THE INFLUENCE OF SOME HEAVY METALS ON SEED GERMINATION AND SEEDLING GROWTH AT *RAPHANUS SATIVUS* L.

INFLUENȚA UNOR METALE GRELE ASUPRA GERMINAȚIEI SEMINȚELOR ŞI CREŞTERII PLANTULELOR LA *RAPHANUS SATIVUS* L.

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Abstract.*The paper presents the results of a study concerning the influence of different concentrations of Pb and Cd on some physiological indicators in the dynamics of the process of seed germination and in the incipient phases of seedling growth.The results underline the specific variations of the analysed indicators (water and dry substance content, content of total mineral elements, content of assimilating pigments, cellular liquid concentration, respiration intensity), depending on the nature of metals and their concentrations used for the seed treatments.Regardless to the concentration, Pb, and Cd determine the decrease of respiration intensity,- indicator for metabolic activities - , and water content as well as the increase of the content of assimilating pigments (exception for Cd in concentration of 10ppm).*

Rezumat. *Lucrarea prezintă rezultatele unui studiu referitor la influența concentrațiilor diferite de Pb şi Cd asupra unor indicatori fiziologici în dinamica procesului de germinație a semințelor şi în primele faze de creştere a plantulelor. Rezultatele evidențiază variații specifice ale indicatorilor analizați (conținut de apă şi substanță uscată, conținut în elemente minerale totale, conținut în pigmenți asimilatori, concentrația sucului vacuolar, intensitate respiratorie) în funcție de natura metalelor şi de concentrațiile acestora utilizate pentru tratarea semințelor. Indiferent de concentrație, Pb şi Cd determină scăderea intensității respirației, indicator pentru evidențierea activității metabolice şi a conținutului de apă precum şi creşterea conținutului de pigmenți asimilatori (excepție face Cd în concentrație de 10ppm).*

The influence of heavy metals on plants is presented in the specialty literature by several authors (1, 2, 4, 6, 7, 8, 10, 12). The paper continues the research regarding the effect of the treatment with heavy metals carried out in the previous years at species with phyto-remediation potential from the *Fabaceae* and *Poaceae* families (9, 11).The paper presents the results of a study concerning the influence of the different concentrations of Pb and Cd on some physiological indicators in the dynamics of the process of seed germination and in the incipient phases of seedling growth in *Raphanus sativus.*

MATERIAL AND METHODS

As vegetal material, we used seeds of *Raphanus sativus*. We had five experimental variants: 1 - control – M (with distilled water) and 4 variants of treatment with heavy metals. They were used in form of acetate in the following concentrations: Pb (50ppm – Pb1, 200ppm – Pb2); Cd (1ppm – Cd1 ;10 ppm – Cd2). The duration of the treatment was 3 hours. After the treatment, the seeds were put for germination in Petri plates, on filter paper, in laboratory conditions.

We analysed the following physiological indicators: at intervals of 1 - 10 days: the content of water and dry substance (gravimetrical method) and intensity of the respiration (Warburg method); at intervals of 4 - 10 days: cellular liquid concentration (refractometrical method) and the content of total mineral elements (dry calcination at 450^0 C method). Moreover, at 1o days after the germination we determined the content of assimilating pigments (spectro-photometrical method).

RESULTS AND DISCUSSIONS

The *Brassicaceae* family comprises numerous representatives mentioned in the specialty literature (3,5,8) as plants hyperaccumulator of heavy metals. Among the representatives of this family we chose for study the species *Raphanus sativus.* This species has the facile germination. Marchiol and collab. 2004 find that the *Raphanus sativus* cultivated on the polluted soils with Cd, Cu, Ni and Zn presents the foliar symptoms of toxicity (chlorosis), and the decrease of biomass with 52% comparing to the witness.

The results obtained are presented in figure 1-5.

The water and dry matter content (fig.1). At every investigated experimental variant, the water content has an ascendant evolution during the analyzed period. This fact is according to the cytophysiological and biochemical changes specific to the germination and seedling growth process.

