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PHYSIOLOGICAL ANSWERS to METAL TRIGGERED EFFECTS in CROPS

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ABSTRACT

Environmental pollution is one of the most important concerns for today's people. The increasing population growth in the world, together with the industrial revolution and its side effects, brings along the risk of reaching healthy food for every newborn and existing population. Contamination and/or accumulation of heavy metals, known as the source of oxidative stress, in foodstuffs and the environment directly affects human health. Since plants are often the first recipients of heavy metals from different environments (such as air, water and soil), it is not surprising that this results in scary scenarios for all living organisms. Therefore, to determine and dispose of the possible downsides in human diet which relates to heavy metals, we examined whether physiological parameters are reliable indicators of oxidative stress in different crop plants. Either with single or combined CdCl2 and PbCl2 treatments, germination percentage, root and shoot lengths and water contents were measured in the roots and shoots of *Triticum aestivum* cv. Gerek and *Hordeum vulgare* cv. Çıldır varieties in this study. According to obtained results, it is concluded that heavy metal treatments trigger the oxidative stress in plants, as all parameters measured are dramatically decreased.

Keywords: Barley, Heavy metals, Germination, Water contents, Wheat

1. INTRODUCTION

Although they occur naturally in the Earth's crust, heavy metals (HMs) have always been a source of problems for the environment due to their tendency to accumulate. Cd and Pb, which are classified as highly toxic metals, form reactive oxygen species (ROS) in cells, which like all other HMs cause oxidative stress. Therefore, increasing HM levels affect the routine metabolic processes and cause some morphological and physiological changes [1]. However, while morphological changes can only be visible after some period of time, physiological changes can be trackable through some parameters.

It is highly important to take advantageous of measurable physiological parameters. Because, oxidative stress caused by HMs affects crop yields. It is known that, seed germination is prevented at higher HM exposures. Again, especially the root growth is easily affected with HMs and loosing water content is a definite consequence in different organs for plants grown in environments with toxic levels of HMs. Therefore, the germination rates, root and shoot growths and water contents are some of the accepted indices as measurable physiological parameters [2, 3]. The importance of these parameters increases even more, because HMs taken by plants are transported to humans through the food chain and cause various health problems.

With this study, we aimed to investigate the relationship between oxidative stress and physiological responses of plants through the application of Cd and Pb toxicity in cultivated plants most commonly used as nutrients in Central Anatolia.

2. MATERIALS AND METHODS

2.1. Seed Germination

For germination, 20 seeds (*Hordeum vulgare* L. cv. Çıldır and *Triticum aestivum* L. cv. Gerek) were treated with CdCl₂ and PbCl₂ solutions of 3 ml single (0, 50, 100 µM) and 3 ml combined (0, 50 + 100,

 $50 + 100 \mu M$) concentrations on Whatman filter paper in petri dishes. Control samples were given the same amount of dH₂O. After 3 days acclimatization period, by applying photoperiod [16h for 7 days, 22° C (\pm 1°C)] in the growth chamber, it was observed that the plants germinated after 10 days [4]. The following formula was used to calculate the germination percentage;

Germination percentage = Number of germinated seeds / Total number of seeds x 100 [5].

2.2. Measurement of Root and Shoot Lengths

The roots and shoots of the plants were separated and measured with a millimeter ruler.

2.3. Determination of Water Contents

Following the wet weight determination, roots and shoots were taken separately into the incubator for 48h at 70°C. Then, they were taken to the desiccator and allowed to reach room temperature. Finally, dry weight measurements were carried out with analytical balances as in wet weight [4]. The formula below is used for the water content calculation;

Water contents (%) = Wet weight - Dry weight / Wet weight x 100 [6].

3. RESULTS

3.1. Germinations of Crop Varieties

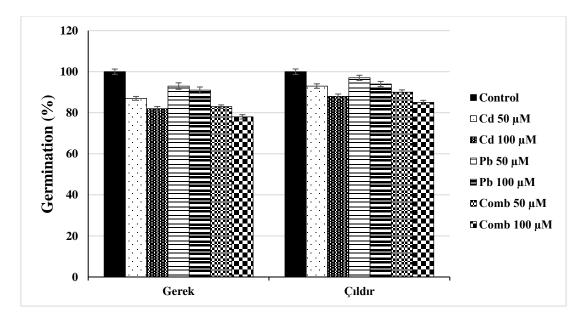


Figure 1. Effects of Cd and Pb treatments on seed germination. Absolute values can be seen in the text. Changes in values are shown as percentage of their controls.

The absolute values of seed germinations for controls of Gerek and Çıldır samples were calculated as 16 and 19 out of 20 seeds, respectively. Figure 1 shows the effects of different metal treatments on germination percentages of Gerek and Çıldır varieties. It was observed that different single and combined Pb and Cd applications decreased the germination percentage in a dose-dependent manner in both varieties. However, single Pb applications were found to be less inhibitive by comparing to their corresponding Cd applications. The germination percentage of the Çıldır variety is more successful than

the Gerek variety, while the highest decrease in germination percentage was belong to Gerek variety with the application of $100~\mu M$ CdCl₂+PbCl₂.

