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EFFECT OF SELENIUM AND MOLYBDENUM CONTENT IN RHIZOBOXES ON ELEMENT UPTAKE OF MAIZE AND SUNFLOWER

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Abstract

Selenium and molybdenum are essential trace elements.
Considering the characteristics of selenium and molybdenum, our research had the following purposes for both elements:
Considering Se the uptake of selenium was investigatein maize and sunflower seedling, moreover the different effect of two selenium species (selenite and selenite) for the examined plants (a monocotyledon (maize, Zea mays L.) and a dicotyledon (sunflower, Helianthus annuus L.)).
Considering Mo on the one sidedue to increasing level of molybdenum treatment, the concentrations of changes of Mo was investigated in maize and sunflower seedlings. On the other sidewe thought it is important to follow the concentration of this element because it plays prior role in nitrate reduction and operation of nitrate reductase. So we would have liked to prove in laboratory circumstances that there is a close relation between molybdenum supply and nitrate reduction: nitrate content of plants can be reduced by supporting their physiological molybdenum demand.
In our experiments for selenite 0 (control), 1, 10 and 100 mg kg^{-1} selenium concentrations, forselenate0, 0.1, 1 and 10 mg kg^{-1} selenium concentrations, while for molybdenum doses 0, 30, 90, 270 mg kg^{-1} were applied.
According to our results it is obvious that concentration of selenium and molybdenum in seedlings significantly was increased due to selenium or molybdenum treatments. Examining roots and shoots of experimental plants separately we found higher selenium and molybdenum content in roots than in shoots. It indicates more intensive selenium and molybdenum uptake (selenium and molybdenum mobility) and high molybdenum content shows nitrate accumulation of shoots under the given experimental circumstances.

Keywords: *maize, molybdenum, rhizobox, selenium, selenite, selenate, sunflower.*

INTRODUCTION

Selenium (Se) is an essential micro-nutrient, particularly in an animal and a human body. Se is a vital component of antioxidant system of organism. Deficiency of selenium is connected with emergence of many diseases among others the heart and vascular system one and the tumorous diseases. Its contradiction derives from above given concentration dangerous to plants and human too. In the periodical system, selenium has the narrowest tolerance concentration range that is essential and toxic contents are close to one another.

Selenium content of a plant is determined by the species of the plant, it's developmental state, selenium supply of soil and the type of selenium species (Lteif et al., 2005). A plant can uptake only the biology available element species from the environment (Chengyi et al., 2005). Selenium bonded to the organic part of the soil is not mobile and this selenium is available to a few indicator plants only. These plants are Se-transformers, which are excellent Se-source for the others plants after their wither (Kádár, 1998). The average content of selenium in plants is 0.01-2 μg kg^{-1} (Kovács et al., 1998). The inorganic selenium is transformed into organically bonded selenium by plants, hence it becomes uptakeable selenium for a human body (Bankhofer, 1994). The selenium content is the largest in the roots generally and smaller

in a young root, in a leaf, in a stalk and the smallest in a grain, which is a special filtering system in a plant (Kádár, 1995).

Certain plants are capable to rich selenium content in their organism, these plants are named as accumulators (Robb and Pierpoint, 1983). The vast majority of plants are not Se-accumulator, but those are Se-sensitive (Terry et al., 2000).

Molybdenum (Mo) as an essential trace element has an important role in nitrogen metabolism.

Comparing the amounts of microelements occurring in soil we can state that molybdenum is in small quantities in soils, however, it is enough for most of cultivated plants (Zimmer and Mendel, 1999). Average molybdenum content of soils is 0.25-5 mg kg^{-1} (Schulte, 2004). In soils molybdenum occurs in minerals and bound to iron and aluminum hydroxides (Fontes and Coelho, 2005).

Under pH 6 soils bind molybdenum rather strongly, therefore in acidic soils molybdenum deficiency can occur easily (Aubert and Pinta, 1977). Molybdenum uptake from basic soils is much more considerable due to the fact that its solubility in contrast to most of the microelements is more increased in basic conditions, so exchange of molybdenate ions of sorption complexes with hydroxide ions of soil solution is probably more intense. Availability of molybdenum can be improved most by raising of hydroxide ion content of soil solution (Berger and Pratt, 1965).