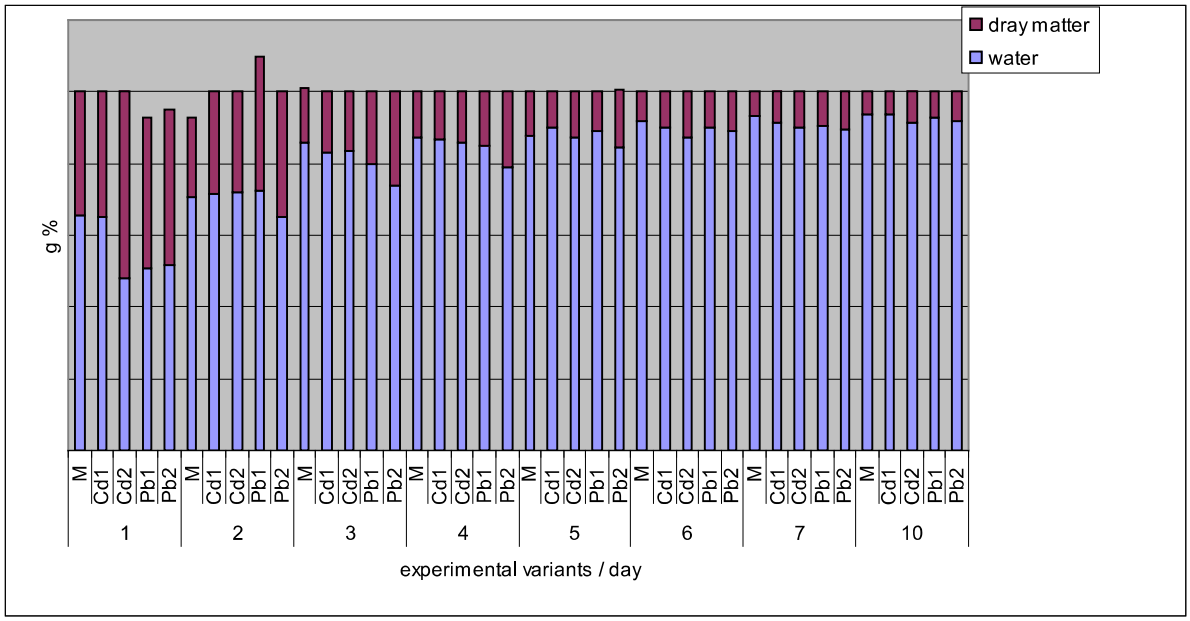

Fig. 1 - The dynamics of the content of water and dry matter

After the first day from the experiment's installing, we ascertain that the water content of the variants treated with Cd 10 ppm and Pb 50 and 200 ppm has the lowest values. This emphasizes the fact that Pb and Cd in the used concentrations slow down the water uptake by the seeds. Within a period of 2-10 days, we ascertain small value differences between the treatment variants.

By comparison with the witness, at the treatment variants, the water content values are generally lower but close to it.

At every experimental variant, the dry matter content decreases from the first day to the 10th day. This reality is due to the fact that the spare substances from the endosperm and cotyledons are hydrolysate, mobilized and used as respiratory sublayer and for the synthesis of new substances necessary for the embryo's nutrition and growth.

The cellular liquid concentration (fig. 2)

At the witness as well as at the treatment variants, the cellular liquid concentration values are smaller on the 4th day and lower on the 10th day. The osmotically active substances from the endosperm and cotyledons' cells also take part in the attainment of the cellular liquid concentration at the first determination. During the germination process and in the incipient stage of the seedling growth, the soluble glucide content decreases, the tissue hydration degree increases and the cellular liquid is diluted.

By comparison with the witness, at the treatment variants the cellular liquid concentration values are inferior to it (1.5-1.65 times smaller than it) on the 4th day and superior (except Cd1) but close to it on the 10th day. We can ascertain an inverted correlation between the cellular liquid concentration and the water content.

