DOI: 10.2429/proc.2013.7(2)063 2013;7(2)

Janina GOSPODAREK[1] and Aleksandra NADGÓRSKA-SOCHA[2]

EFFECT OF SOIL CONTAMINATION WITH HEAVY METAL MIXTURES ON IRON CONTENT IN BROAD BEAN (*Vicia faba* L.) PODS AND SEEDS*

WPŁYW SKAŻENIA GLEBY MIESZANINAMI METALI CIĘŻKICH NA ZAWARTOŚĆ ŻELAZA W STRĄKACH I NASIONACH BOBU *(Vicia faba* L.)

Abstract: The investigations were conducted to determine the effect of soil contamination with mixtures of: lead, copper and cadmium with zinc and nickel on two levels of pollution (acc. to II and III pollution class in IUNG classification) on iron concentrations in broad bean pods and seeds. Broad bean, White Windsor c.v. was cultivated in a control soil with natural heavy metal concentrations (Control and Control+NPK) and in the soil contaminated with the mixtures of heavy metals (Ni+Zn, Ni+Cd, Ni+Pb, Ni+Cu, Zn+Cd, Zn+Pb, Zn+Cu) applied in two doses, or with single heavy metals (Cd, Cu, Ni, Zn and Pb) used in a higher dose. Most of the analyzed heavy metal mixtures did not affect significantly iron concentrations in broad bean pods. Soil contamination with cadmium according to the dose established on the III pollution level acc. to IUNG classification and with a mixture of Ni with Cu and Zn with Cd acc. to the dose corresponding to II level of pollution in IUNG classification led to a decrease in iron concentrations in broad bean seeds.

Keywords: heavy metals, broad bean, accumulation, Fe

Iron is an element crucial to plants for their proper functioning. It participates in the photosynthesis, stimulates chlorophyll formation, takes part in nucleic acid metabolism and regulates oxidation-reduction reactions [1]. There is a considerable interdependence of iron and heavy metals metabolic activity [2]. Excess of metals may cause disturbances in the acquisition and transfer of iron, which in consequence influences development, chemical composition and nutritional value of plants. Pulse seeds are very valuable source of protein and vitamins from groups B and C, so they should be more frequently consumed by people [3]. Broad bean pods are also valuable source of important minerals like potassium, calcium and iron [4]. Heavy metals jointly present in soil often show a different effect upon plant than when occurring singly. Because out of the five heavy metals (Pb, Cd, Cu, Zn and Ni) analysed so far and applied separately, zinc and nickel revealed the strongest (negative) effect on broad bean plants growth and also caused considerable changes in macroelement concentrations in plants, including iron, it seemed purposeful to test the effect of soil contamination with mixtures of the above mentioned metals on two levels of pollution (acc. to II and III pollution class in IUNG classification) on iron concentrations in broad bean plants.

Materials and methods

The experiment was conducted on degraded chernozem developed from loess with acid reaction and organic carbon content 1.13%. Broad bean, White Windsor c.v. was

[1] Department of Agricultural Environment Protection, Agricultural University of Krakow, al. A. Mickiewicza 21, 31-120 Kraków, Poland, email: rrjgospo@cyf-kr.edu.pl
[2] Department of Ecology, University of Silesia, ul. Bankowa 9, 40-007 Katowice, Poland, email: olan@hoga.pl
*Contribution was presented during ECOpole'12 Conference, Zakopane, 10-13.10.2012

cultivated in a control soil with natural heavy metal concentrations (Control and Control+NPK) and in the soil contaminated with the mixtures of heavy metals (Ni+Zn, Ni+Cd, Ni+Pb, Ni+Cu, Zn+Cd, Zn+Pb, Zn+Cu) applied in two doses (marked respectively "II" and "III" corresponding to II and III class of pollution acc. to the classification suggested by IUNG [5], or with single heavy metals (Cd, Cu, Ni, Zn and Pb) used in a higher dose ("III"). The metal mixtures were used for the soil contamination in the same year when the plants were sampled for analysis, whereas in case of single metals the soil which was contaminated in the year preceding the experiment, *ie* in 2007 was used (it was marked respectively "(1)"). Doses of metals corresponding to II class were as follows: Cd - 2.25 mg $\cdot$ kg^{-1} d.m., Cu - 65 mg $\cdot$ kg^{-1} d.m., Pb - 175 mg $\cdot$ kg^{-1} d.m., Ni - 62.5 mg $\cdot$ kg^{-1} d.m., Zn - 350 mg $\cdot$ kg^{-1} d.m. and to III class respectively: Cd - 4 mg $\cdot$ kg^{-1} d.m., Cu - 85 mg $\cdot$ kg^{-1} d.m., Pb - 530 mg $\cdot$ kg^{-1} d.m., Ni - 110 mg $\cdot$ kg^{-1} d.m., Zn - 1000 mg $\cdot$ kg^{-1} d.m. Detailed data about the way in which metals were supplied to the soil and applied fertilization were presented in previous paper [6]. The samples for chemical analyses were collected at the seed full maturity. Plant material was washed in tap and in distilled water, dried in 105ºC to a constant weight and ground to fine powder, then mineralized and dissolved in 10% HNO_3. After filtration Fe content was measured using Flame Atomic Absorption Spectrometry (FAAS) [7, 8]. The quality of the analytical procedure was controlled by using samples of the reference material in each series of analysis (Certified Reference material CTA-OTL-1 Oriental Tobacco Leaves). The data were processed using software Statistica to compute significant statistical differences between samples ($p < 0.05$) according to Tukey's multiple range test.