Plants are much more sensitive to molybdenum deficiency than to excess. In case of insufficient molybdenum supply sugar content and intensity of photosynthesis decrease, biosynthesis of ascorbic acid is restrained, therefore eg. C-vitamin content of leaves may decrease to the quarter of normal level (Wang et al., 1999). Growth of these plants slows down, leaves turn pale, although blooming is disturbed. Signs of deficiency appear mostly on central and older leaves. Color of leaves is yellowish, leaf margins may be twirled and chlorosis between leaf veins is frequent (Bambara and Ndakidemi, 2010; Zaijun et al., 2005; Schulte, 2004).

Molybdenum demand of different plants is not equal. Papilionaceous plants (soy bean, pea, bean, alfalfa) accumulate much more molybdenum than others and distribution of it is not constant in plant parts. According to Szalai (2006) there is much more molybdenum in seed than in vegetative organs. Higher molybdenum demand of papilionaceous plants is in connection with the presence of Rhizobium bacterium on their roots. These bacterium need molybdenum to fix nitrogen since the enzyme catalyzing this process called nitrogenase contains molybdenum ((Williams and da Silva, 2002; Gupta et al., 2011; Loch and Nosticzius, 1992).

Certain plants (eg. cauliflower, cabbage, spinach, lettuce and tomato) are especially sensitive to molybdenum deficiency thus they can be used as indicator plants (Katyal and Radhawa, 1983; Duval et al., 1991).

Appropriate molybdenum supply is necessary not only because of avoiding different deficiency symptoms but because the fact that molybdenum has a key function in nitrate reduction as the cofactor of nitrate reductase(Berks at al., 1995; Yaneva et al., 1996; Tunçeli and Türker, 2004). In lack of molybdenum nitrate reductaselooses its activity, nitrate reduction declines resulting in the accumulation of nitrate.

Molybdenum is essential for plants and animals as well. It is a component of several enzymes and enzyme systems contributing to appropriate operating of cells and growth (Reilly, 1991).

Our attention to the harmful effect of nitrate was drawn by the appearance of methaemoglobinaemia, what leads to anaemic disease or in worst case to death by suffocation of infants. In case of methaemoglobinaemia nitrate is reduced to nitrite in the ventricle. Nitrite transforms haemoglobin to methaemoglobin by oxidising central Fe^{2+} ion to Fe^{3+} making it unable for transporting oxygen. Methaemoglobin can be found at lower level (0.5-3.0%) also in blood of healthy people not causing any abnormal symptoms up to 10%, but over it cyanosis may occur and over 25% increasing pulse rate and short breathing can be observed (Knowles et al., 1989). Methaemoglobin concentration over 50% may cause death by suffocation. These present the importance of optimal operating of nitrate reductase, which demands optimal amount of molybdenum.

MATERIALS AND METHODS

A monocotyledon (maize, *Zea mays L. cv Norma SC*) and a dicotyledon (sunflower, *Helianthus annuus L. cv Arena PR*) were chosen for our research studying selenium and molybdenum moreover nitrogen speciesin its root and shoot parts,separately.

The concentration of selenium and molybdenum were analyzed in root and shoot, furthermore in soil samples. Beside molybdenum concentration,the change of NO_3-N, NO_2-N and NH_4-N concentration was also observed since the nitrate reduction processwas also examined in the rhizobox experiment applied different molybdenum levels.

Maize and sunflower plants were grown in the climate room of Institute of Crop Science, Department of Agricultural Botany and Crop Physiology where environmental conditions were regulated: 65-75% relative humidity (RH), 25/20 °C temperature periodicity (day/night), 220 $\mu E\ m^{-2}\ s^{-1}$ light intensity, 16 hours/8 hours light/dark period.

After disinfection seeds were germinated between moistened filter papers, stimulated geothropically at 22 °C. Seedlings with 2-3 cm coleoptils were placed into rhizoboxes (Photos 1-2.).

Photo 1: Maize seedlings grown in rhizobox (control)

Photo2: Sunflower seedlings grown in rhizobox (control)

Experiments carried out in rhizoboxes:

Advantages of experiments carried out in rhizoboxes are that growing and daily growing rhythm of roots of maize can be followed up moreover also possible phytotoxic symptoms of roots resulting from increasing molybdenum dose can be seen.

Calcareous chernozemsoil type from Látókép Experimental Station of University of Debrecen was applied for our studies.

Composition of the applied soil can be seen in Table 1. Parameters correspond with the ones of soil used in experiment of Nagy et al. (2010), there was no NPK fertilization.