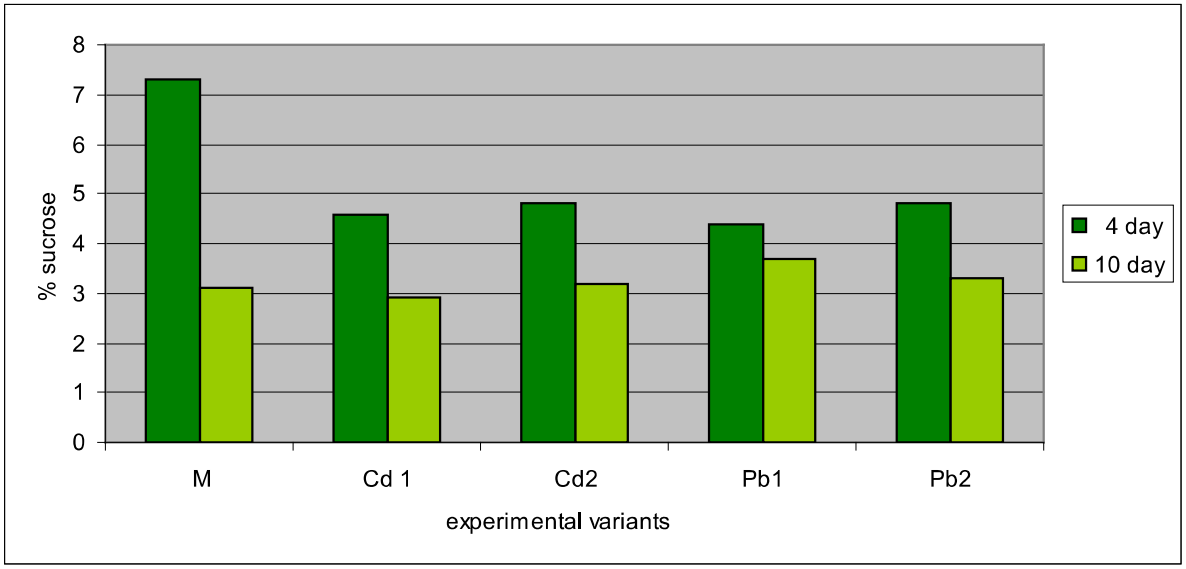

Fig. 2 The dynamics of the concentration of cellular liquid

The total mineral elements contents (fig.3) has values that range between: 4.56 g % and 5.99 g % for the witness and 4.30 g % and 6.69 g % for the treatment variants.

At every experimental variant, the values of the total mineral elements content are lower on the 4th day by comparison with the 10th day. We can ascertain small value variations between the witness and the treatment variants.

