

EFFECT OF CADMIUM DOSES ON SEED GERMINATION AND MORPHOLOGY PARAMETERS OF WHEAT (*TRITICUM AESTIVUM* L.)

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INTRODUCTION

Pollution from heavy metals is a global problem that is very dangerous to the environment. The term "heavy metals" refers to any metallic element that has a relatively high density and is toxic at low concentration (Lenntech, 2004). Among all other heavy metals, cadmium is receiving more and more attention because it is one of the most toxic heavy metal. Cadmium (Cd) is a toxic metal that is naturally present in trace quantities in almost all soils. The main source of Cadmium in the environment by power stations, heating systems, metal industries, Ni-Cd batteries, and phosphate fertilizers (Toppi and Gabbrielli, 1999) as well as from geo-chemical weathering of rocks. Cd is very harmful for biological activity in soil, it also causes morphological, anatomical, and physiological changes in plants including growth inhibition, water imbalance, and reduction in seed germination (Benavides *et al.*, 2005; Mishra *et al.*, 2006). The regulatory limit of cadmium (Cd) in agricultural soil is 100 mg/kg soil (Salt *et al.*, 1995). The presences of Cd in the soil is taken up by roots and then transported across plant tissues, and finally accumulated in roots, shoots, fruits and grain (Qian *et al.*, 2009). Wheat is one of the most important crops all over the world and wheat is the second most important food grain of India and it is the staple food for millions of Indians. Accumulation of cadmium in wheat is higher than other common cereals such as oats and barley (Jansson, 2002). The concentration of cadmium at 0.2 mM is able to cause 50% loss in whole chain as well as photosystem II catalyzed electron transfer activities in wheat thylakoids. Compared to photosystem II, the activities catalyzed by photosystem I are less sensitive to cadmium stress (Hasan and Murthy 2008). Recently the (Codex Alimentarius Commission, 2005) of FAO/WHO has set a level for Cd in wheat grain of 0.2 mg kg⁻¹. The aim of this study is to evaluate the effect of cadmium toxicity and different doses of Cd concentration which is more effective to the germination and also shows negative effect on different morphological parameters of wheat crops.

MATERIALS AND METHODS

Effect of cadmium doses on seed germination and morphology parameters of wheat (*Triticum aestivum* L.) the experiment was performed in Ranjan Plant Physiology and Biochemistry Laboratory, Botany Department, University of Allahabad and Department of Environmental Science, School Of Forestry And Environment, SHIATS, Allahabad.

ABSTRACT

Present study carried out to evaluate the Cd effect on growth parameters in wheat (*Triticum aestivum* L.). Healthy seeds were sown in sterilized sand filled with plastic glass and kept in darkness for 2 days, seedling placed in growth chamber for 15 days at 24 ± 2°C with highest intensity 100 μmol photon m⁻²s⁻¹. Plant uprooted with jet of water and uniform size seedling were placed in aqueous growth medium. Uniform seedling of wheat were germinated in Hoagland's solution containing different concentrations of Cd (200, 500, 1000, 2000, 5000, 10,000 μmol). The result shows that the maximum germination of seedling was found in control which was (98%) and minimum was recorded in treatment 10,000 μmol (36%) due to higher concentration present in stock solution. As well as higher concentration of Cd (10,000 μmol) decreases plant height 57.44%, shoot length 62%, root length 54.97%, fresh weight 59.01% and dry mass 40% as compare to control. Shoot length of wheat is highly affected by higher concentration of Cd as compare to other parameters of wheat seedling. This experiment revealed that this variety of wheat show higher accumulation in shoot length and also Cd produced a significant reduction in seed germination, plant height, shoot and root length, fresh and dry mass of wheat crop.

KEY WORDS

Heavy metal cadmium
Wheat (*Triticum aestivum* L.)
Morphology
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Seed material

Seeds of wheat cultivars (Lok 1 variety) were used by the cereal section. The healthy and robust seeds of each cultivar were surface sterilized with sodium hypochlorite (4%) for ten minutes followed by five washings with double distilled water.

Cadmium stock solution

Cadmium chloride ($\text{CdCl}_2 \cdot \text{H}_2\text{O}$) was used to prepare the stock solution (in 20,000 μmol) by dissolving required mass of (400mg) CdCl_2 into sterilized Hoagland's nutrient medium, (Hoagland's solution was given by Hoagland and Arnon, 1950. This is the one of the most popular solution composition for growing plants). From this stock solution finally desired different doses of cadmium are (200, 500, 1000, 2000, 5000, 10, 000 μmol) were used in the experiment along with control (without Cd).

Experimental Setup

Sand was sieved through 2 mm sieve before filled in thermophore plates which has dimensions 43 x 43 x 23 length, width and depth, respectively. Each thermophore plate contained 200g sand and Cd levels were developed in sand by adding 50 ml of each concentration before sowing. Seeds of each cultivar were dipped in distilled water for 24 hours at 28°C in incubator. Seven imbibed seeds of each cultivar were placed in the sand at uniform depth. These plates were placed in growth room at 25°C under 14 hrs photoperiod.