In our experiments in the case of selenite 1, 10 and 100 $mg\ kg^{-1}$ selenium concentrations (Photos 3-4.), in the case of selenate 0.1, 1 and 10 $mg\ kg^{-1}$ selenium concentrations were applied, furthermore there was also control (Ø) treatment. Molybdenum doses (Photos5-6.)were as follows: Ø (control), 30, 90, 270 mg kg^{-1}. Selenium was applied in the form of selenite ($Na_2SeO_3.5H_2O$) (Fluka, Buchs, Switzerland) and selenate (Na_2SeO_4) (Sigma-Aldrich, Steinheim, Germany) dissolved in ultrapure water. The applied molybdenum salt ($(NH_4)_6Mo_7O_{24}.4H_2O$) was dissolved in distilled water. The required concentrations were calculated for selenium and molybdenum. Each treatment was applied in three repetitions and in calcareous chernozem.

Table 1: Parameters of soil applied in the experiments carried out in rhizoboxes

Depth	0-0.3 m
pH (KCl)	5.71
pH (H_2O)	6.58
Soil texture category	loamy clay
Total water-soluble salt	0.015 %
$CaCO_3$	0.202 %
Humus	3.54 %
KCl-soluble NO_3-N+NO_2-N	8.04
AL-soluble P_2O_5	199 mg kg^{-1}
AL-soluble K_2O	451 mg kg^{-1}
AL-soluble Na	332 mg kg^{-1}
KCl-soluble Mg	176 mg kg^{-1}
KCl-soluble SO_4^{2-}-S	6.04 mg kg^{-1}
KCl-EDTA-soluble Cu	5.79 mg kg^{-1}
KCl-EDTA-soluble Zn	7.9 mg kg^{-1}
KCl-EDTA-soluble Mn	262 mg kg^{-1}

Before placing soil into the rhizoboxes, moistened filter paper was placed onto the bottom of boxes, thus steady water uptake of plants was ensured. After putting seedlings into the prepared soil, side plastic wall of rhizoboxes were covered by black foil. Plants were gheothropically stimulated so roots grew along the plastic wall of box making it possible to follow growing of roots.
Weight of rhizoboxes and length of roots were measured daily and also evaporated water was added each day.

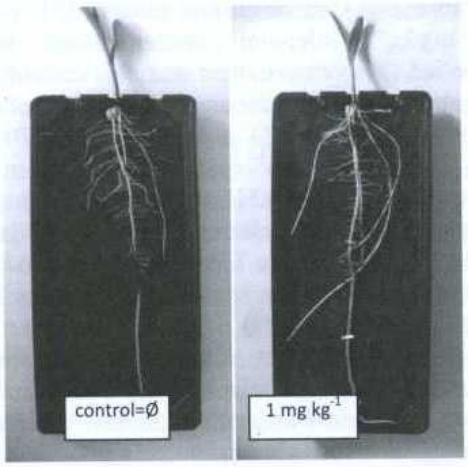

Photo 3.Maize in rhizoboxes, selenite treatments (control and1mg kg^{-1} selenium)

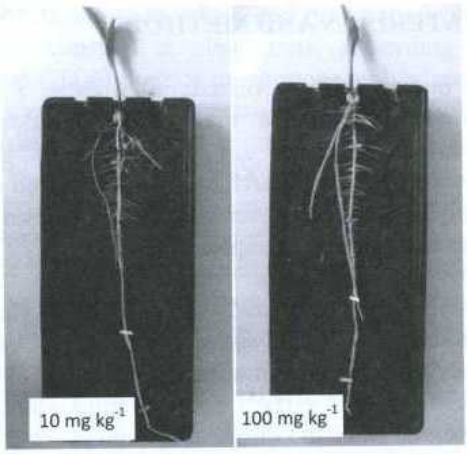

Photo 4.Maize in rhizoboxes, selenite treatments (10 and 100 mg kg^{-1} selenium)

