined the physiological response to elemental Sulphur (S) and Cadmium (Cd) in maize (*Zea mays L.*) for 60 days in pot culture. The Cd was added in solution in 4 doses (0, 20, 50, 100 mg/kg) before planting the maize seeds. The results suggested that chlorophyll content, shoot biomass and crop growth got reduced due to Cd accumulation.

Hassett *et al*. (2006) concluded that the collective effects of Cadmium (Cd) and lead (Pb) strongly reduce the radicle development and elongation even at low concentration. Jamal *et al*., (2006) reported that the root/shoot length, root/shoot fresh & dry weight are significantly reduced by the toxic effects of Cr and Pb. Lahouti and Peterson (2006) used nutrient solution lebelled with Cr-III and Cr-II to conclude that 98 % of chromium accumulated by nine crop plants retained in the roots. Cauliflower plants accumulated maximum cromium while, Mung bean seedlings the least.

Pal *et al*. (2006) studied that the directly or indirectly accumulation of heavy metals e.g. Cd into the plants induced many physiological changes (free radicals formation, inhibition of photosynthesis, modifications in the ions & water metabolism, growth inhibition and changes in the enzyme activities). Chen *et al,* (2007); Zhong-Qiu *et al*., (2005) and Zhanga *et al*., (2002) evaluated the changes in growth and development of maize grains in the presence of Cd (Cadmium). The Cd caused the reduction in the growth, production, xylem & phloem functioning at 0.5, 2, & 8 M concentrations, after 15-day of exposure. Cd toxicity had the order: glume < rachis < grain < stem < awn. Maximum amount of Cd was found in maize grains (about 51.0%).

Fiala *et al*. (2007) studied the two maize cultivars named Blitz-160-MB and Premjia-190-MB. They exposed them to Ni and Cd consecutively for three days (individually and also in combination). They reported reduction in primary root length due to the individual effect of Cd and Ni ions. Cd & Ni conjointly reduced the cell viability. Further, growth and morphological parameters under study were said to be more sensitively at the stage of 5-6 days old seedlings in the Blitz-160-MB as in Premjia-190-MB. Maiti *et al.* (2012) evaluated two maize varieties Sartaj and Deccan to tell the effects of Cr on their physiological and biochemical characteristics. They reported the alterations in the protein oxidation and lipid peroxidation at 0, 50, 100, 200 and 300 μM doses. Further, it revealed that Sartaj was more sensitive to oxidative stress contrasting the variety Daccan.

Krantev *et al*. (2008) proposed that the toxic effects of Cd caused the reduction in the root and shoot fresh and dry weights of maize (*Zea mays* L.) plants. Juneja (2008) in his in-vitro studies, mixed radio tagged Cr with xylem sap (pure and carboxylic acid fraction) of maize crop at three stages of plant growth. Xylem sap transported both nutrient and non-nutrient ions after absorption by roots to the aerial plant parts. Speciation analysis by paper electrophoresis revealed that cationic Cr reacted with organic ligands, mobile & soluble complexes stored in leaves and other edible plant parts. While, in the detoxification mechanism, the harmful effects of toxic Cr were reduced and complexed. Liang *et al*. (2009) studied the toxic effects of Pb, Zn, and Cd complex in the soil and water, which caused the reduction of root/shoot fresh and dry weights and led towards the reduction in the production of maize plants.

Ghani (2010) investigated the toxic effect of heavy metals (Cr, Co, Pb, Mn and Cd) on the plant growth and seed yield of maize plants. After 80 days of germination, plants were harvested and then evaluated for nitrogen, protein and heavy metal content. As a result heavy metals caused significant decrease in growth and protein content. Cd was the most toxic metal followed by Co, Hg, Mn, Pb and Cr. Individual effects of these heavy metals were more intense as compared to their combined effects. Ali *et al*. (2011) evaluated two barley genotypes which were used to study the effects of Aluminium (Al) and Cromium (Cr) on plant growth, development, root growth enzyme (dehydrogenase), oxidative stress and antioxidative enzymes. They observed that plant growth was reduced after application of Both Al and Cr. Prominent physiological and chemical alterations were also noted in two species of Maize.

Data *et al.* (2011) conducted a laboratory experiment to determine the phytotoxic effect of chromium on seed germination and seedling growth of some wheat cultivars (*Triticum aestivum* L.). The plants were observed for germination percentage, root and shoot length, fresh weight and dry weight of seedling, sugar, protein and chlorophyll content in the leaves of both treated and control plants. Cr accumulation significantly affected the plants by inhibition of seed germination, root growth and other parameters. Gupta *et al*. (2011) determined the toxic effects of cadmium (Cd) on seed germination, root/shoot length and dry biomass of maize seedlings in comparative studies with control condition in a laboratory. Treatments used at 50, 100, 150 and 200 mg/L concentration significantly affected the seed germination and seedling growth as compared to the control. The concentration of Cd (50-200 mg/L) adversely reduced the seed germination, root/shoot length and dry biomass as compared to the control condition.