Determination of Seed germination

Seed germination rate was determined by counting number of germinated seeds after seven days of treatment. Seed germination percent was calculated using the formula:-

$$\text{Seed germination percent} = \frac{\text{Number of seeds germination}}{\text{Total number of seeds sown}} \times 100$$

Plants growth

After last treatment of CdCl_2 on 12th day, seedling was harvested and various growth parameters were determined. Length and height were measured with the help of a meter rod. Fresh and dry weight was measured with the help of weighing machine.

Statistical analysis

All statistical analysis was performed on observed data during the experiments and analysis with the help of statistical software (SPSS version 10).

Table 1: Effects of Cd on seed germination (%) of (*Triticum aestivum*).

Treatment	Germination percentage (%) of seedling		
	No. of seeds placed	Germinated seeds	Germination(%)
Control	50	49	98
200 Cd μmol	50	48	96
500 Cd μmol	50	47	94
1000 Cd μmol	50	43	85
2000 Cd μmol	50	37	75
5000 Cd μmol	50	30	61
10,000 Cd μmol	50	18	36

Data are mean of the three replication \pm standard error of the mean.

RESULTS AND DISCUSSION

Effect of cadmium on Seed germination

The effect of cadmium treatment on seed germination of wheat seedlings significantly decreased with increasing cadmium concentration shown in (Table 1). The rate of reduction were significantly recorded with 36% decreases, due to higher concentration (10000 μmol) of Cd presences in stock solution as compare to control which was recorded (98%). Cadmium reduce seed germination of wheat seedling at higher concentration. Similarly work also reported by (Asgharipour *et al.*, 2011), Cadmium exhibited toxic impact on germination and seedling growth. Negative effect of cadmium on seed germination and seedling growth of four wheat (*Triticum aestivum* L.) cultivars (Ahmed *et al.*, 2012).

Effect of cadmium on Plant height

The effect of cadmium treatment on plant height show the adverse effects which was significantly decreases (57%) at higher Cd concentration presences in stock solution as compare to control. The result shown in (Table 2) which was recorded plant height (27cm) at higher concentration and 47cm in control. The standard error was ranges between ± 0.64 to ± 2.82 . Higher concentration of Cd show the negative effect on plant height of wheat crops. Cadmium is unnecessary for plant height and can be easily accumulated by plant (Miao and Wang, 2006).

Effect of cadmium on shoot length

The effect of cadmium treatment on shoot length show the more significant reduction as compare to another part of the wheat seedling the rate of reduction were recorded 62% decreases due to higher concentration (10000 μmol) of Cd presences in stock solution as compare to control. The result shown in (Table 3) which was recorded 17 cm at higher concentration and 27.4 cm in control. The standard error was ranges between ± 0.39 to ± 1.19 . Higher concentration (10,000 μmol) of Cd adversely affects the shoot length. The concentration of Cd in shoot was always higher than that in roots. The accumulation of cadmium concentration in shoot was found much high as compare to root which was recorded highest percent decrease 54.97% in root and 62% in shoot at higher cadmium treatments. The increase in Cd concentrations in plants with Cd treatment was previously reported in many studies (Koleli *et al.*, 2004; Ouzounýdou *et al.*, 1997; Yildiz, 2005). Reduced shoots length and weight in winter wheat (*Triticum aestivum* L.) and canola (*Brassica napus* L.) plants subjected to cadmium metal toxicity have been reported (Khan *et al.*, 2007 and Lamhamdi *et al.*, 2013). Increasing the Cd-concentrations led to transition of leaf chlorosis into yellowing and necrosis of leaf tips (Foy *et al.*, 1978). (Prasad, 1995) has suggested that Cd is easily absorbed by the root system of plants and translocated by xylem to the aerial portion

Effect of cadmium on Root length

The effect of cadmium on root length of wheat seedling was recorded maximum in control 19.1cm and minimum was recorded 10.5cm decreased with the increasing of Cd concentration 10,000 μmol . The result shown in (Table.4) which was significantly decrease 54.97% on root length of wheat as compare to control. The standard error was ranges

Table 2: Effect of cadmium on plant height (*Triticum aestivum*) .

Treatment	Plant height (cm)
Control	47 ± 2.31
200 Cd μ mol	39 ± 2.71
500 Cd μ mol	38 ± 2.82
1000 Cd μ mol	36 ± 1.35
2000 Cd μ mol	34 ± 1.47
5000 Cd μ mol	32 ± 0.64
10,000 Cd μ mol	27 ± 0.99

Data are mean of the three replication \pm standard error of the mean.

Table 3: Effect of cadmium on shoot length (*Triticum aestivum*).