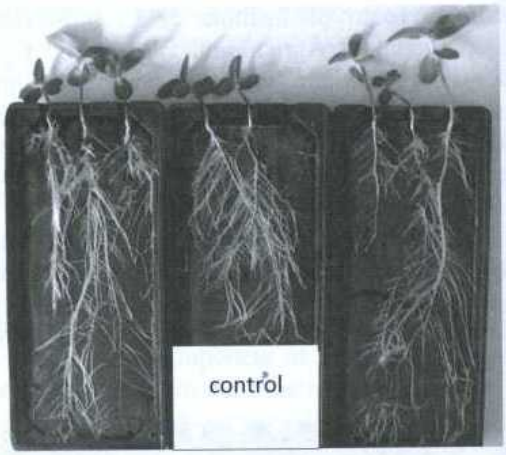

Photo 5.Three replicates of the control Mo treatment, sunflower in rhizoboxes

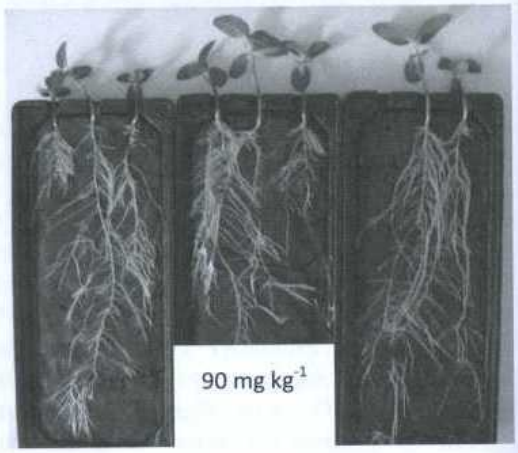

Photo 6.Three replicates of the 90 mg kg^{-1} molybdenum treatment, sunflower in rhizoboxes

Finishing experiment plants grown in rhizobox were dried at 85 °C to constant weight and weighed by analytical balance (OHAUS) after cooling them to room temperature.

After drying and homogenization samples were subjected to HNO_3-H_2O_2 wet digestion (Kovács et al., 1996). 1 g (±0.01 g) sample was measured. After adding 10 cm^3 cc. HNO_3 they were predigested for a night. Next day the samples were digested by LABOR MIM OE 718/A digestion instrument: 45 min at 60 °C; then 90 min at 120 °C (after addition of 3 cm^3 30% H_2O_2). After cooling they were filled up with deionized water to 50 cm^3, mixed by shaking and filtered through FILTRAK 388 type filter paper. Also blank experiment was done.

Concentrations of elements were determined by inductively coupled plasma optical emission spectrometry (ICP-OES) (Perkin Elmer OPTIMA 3300 DV) and inductively coupled plasma mass spectrometry (ICP-MS) (Thermo Elemental X7). Setting and measuring parameters correspond with the ones applied by Puskás-PresznerandKovács(2009).The interfering polyadducts were eliminated by the use of collisional cell technique (CCT) in kinetic energy discrimination (KED) mode (Szebeniet al., 2011). For selenium the 196.026 nm, while for molybdenum the 202.031 nm wavelengths in inductively coupled plasma optical emission spectrometerand two different selenium isotopes (^{78}Se and ^{80}Se) and three different molybdenum isotopes (^{95}Mo, ^{96}Mo and ^{98}Mo) were used in inductively coupled plasma mass spectrometer for analysis of selenium and molybdenum concentrations in plant and soil samples.

NO_3-N, NO_2-N and NH_4-N concentration of plant samples were determined by FIAstar 5000 Analyzator.

Statistical analysis:

Experimental data were evaluated by bivariate general linear model (GLM), which is a combination of analysis of variance and linear regression analysis. Statistical analysis was done by SPSS 13.0.

RESULTS AND DISCUSSIONS

Experiments with selenium:

The selenium concentrations of shoots and roots of maize and sunflower plants are shown in Tables 2-3., where the plants were grown in rhizoboxes and in different selenium levels of contaminated soil. The elongation of roots of applied plants cultivated in a rhizobox was retarded and the differentiation of the lateral roots was moderate also.

*Table 2.Se concentration (mg kg^{-1}) of maize and sunflower in rhizobox as a result of selenite treatment (***: 0.1% level of significance)*

	Se concentration			
Selenite	Maize		Sunflower	
Treatments	Shoot (***)	Root (***)	Shoot (***)	Root (***)
Ø	0.736±0.248	0.339±0.04	0.282±0.163	0.12±0.034
1	4.54±0.19	3.10±1.40	3.81±1.64	7.11±0.61
10	5.85±0.15	11.6±1.0	1.93±0.93	13.8±2.7
100	32.8±4.6	162±15	10.3±0.9	341±21

The higher the selenium level in the soil the higher the selenium concentration in a plant sample. The same selenate concentration caused much higher selenium content in a plant sample then the same selenite concentration produced.