3.2. Root and Shoot Lengths of Crop Varieties

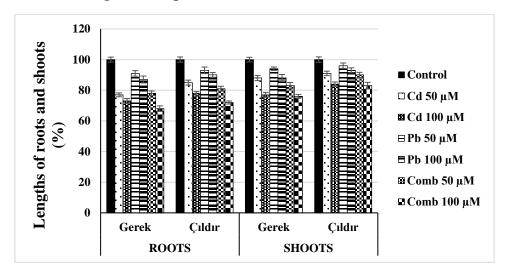


Figure 2. Effects of Cd and Pb treatments on root and shoot lengths. Absolute values can be seen in the text. Changes in values are shown as percentage of their controls.

The absolute values of root and shoot lengths of the control samples of Gerek and Çıldır varieties were measured as 12.60 and 13.80 cm for shoots and 9.50 and 11.80 cm for roots, respectively. Figure 2 shows the effects of different metal applications on the root and shoot lengths of Gerek and Çıldır varieties. According to our results, while increasing HM applications show dramatic dose depended effect on both shoot and root lengths, similar to trend in germination percentages, Cd effect was more significant than Pb. On the other hand, plant shoots were less affected by increasing HM doses by comparing to roots and the highest decrease in growth was belong to roots of Gerek variety with the application of 100 μ M CdCl₂+PbCl₂.

3.3. Water Contents of Crop Varieties

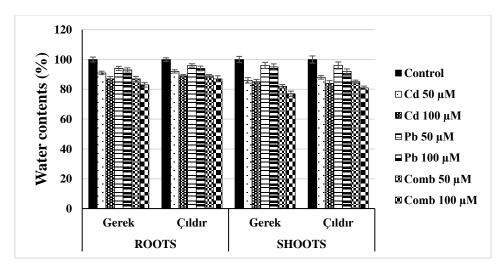


Figure 3. Effects of Cd and Pb treatments on water contents. Absolute values can be seen in the text. Changes in values are shown as percentage of their controls.

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The absolute values of water contents for controls of Gerek and Çıldır samples were calculated as 58.00% and 66.30% in shoots and 70.20 and 77.00% in roots, respectively. Figure 3 shows the effects of different metal applications on the water content of the Gerek and Çıldır varieties. Results show that, water contents decreased in a dose-dependent manner due to single or combined HM applications in both plants and single Pb applications were less inhibitory than Cd applications. During the course of treatments, shoots were found to be more sensitive to increasing HMs in comparison to roots and the highest decrease in water content was belong to shoots of Gerek variety with the application of $100~\mu\text{M}$ CdCl₂+PbCl₂.

4. DISCUSSION

4.1. Germinations of Crop Varieties

Germination percentages are considered as an indicator of the adaptation of plants to their environments, due to effects of HMs on plant germination and growth [7]. In the literature, there are some reports indicating the inhibition of seed germination with increasing HMs (especially Pb and Cd) doses in different plant organisms [8, 9], similar to ours.

On the other hand, in some studies supporting current results, it has been reported that Cd causes a greater reduction in germination compared to other HMs and Pb [10]. In our study, it was observed that the germination percentage of Çıldır plant was higher by comparing to Gerek variety. This can be interpreted as suppressions on water intake and other mechanisms relating to nutrition causes an interruption of germination [11] and these processes were seem to be more affected by HM treatments in Gerek variety.

4.2. Root and Shoot Lengths of Crop Varieties

Since root and shoot lengths are the expression of growth, their growth potential was accepted as an indicator [12]. Several reports in literature have shown the inhibitory effects of Pb and Cd on root [8] and shoot [13] growths, as we observed in the current study with increasing doses of Pb and Cd treatments.

Similar to situation discussed for germination above, it has been shown that Cd had the highest effect on both shoot and root lengths among other tested HMs, including Pb [10] and this is also support our results. In the present study, the suppressor effects of Pb and Cd on roots and shoots were more pronounced in Gerek variety. The reason for this is that with the increase of HM doses, the endodermis barrier disappears and the metal flow reaches the vascular tissues.

4.3. Water Contents of Crop Varieties

Among other causes, the main accepted reason for the decreased water content is the reduction in the absorption area due to the reduction in the number of hairy roots under the influence of HMs [14]. Therefore, water is a limiting factor, especially where high concentrations of HMs available.

As reported by previous researchers, the water content of plants treated with Cd and Pb decreased [15, 16] and they are in agreement with our results. On the other hand, we observed that the combined treatments of Pb and Cd more limiting in terms of roots and shoots water contents by comparing to their individual effects. This is also declared after examining the combined effects of NaCl and Cd in wheat [17]. The current study show that, water content of Gerek variety found to be more sensitive to HM applications in comparison to Çıldır variety.

5. CONCLUSIONS

As a conclusion we can declare that, the tested physiological parameters can be used as biomarkers for the early determination of oxidative stress, because decreases were observed in all parameters and this is an indication that plants are exposed to oxidative stress under HM treatments. Another inference that can be made according to the results of this study is, Çıldır variety can be used as an agent in rhizofiltration processes for cleaning farming areas, as it is more resistant to HM stress by comparing to Gerek variety. However, for the same reason, precaution should be taken when using it as animal or human food.

CONFLICT OF INTEREST

The author stated that there are no conflicts of interest regarding the publication of this article.

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