Kleckerova *et al*. (2011) studied the growth parameters and enzyme activity at various levels of Cadmium and Zinc ions in plant tissue. They found the reduction in growth parameters (root/shoot ratio and fresh/dry weight) in both cases. Sarvajeet *et al.* (2011) determined that Cadmium (Cd) is a non-essential and toxic metal, rapidly taken up by roots and accumulated in various plant tissues which hamper the crop growth and productivity worldwide. Plants employ various strategies to counteract the inhibitory effect of Cd, among which nutrient management is one of a possible way to overcome Cd toxicity. Solti *et al.* (2011) estimated that Fe is required to plants in a very small amount for uptake of minerals and ions. The shortage of Fe, due to storage of Cadmium (Cd), in plants affects xylem and ultimately uptake of water and minerals from the soil to aerial parts of plants. Cd caused deficiency of Fe that results in chlorosis and ultimately leads to the plant death.

Stingu *et al.* (2011) suggested that the maize plant responses, in terms of growth and metal uptake to different concentrations of cadmium ions (4, 20 µM). Plants were analyzed in a hydroponic culture for 2 weeks. For a 4 µM cadmium-contaminated environment, the maize plant presents the highest bioaccumulation level after 192 h, with a recovery degree of 52%, meanwhile, at a 20 µM concentration; the highest bioaccumulation was registered after 366 h, with a corresponding recovery of 10.56%. The translocation factor presented higher values for 20 µM induced contamination than for 4 µM, which means that increasing metal concentration in the medium increased the concentration of Cd ions in the upper parts of the plant. Anatomical sections of a maize plant (in a 4 and 20 µM cadmium-contaminated environment) were observed to evidence the changes in plant morphological structure.

Anwer *et al*. (2012) conducted an experiment to investigate the potential of citric acid for translocation and accumulation of Cd and to check its impact on maize plants growth. They grow the plants in small plastic bags and treated them with 300 mg $kg^{-1}$ of $CdCl_2$. The plants were harvested after 10 days then dried and weighted for root/shoot biomass to study the proficiency of maize plants to bioaccumulate metal. Results showed heavy metal accumulation more in roots than shoots and application of citric acid depressed the uptake at all concentration levels. That, proved maize plant to be an effective accumulator for Cd. Hussain *et al.* (2012) studied the toxicity of Cadmium and suggested that the Cd cause reduction in the plant growth and development. They exposed two maize cultivars Agati-2002 and Ev-1098 to $CdCl_2$ by growing the plants in the pots filled with sand in growth chambers where the day/night temperature was pre-set. After the 15 days of germination they exposed the seedlings to 0, 25, 50 and 75 µM Cd. High concentration (75 µM) of Cd reduced the growth attributes (chlorophyll content, relative water content of leaves) in Agati-2002 and induced the oxidative stress that resulted in plant growth reduction in both cultivars. Further, Agati-2002 was more susceptible to the cadmium (Cd) stress.

Hussein *et al*. (2013) determined the inhibitory effects of cadmium on various biochemical parameters and plant growth in maize. They used two maize lines MTM-1 and MTM-2 and applied the heavy metals at different time intervals. During this experiment 20 days old seedlings were exposed to 0, 3, 6, 9 and 12 mg $CdCl_2$ $kg^{-1}$ concentrations. The results revealed that the higher concentration of Cd reduced the plant growth in MTM-1 on the 5$^{th}$, 10$^{th}$ and 15$^{th}$ day after the treatment. But the plants did not show the inhibitory effects of Cd on biomass and leaf area in line MTM-2. Pooja *et al*. (2013) estimated that the potential toxicity of heavy metal (Cd) in plants cause reduction in plant productivity by disturbing physiological process like photosynthesis (mineral uptake, ion metabolisim and water relations).

Dikkaya *et al*. (2014) investigated the effects of Zinc (Zn), Cadmium (Cd) and their interaction on root/shoot dry weight ratio, root/shoot elongation and ascorbate peroxidase enzyme activity in maize (*Zea mays* L.). Results indicated that plants treated with high concentration of Zn had significant effect on root and shoot elongation. Cd treatment at 10 and 15 µM level shown significant decrease in root and shoot dry weights as compared to control. Qaisar *et al*. (2014) said that the chromium (Cr) is a heavy metal and has toxic effects on plants. Pretreatment of seeds with hydrogen peroxide ($H_2O_2$) can improve stress tolerance. They studied the response of maize (*Zea mays* L.) to $H_2O_2$ (80 µM) priming under chromium stress (100 and 200 µM) at seed germination and seedling stage. Experiments were conducted in Petri plates in completely randomized block design with three replications. A significant decrease was found in photosynthetic pigments, germination percentage and tolerance index of seedlings under chromium stress. Sugar and proline content were increased under chromium treatment and tolerance was improved by the application of hydrogen peroxide (80 µM).

**Conclusion**

It is concluded that the toxic effects of heavy metals; Cr and Cd affects the vital morphological and physiological parameters. They cause the reduction in normal growth and development pattern of maize plant. Even, severe heavy metal toxicity result in oxidative stress, nuclear structural changes, disturbs cell cycle, ions imbalance, affects cell viability and can cause death of plants. Hence, serious yield losses occur.

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5/19/2018