*Table 3.Se concentration (mg kg^{-1}) of maize and sunflower in rhizobox as a result of selenate treatment (***: 0.1% level of significance)*

	Se concentration			
Selenate	Maize		Sunflower	
Treatments	Shoot (***)	Root (***)	Shoot (***)	Root (***)
Ø	0.736±0.249	0.339±0.04	0.282±0.163	0.120±0.034
0.1	13.1±1.6	3.9±0.1	13.9±2.0	8.47±0.37
1	213±43	42.7±5.7	209±39	33.6±3.0
10	570±50	837±163	213±46	258±62

The maize and the sunflower plants have taken up comparatively small quantity of selenium from the control soil, however the higher selenium treatments showed much higher selenium concentration both in the shoot and in the root also. It is established that the roots of the maize and the sunflower have taken up two times concentration of selenium then of the

shoots. It is assumed that the transport into the shoot was retarded.

In the case of phytoremediation of soil contaminated by selenium it is important to know what amount of selenium can be taken up by a plant from a contaminated soil, hence what duration is needed to eliminate the selenium contamination.

Table 4.shows the ratios between the selenium contents of shoots in the lowest (control) and in the largest treatments of maize and sunflower in the rhizoboxes (soil).

Table 4. Ratios and the selenium concentrations of selenite and selenate treatments in maize and sunflower

	monocotyledons (maize)	dicotyledons (sunflower)
selenite treatment:	45X (0.736 and 32.8 mg/kg)	41X (0.249 and 10.3 mg/kg)
selenate treatment:	775X (0.736 and 570 mg/kg)	859X (0.249 and 215 mg/kg)

The concentration values of plant shoots in parentheses are the selenium contents of the lowest, i.e. control (Ø) and the largest selenite (which is 100 mg/kg), and selenate (which is 10 mg/kg) treatments.
On the basis of Table 4.the largest selenite treatment (100 mg/kg selenium) caused approximately 40-50 times increment in selenium concentration, while the applied largest selenate treatment (what was only 10 mg/kg selenium) resulted approximately 800-900 times increment in selenium concentration of the shoot. This means, the selenium uptake applying selenate is much better approximately (200 times) greater than the uptake of selenite by a monocotyledon (eg. maize) or a dicotyledon (eg. sunflower).

Experiments with molybdenum:

In Figure 1. change of molybdenum concentration in shoots and roots of maize and sunflower seedlings depending on the molybdenum treatment is shown.

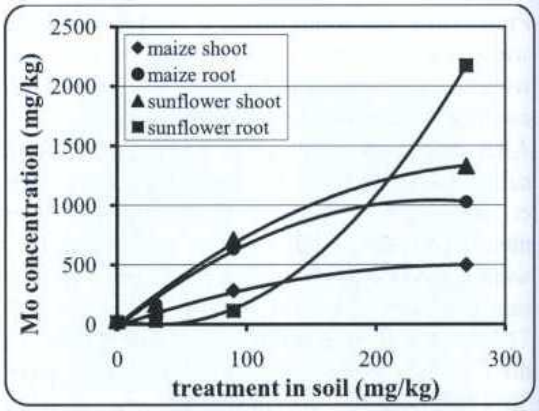

Figure 1: Molybdenum concentration of shoots and roots of maize and sunflower seedlings grown in rhizoboxes

Considering Figure 1.themaize and sunflower seedling took up small amount of molybdenum from control soil. Molybdenum concentration was low in shoot and root as well but owing to molybdenum treatments increase was observed. Kádár (1995) came to similar results during his microelement-load field experiment set on calcareous chernozem soil. According to his result load caused extremely high molybdenum accumulation in maize and sunflower, however excepting control treatment there was only inconsiderable difference between molybdenum content of organs above and under the surface. These data show us that in our research a more intensive concentration growth occurred. Furthermore a higher increase of molybdenum concentration was observed in the case of sunflower as a dicotyledon shoot and root as well.
Molybdenum treatments had effects also on N-forms. NO_3-N content of maize and sunflower shoots was variable (Tables 5-6.), the lowest and the highest molybdenum treatment in maize experiment served the highest values (Table 5.). Comparing these results to the maize experiment of Kádár et al. (2000) differences can be found. In their experiment in case of ammonium-paramolybdenate a definite increase of NO_3-N concentration occurred. We suppose that N added in the form of ammonium had been nitrified by that time and this increased supply was reflected by the leaves. Concentration of NH_4-N grew in shoots in compliance with increasing molybdenum

concentration, which indicated a definite connection between molybdenum treatments and more intensive nitrate reduction.