Results and discussion

Some of tested heavy metal mixtures led to a significant limiting of plant growth, which could be seen in average weight of single seeds (Fig. 1). Particularly strong negative effect was observed for mixtures of cadmium and lead with nickel and zinc applied in a dose according to III class of pollution acc. to IUNG classification (Fig. 1). Much smaller than control were also broad bean seeds collected from plants cultivated in the soil contaminated with mixture of zinc and nickel in a dose established according to II class of pollution. On the other hand, metals used separately did not cause any significant differences in the weight of seeds with reference to the control. Weakening of broad bean growth under conditions of soil contaminated with zinc in a higher dose used separately and zinc in mixture with cadmium, resulted in non-formation of seeds by these plants. Zinc and nickel were supplied to the soil in the form of sulphate. Sulfur helped to lower the pH of soil, which had a negative impact on plant growth and also increased solubilization of heavy metals [9]. This may in turn lead to increased accumulation of heavy metals by plants [10].

Iron concentrations fluctuated from 20 to 34 mg $\cdot$ kg^{-1} d.m. in pods and from 7 to 31 mg $\cdot$ kg^{-1} d.m. in seeds of broad bean (Figs. 2 and 3). Iron concentration in bean pods reported by Kabata-Pendias [2] was 84 mg $\cdot$ kg^{-1}d.m., whereas in pea seeds 86 mg $\cdot$ kg^{-1} d.m. Average content of this element in common bean grains ranged between 34 and 89 mg $\cdot$ kg^{-1} d.m. [11, 12]. There is also a diversification in iron concentrations in various plant organs [13]. Iron concentrations in shoots and leaves of broad bean fluctuated from 73 to 318 mg · kg^{-1} d.m. depending on the analyzed objects, and in roots ranged from 164 to 666 mg · kg^{-1} d.m. [14], so they were much higher than in pods and seeds. In a majority of objects with contaminated soil no significant changes were observed in iron concentrations in pods (Fig. 2). It reached the highest value in broad bean pods exposed to contamination with copper or with a mixture of nickel and copper, both in a lower and higher dose. The uptake of iron (Fe) changed during Cu stress in *Prunus persica* L. Batsch grafted onto two different rootstocks [GF677 (p. perscia x P. amygdalus) and Mr.S2/5 (*P. cerasifera*)] when grown in the presence of 0.1, 10 or 100 μM copper sulphate ($CuSO_4$) [15].