Treatment	Shoot length (cm)
Control	27.47 \pm 0.75
200 Cd μ mol	24.63 \pm 0.84
500 Cd μ mol	22.10 \pm 1.19
1000 Cd μ mol	20.86 \pm 1.08
2000 Cd μ mol	19.13 \pm 0.72
5000 Cd μ mol	18.66 \pm 0.39
10,000 Cd μ mol	17.00 \pm 0.57

Data are mean of the three replication \pm standard error of the mean.

Table 4: Effects of cadmium on root length of (*Triticum aestivum*).

Treatment	Root length (cm)
Control	19.17 \pm 0.53
200 Cd μ mol	14.96 \pm 0.49
500 Cd μ mol	13.92 \pm 0.88
1000 Cd μ mol	13.79 \pm 0.30
2000 Cd μ mol	12.69 \pm 0.28
5000 Cd μ mol	11.43 \pm 0.34
10,000 Cd μ mol	10.53 \pm 0.27

Data are mean of the three replication \pm standard error of the mean

Table 5: Effect of cadmium on fresh weight of (*Triticum aestivum*).

Treatment	Fresh wt mg plant ⁻¹
Control	122 \pm 3.21
200 Cd μ mol	87 \pm 2.71
500 Cd μ mol	84 \pm 2.82
1000 Cd μ mol	82 \pm 1.35
2000 Cd μ mol	81 \pm 1.47
5000 Cd μ mol	79 \pm 0.64
10,000 Cd μ mol	72 \pm 0.99

Data are mean of the three replication \pm standard error of the mean.

Table 6: Effect of cadmium on dry mass of (*Triticum aestivum*)

Treatment	Dry mass mg plant ⁻¹
Control	60 \pm 1.05
200 Cd μ mol	49 \pm 0.99
500 Cd μ mol	41 \pm 1.15
1000 Cd μ mol	38 \pm 0.72
2000 Cd μ mol	34 \pm 0.68
5000 Cd μ mol	29 \pm 0.43
10,000 Cd μ mol	24 \pm 0.49

Data are mean of the three replication \pm standard error of the mean

between \pm 0.27 to \pm 0.88. Similarly result was reported by (Oncel *et al.*, 2000) Cr⁺⁶, Cu⁺² and Ni⁺² showed a concentration dependent inhibition of root growth at 20 and 40 ppm doses and also obtained similar effects using cadmium in wheat seedlings. High levels of Cd consequently induce growth abnormalities including short, thick roots (Breckle, 1991). This was observed by other author also that Cd responsible for abnormalities as legends, bridges, stickiness, precocious separation and fragments were most common

(Siddiqui *et al.*, 2009). Cd⁺² reduced the root size by 5.0% as compared to the control root elongation. The Cd accumulated in the root remains in this organ, and only 2% is transported to leaves in higher plants (Xue *et al.*, 2013).

Effect of cadmium on Fresh weight

The effect of cadmium treatment on fresh weight show the negative effects which was significantly decreases (59.01%) at higher conc. of Cd presences in stock solution as compare to control. The result shown in (Table.5) which was recorded 72cm at higher concentration and 122 cm in control. The standard error was ranges between \pm 1.36 to \pm 3.20. Cadmium had adverse effects on fresh weight of wheat seedling. Similarly work also reported by (Hemalatha *et al.*, 2010) with increase in Al and Cd concentration, considerable reduction in seed germination rate, root and shoot lengths of seedlings, their fresh weights and chlorophyll content was noticed among the four cultivars, of finger millets.

Effect of cadmium on Dry mass

The effect of cadmium treatment on dry mass of wheat seedling show the adverse effects which was significantly decreases (40%) at higher conc. (10,000 μ mol) of Cd presences in stock solution as compare to control. The result shown in (Table. 6) which was recorded dry mass decrease 24 cm at higher concentration and 60 cm in control. The standard error was ranges between \pm 0.43 to \pm 1.15. Dry mass of wheat seedlings was decreased under higher concentration of Cd. DM decreased under stress and more prominently in metal stress. Decrease in DM corresponded to level of chlorophyll content in respective treatments (Dube *et al.*, 2002). Such an observation parallels earlier reports the negative effect of Cd on plant growth was accompanied by an increase in dry to fresh mass (DM/FM) ratio in all organs (Moya *et al.*, 1993). That seedling weight accumulation is severely affected in plants growing in metal-contaminated soils (Verkleij and Prast, 1989; Ouzounidou *et al.*, 1995).

In this study, it was observed that the heavy metal accumulation depend on different concentration of cadmium. Cadmium is one of the most toxic metal which show the negative effect if it is present in higher concentration. From the present research it concludes that the higher concentration of cadmium chloride was adversely affected the seed germination and morphological parameters of wheat seedling. The cadmium concentration in shoot length was much higher as compare to other morphology parameters of wheat crops. Based on results Lok 1 variety of wheat was more sensitive to cadmium concentration in shoot length and it should be considered that higher concentration of cadmium decline the seedling percentage as well as plant growth of wheat crops.

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