The effect of increasingmolybdenum treatments increased the measuredNO_3-N concentration in theshoots androots of sunflower seedlings, and the NH_4-N concentration did not show a similar tendency, which indicated that thenitrate reductasesystem capacity was not unlimited, saturated, which led to the accumulation of nitrate (Table 6.). This is a kind of protective reaction of the plant (sunflower), because in this way prevents the accumulation of toxic ammonia, while non-toxic nitrate is selected into vacuoles for the plant, thus the ammonia temporarily is excluded from the metabolism.

Table 5: Nitrate- (NO_3-N), nitrite- (NO_2-N) and ammonium-nitrogen (NH_4-N) concentration(mg kg^{-1}) of shoots of maize seedlings grown in rhizoboxes in case of different molybdenum doses (mg kg^{-1})

Mo-treatment	NO_3-N	NO_2-N	NH_4-N
control	41.5	0.424	237
30	0.736	0.681	331
90	8.44	0.102	308
270	214	1.51	401

Table 6: Nitrate- (NO_3-N), nitrite- (NO_2-N) and ammonium-nitrogen (NH_4-N) concentration (mg kg^{-1}) of shoots of sunflower seedlings grown in rhizoboxes in case of different molybdenum doses (mg kg^{-1})

Mo-treatment	NO_3-N	NO_2-N	NH_4-N
control	73.3	0.040	647
30	173	0.040	446
90	313	0.445	547
270	899	0.382	549

In the root of maize and sunflower seedlingsthe capacity of nitrate reductase was higher, asowing to the effect of increasingmolybdenum treatments, the measured NO_3-N concentration was increased in the root of maize and sunflower also (Tables 7-8.), moreover the NH_4-N concentration of sunflower was also increased (Table 7.), while in the maize it was decreased (Table 6.). A major part of the absorbed nitrate was reduced to ammonium that linking to glutamic acid took part in amino acid synthesis through transaminating reactions and thus in protein synthesis.

On the basis of our results, thereis obvious close relation among the molybdenum level, NO_3-N absorption and nitrate reduction. The system is finely controlled, ammonium is not able to accumulate in tissues.

High concentration of free ammonium cytotoxic because infiltrating in the energizing (mainly mitochondrial) membranes acts as disconnecting factor that is depolarises the membrane without synthesizing ATP. Since plants didn't show toxic symptoms we suppose that there was enough glutamic acid for absorbing ammonium. From these we conclude that citric acid cycle that is the part of dehidrogenating phase of respiration serving carbon skeleton of glutamic acid was also active. All these refer to intensive metabolism in which role of molybdenum was confirmed by our experiments too. Establishments set in relation with shoot are relevant also in case of maize roots, which means nitrate reductase activity of roots and shoots were roughly equal in experiment carried out in rhizobox (Table 7.).

As a result of our experiments it can be concluded that molybdenum plays an important role in reduction of nitrate.

Table 7: Nitrate- (NO_3-N), nitrite- (NO_2-N) and ammonium-nitrogen (NH_4-N) concentration (mg kg^{-1}) of roots of maize seedlings grown in rhizoboxes in case of different molybdenum doses (mg kg^{-1})

Mo-treatment	NO_3-N	NO_2-N	NH_4-N
control	35.5	0.040	280
30	15.8	0.040	334
90	15.8	0.459	333
270	134	2.84	200

Table 8: Nitrate- (NO_3-N), nitrite- (NO_2-N) and ammonium-nitrogen (NH_4-N) concentration (mg kg^{-1}) of roots of sunflower seedlings grown in rhizoboxes in case of different molybdenum doses (mg kg^{-1})

Mo-treatments	NO_3-N	NO_2-N	NH_4-N
control	15.6	0.040	443
30	50.3	0.040	507
90	162	1.76	544
270	285	3.66	824

CONCLUSIONS

The consumption of selenium sources of natural products, such as whole-grain cereals, wheat germ, brown rice, nuts, sesame seeds, soy and garlic, not in any case provides enough amount of selenium for human-input, especially if the soil is lack of selenium. In many European countries the soils contains quite low selenium. Therefore, selenium supplementation becomes necessary in these areas, and where people are proven to suffer from selenium deficiency. Today, the number of patients which are lack of selenium is decreasing, so selenium supplementation seems to be inevitable. One way you can replace the selenium, that a selenium salt (selenite or selenate) is added into the soil for the cultivated plants. One of the problems with the supply of adequate selenium to the plants in most cases do not show well defined deficiency symptoms among selenium deficient growing conditions.