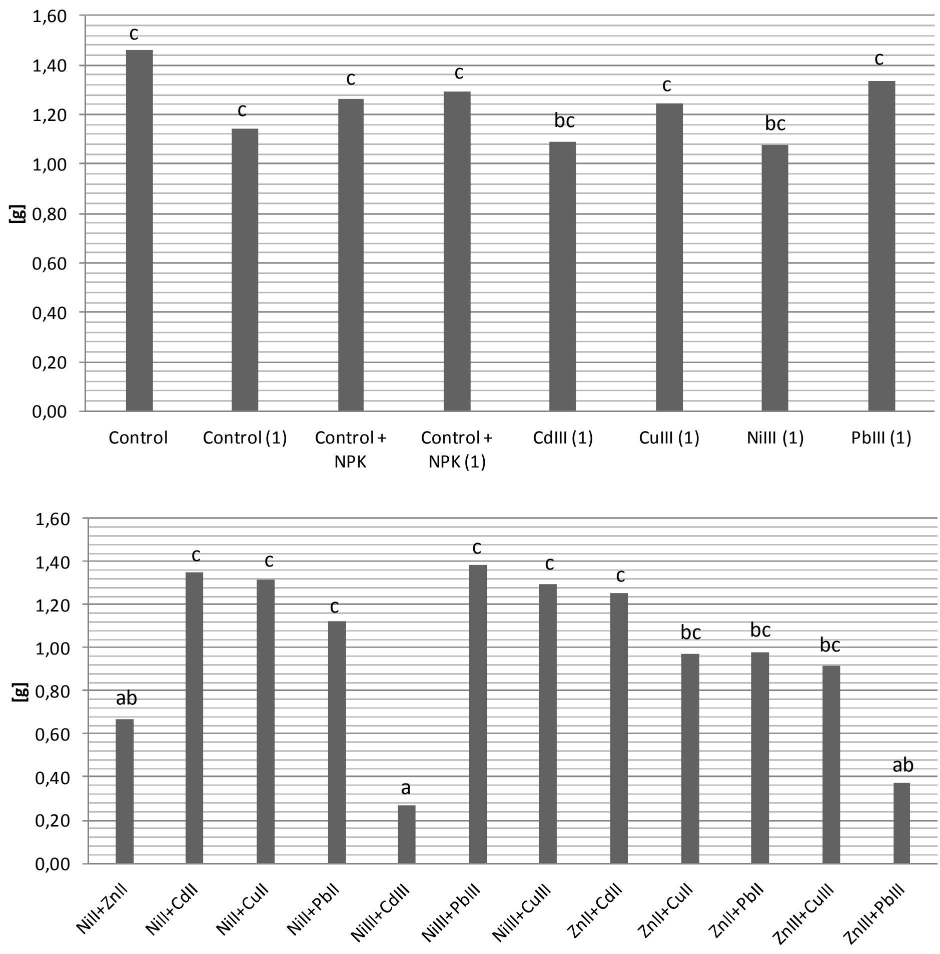

Fig. 1. Average weight of single seed of broad bean (*Vicia faba* L.) cultivated in unpolluted soil (Control, Control+NPK) and in soil contaminated with heavy metals. The (1) means the soil, which was polluted and used last year (2007). Values marked with different letters are statistically different at $p < 0.05$

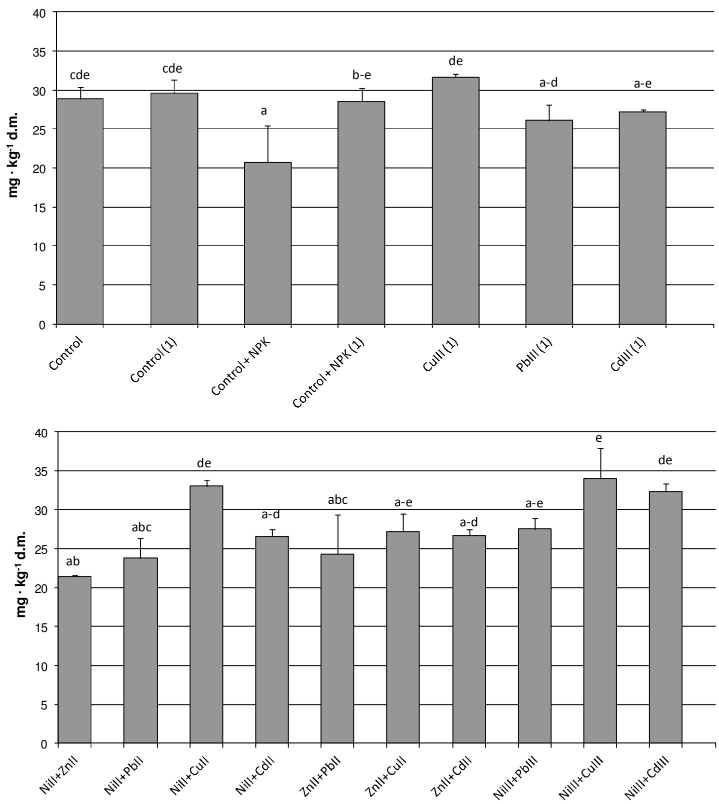

Fig. 2. Iron content in pods of broad bean (*Vicia faba* L.) cultivated in unpolluted soil (Control, Control+NPK) and in soil contaminated with heavy metals. The (1) means the soil, which was polluted and used last year (2007). Values marked with different letters are statistically different at $p < 0.05$