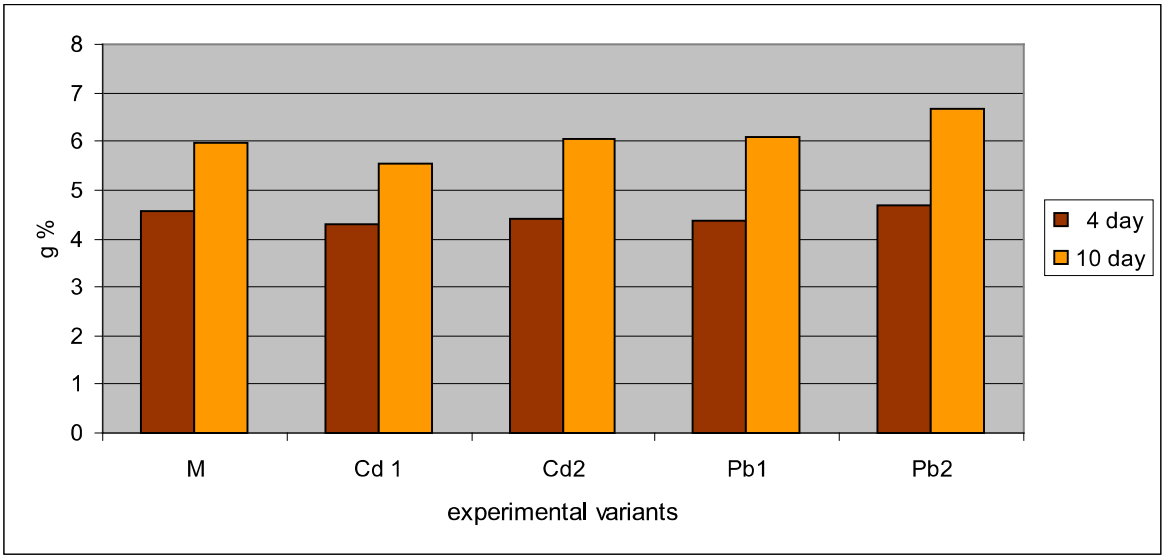

Fig. 3 The dynamics of the content of total mineral elements

From among the **forms of** analyzed **assimilated pigments** there prevails in the quantitative aspect in all experimental variants, chlorophyll a, followed in decreasing order by chlorophyll b and by carotenoidic pigments. An aspect to be noticed is the fact that the treatments with Cd 10 ppm determined the decrease of the content of total assimilating pigments, in comparison with the witness, while the treatments with Cd 1ppm and Pb (50ppm and 200ppm) have opposite effects (fig. 4).

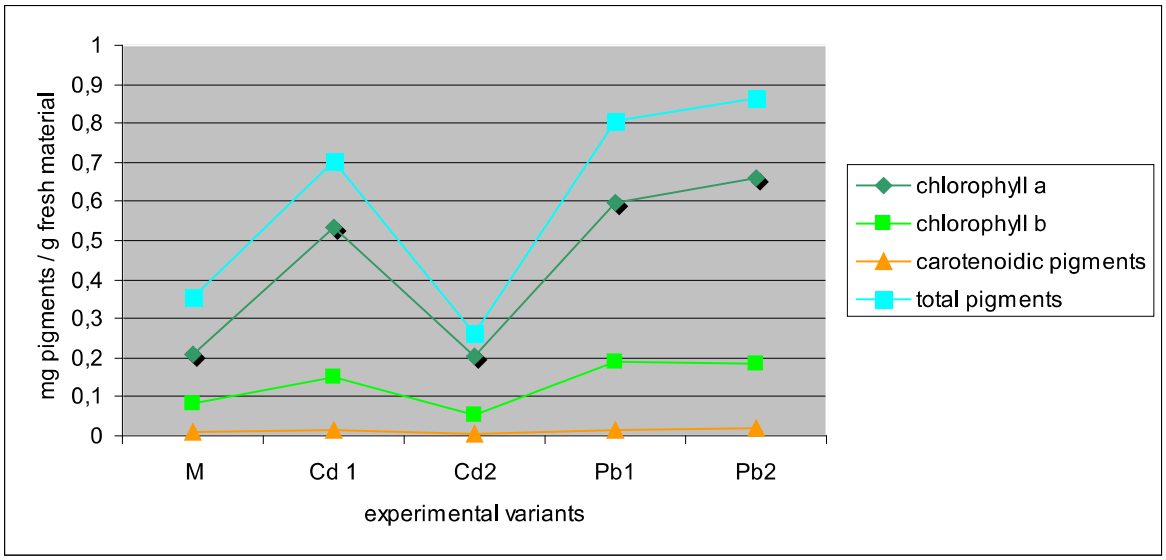

Fig. 4 The content of the assimilating pigments

We can ascertain an increase of the a chlorophyll content - which is more resistant to oxidative stress conditions – at all the experimental variants, more obvious at the Cd2, perhaps as a defence mechanism.Our results for the treatments with Pb and Cd 10 ppm confirm some of the literature data in the field

regarding the positive or negative effects of the two heavy metals used. According to Hernandez and collab. (2000) Pb in a concentration of 200 ppm caused the growth of the chlorophyllian pigment content.