It proved impossible to gather a sufficient quantity of seeds for analysis from all treatments due to a phytotoxic effect of the applied metals [16]. A decrease in iron content was noted in seeds collected from contaminated plants. The most obvious decline (*ca* 5-fold) was registered in the object contaminated with a mixture of copper and nickel in a lower dose (Fig. 3). A significant decrease in iron content in comparison with the control plants was observed also in seeds collected from the plants growing in conditions of soil contaminated with a higher dose of cadmium and polluted with a mixture of zinc and cadmium in a dose calculated acc. to II level of pollution in IUNG classification. Iron deficiency may result from its complex joint effect with other elements in soil. Nickel is mentioned as one of the metals whose excess in soil leads to reduced absorption and translocation of iron, although a considerable decline in iron in seeds from plants polluted with a mixture of copper and nickel was not connected with a decreased level of this element in other plant parts [14]. However, iron does not migrate easily in plant tissues, so its deficiency appears in the first place in young plant tissues and in generative parts. The consequences of iron deficiency are disturbances of metabolic processes leading to reduced yield. In plants from the object polluted with a mixture of copper and nickel seed yield was 3-fold lower than obtained from the control plants receiving mineral fertilization. In the other objects, where metal mixtures were used, seed yield was twice lower than in the control plants [16]. A decline in iron content in broad bean seeds was registered also under the influence of swine liquid manure fertilization, although on the contrary, in leaves this measure caused increased content of iron and other heavy metals (Cd, Cu, Mn and Zn) [17]. The authors associated this with a decrease in the soil pH under the influence of this factor.

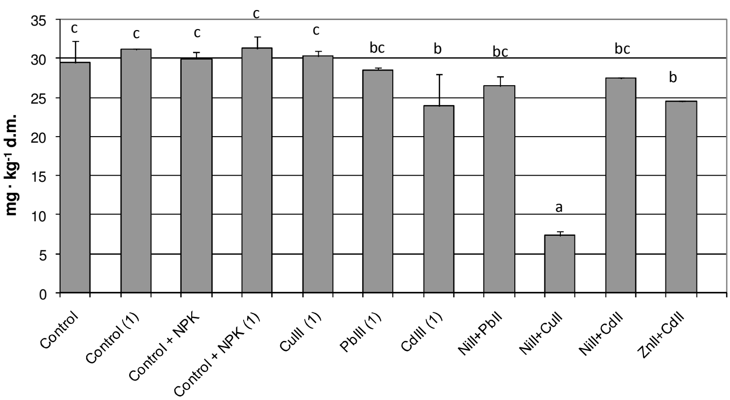

Fig. 3. Iron content in seeds of broad bean (*Vicia faba* L.) cultivated in unpolluted soil (Control, Control+NPK) and in soil contaminated with heavy metals. The (1) means the soil, which was polluted and used last year (2007). Values marked with different letters are statistically different at $p < 0.05$

Conclusions

1. Broad bean pods and seeds are characterized by lower iron concentrations than roots and shoots.
2. Most of the analyzed heavy metal mixtures did not affect significantly iron concentrations in broad bean pods. A slight increase in this element content was registered in the presence of Cu in soil or when mixtures of Ni and Cu and Ni and Cd were applied. On the other hand, on the other treatments Fe content was similar to the control object receiving mineral fertilizers.
3. Soil contamination with cadmium according to the dose established on the III pollution level acc. to IUNG classification and with a mixture of Ni with Cu and Zn with Cd acc. to the dose corresponding to II level of pollution in IUNG classification led to a decrease in iron concentrations in broad bean seeds.

References

[1] Graziano M, Lamattina L. Nutric oxide and iron in plants: an emerging and converging story. Trends in Plant Sci. 2005;10(1):4-8.
[2] Kabata-Pendias A, Pendias H. Biogeochemia pierwiastków śladowych. Warszawa: Wyd Nauk PWN; 1993.
[3] Messina MJ. Legumes and soybeans: overview of their nutritional profiles and health effects. Am J Clin Nutr. 1999;70(3 Suppl):439-450.