Authors cited by Linger and collab. (2005), find that the Cd interferes with chlorophyll synthesis, assembly of pigment protein complexes and thylakoids, the electron transport chain, Calvin cycle enzymes, sugar transport and consumption, chloroplast replication and oxidative stress.

The **respiration process** generally has a dynamics which is similar at all the experimental variants and its intensity is higher in the first period of time analyzed and diminishes in the second. By comparison with the witness, at the treatment variants, the respiration intensity has (with some exceptions) lower values in the period of time analyzed. These results confirm the ones presented in the specialty literature consulted (2,6,7).Cd is highly reactive, inactivates different enzymatic processes and modifies the cellular redox balance (Jonak şi collab., 2004; Schutzendubel and Polle, 2002) and plants respiration (authors cited by Linger and collab., 2005).

At the Cd 1 variant, the respiration intensity constantly remains at values close to the witness; this emphasizes the fact that Cd in a concentration of 1 ppm does not affect this process.

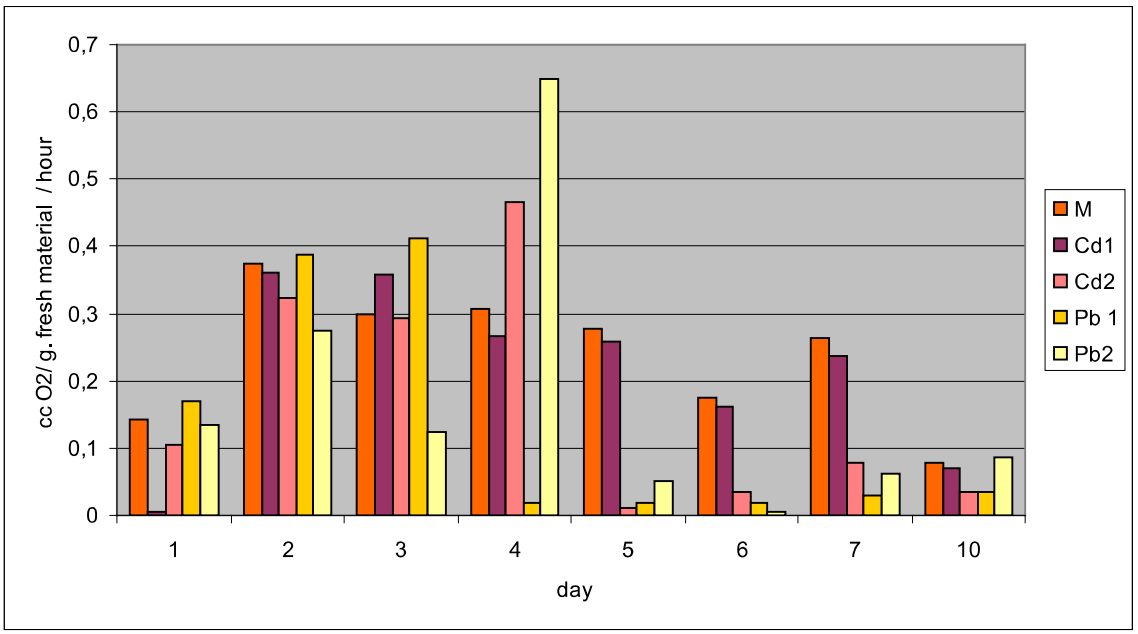

Fig. 5. The dynamics of the intensity of respiration

CONCLUSIONS

We underlined specific variations of the indicators analysed according to the nature of the metals and their concentrations used for the treatment.

The concentration of Pb and Cd used in the treatment determine a easy decrease of respiration intensity - indicator for metabolic activities - and water

content as well as the increase of the content of assimilating pigments (exception for Cd in concentration of 10 ppm).

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