[4] Mateos-Aparicio I, Redondo-Cuenca A, Villanueva-Suarez MJ, Zapata-revilla MA, Tenorio-Sanz MD. Pea pod, broad bean pod and okara, potential sources of functional compounds. Food Sci Technol-Leb. 2010;43(9):1467-1470. http://dx.doi.org/10.1016/j.lwt.2010.05.008.
[5] Kabata-Pendias A, Motowicka-Terelak T, Piotrowska M, Terelak H, Witek T. Ocena stopnia zanieczyszczenia gleb i roślin metalami ciężkimi i siarką. Puławy: Wyd IUNG; 1993;ser. P:53.
[6] Gospodarek J, Nadgórska-Socha A. The effect of soil contamination with heavy metals in a mixture with zinc and nickel on their content in broad bean (Vicia faba L., ssp. maior) roots and shoots. Ecol Chem Eng. 2010;17(12):1571-1578.
[7] Ostrowska A, Gawliński S, Szczubiałka Z. Metody analizy i oceny właściwości gleb i roślin. Warszawa: Katalog Instytutu Ochrony Środowiska; 1991.
[8] Azcue J, Murdoch A. Comparison of different washing, ashing, and digestion methods for the analysis of trace elements in vegetation. Intern J Environ Chem. 1994;57:151-162.
[9] Seidel H, Wennrich R, Hoffmann P, Loser Ch. Effect of different types of elemental sulfur on bioleaching of heavy metals from contaminated sediments. Chemosphere. 2006;62(9):1444-1456. http://dx.doi.org/10.1016/j.chemosphere.2005.06.003.
[10] Kaplan M, Orman S, Kadar I, Koncz J. Heavy metal accumulation in calcareous soil and sorghum plants after addition of sulphur-containing waste as a soil amendment in Turkey. Agr Ecosys Environ. 2005;111(1-4):41-46. http://dx.doi.org/10.1016/j.agee.2005.04.023.
[11] Bassinello PZ, Oliveira BR, Lorrana NNN, Wellington MRS. Effect of the environment on zinc and iron levels in common beans. Annual Report of common bean Improvement Cooperative. 2010;10:146-147.
[12] Beebe S, Gonzalez AV, Rengifo J. Research on trace minerals in the common bean. Food and Nutrition Bulletin. 2000;21(4):387-391.
[13] Frossard E, Bucher M, Mächler F, Mozafar A, Hurrell R. Potential for increasing the content and bioavailability of Fe, Zn and Ca in plants for human nutrition. J Sci Food Agr. 2000;80(7):861-879. DOI: 10.1002/(SICI)1097-0010(20000515)80:7<861::AID-JSFA601>3.0.CO;2-P.
[14] Gospodarek J, Nadgórska-Socha A. The effect of soil contamination with heavy metal mixtures of: lead, copper and cadmium with zinc and nickel on iron content in broad bean (Vicia faba L.). Ecol Chem Eng. in press.
[15] Francini A, Sebastiani L. Copper effects on Prunus persica in two different grafting combinations (P. persica × P. amygdalus and P. cerasifera). J Plant Nutr. 2010;33(9):1338-1352. DOI: 10.1080/01904167.2010.484094.
[16] Gospodarek J. Effect of soil contamination with a mixture of heavy metals on broad bean (Vicia faba L.) seed quality. Ecol Chem Eng A. 2011;18(12):1651-1657.
[17] Li F, Ni L, Yuan J, Sheng GD. Cultivation practices affect heavy metal migration between soil and Vicia faba (broad bean.). Chemosphere. 2010;80(11):1393-1398. DOI: 10.1016/j.chemosphere.2010.06.001.

WPŁYW SKAŻENIA GLEBY MIESZANINAMI METALI CIĘŻKICH NA ZAWARTOŚĆ ŻELAZA W STRĄKACH I NASIONACH BOBU *(Vicia faba* L.)

Katedra Ochrony Środowiska Rolniczego, Uniwersytet Rolniczy w Krakowie

Abstrakt: Celem pracy było określenie wpływu skażenia gleby mieszaninami metali ciężkich: ołowiu, miedzi i kadmu z cynkiem i niklem na dwóch poziomach zanieczyszczenia (wg II i III klasy zanieczyszczenia zgodnie z klasyfikacją IUNG) na zawartość żelaza w strąkach i nasionach bobu. Bób odmiany Windsor Biały był uprawiany w glebie o naturalnej zawartości metali ciężkich (Kontrola i Kontrola+NPK) oraz w glebie skażonej mieszaninami metali ciężkich (Ni+Zn, Ni+Cd, Ni+Pb, Ni+Cu, Zn+Cd, Zn+Pb, Zn+Cu), zastosowanymi w dwóch dawkach, lub pojedynczymi metalami ciężkimi (Cd, Cu, Ni, Zn i Pb) zastosowanymi w wyższej dawce. Większość z badanych mieszanin metali ciężkich nie wpłynęła istotnie na zawartość żelaza w strąkach bobu. Skażenie gleby kadmem wg dawki ustalonej na poziomie III stopnia zanieczyszczenia wg klasyfikacji IUNG oraz mieszaniną Ni z Cu i Zn z Cd wg dawki ustalonej na poziomie II stopnia zanieczyszczenia wg klasyfikacji IUNG prowadzi do obniżenia zawartości żelaza w nasionach bobu.

Słowa kluczowe: metale ciężkie, bób, akumulacja, Fe