In order to produce healthy food, therefore it may be appropriate to monitor selenium contents of soils and the crops.

In our experiment in soil (in rhizoboxes) the effect of selenium supply was studied in a monocotyledon (maize) and a dicotyledon (sunflower) plants among controlled conditions. In the experiments the dose was calculated and added as selenium in selenite form (concentrations: Ø, 1, 10, 100 mg kg^{-1}) and in selenate form (Ø; 0,1; 1; 10 mg kg^{-1}). On the basis of our results the selenium content of plants was increased significantly by the effect of selenium treatments. This increase of selenium concentration was more intensive by the effect of selenate treatment than by the effect of selenite treatment applying the same level of treatment. The selenium concentration of shoot and root samples was analyzed respectively. Se content was higher in roots than in shoot samples in the case of maize and sunflower as well. This shows that the selenium accumulation in roots was more intensive than in shoots of the applied plants among the applied conditions.

The rhizobox experiment can not be performed in selenium free environment, since the control soil contains also certain amount of selenium. Despite this, the sunflower plant could uptake only a relatively small amount of selenium from control soil. However, the maize has got more selenium from the control soil then the sunflower and it continued to increase as a result of the selenate treatments. This difference is explained by the difference nutrient uptake mechanism of the two types of plant (maize and sunflower).

In conclusion it can be stated that it is extremely important for proper selenium content in soil. Considering the selenium content of a soil, although the presence of selenium is important for the plants, however too high concentration is a serious threat to the plants, animals and humans. Owing to the relatively high tolerance of maize and sunflower to selenium they can be applied for phytoremediation of selenium-contaminated areas, which is an environmentally friendly and relatively inexpensive method of decontamination of a soil, however it is time consuming activity.

The uptake of molybdenum by two experimental plants (maize and sunflower) was examined with increasing molybdenum concentration; moreover the effect of molybdenum supplying was also investigated on the process of nitrate reduction. Examination of the relationship between the molybdenum supplying and the nitrate reduction was also investigated, as the presence of molybdenum which has key role of nitrate reduction to enzyme activity of nitrate reductase is essential. In the absence of molybdenum the process of the nitrate reduction will be slow down and this results the accumulation of nitrates in the plant.

In our rhizobox experiment, molybdenum was not added to the control treatment of soil, furthermore the molybdenum concentrations of the other treatments were as follows: 30, 90, 270 mg kg^{-1}.

On the basis of our results it seems that as a result of Mo-treatments, the content of molybdenum in the plants has increased substantially. This increase of sunflower seeds was more intensive, than of the maize. Analyzing the plant parts (eg. root and shoot) separately, the concentration of molybdenum in the root of maize was higher than in the shoot of maize, however only in the case of the largest sunflower treatment (270 mg kg^{-1})

resulted higher molybdenum concentration in the root.

The Mo treatments influenced by various forms of nitrogen. In those cases when the Mo treatments reduced the roots NO_3-N content and increased the NH_4-N concentrations, then more intensive nitrate reductase activity was postulated. The examples were also found for those NO_3-N concentration of the root and shoot increased by the influence of molybdenum treatments, but those were not followed higher NH_4-N concentration in the examined plants, which according to our assumption this phenomenon is explained by a kind of defense mechanism of the plant.

Based on the results of the attempt, we found clear evidence that there is a close correlation between the molybdenum supply of the plants and the reduction of nitrate.

Knowing this fact, in the case of intensive addition of NO_3-N, we have to take the original molybdenum concentration in the soil into consideration, moreover if the free molybdenum concentration in the soil does not reach the plant's physiological needs (that is approximately 0.01 μM Mo) into account also. If it is true then a micro-nutrient fertilizer containing molybdenum is reasonable to add into the soil. To ensure adequate supply of Mo the nitrate content in the leaf and root vegetables can be reduced, to produce and to consume healthier raw materials and foods.

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REFERENCES

Aubert H., Pinta M.: 1977. Trace elements in soils. Elsevier Scientific Publishing Company, 55-61.

Bambara S., Ndakidemi P.A.: 2010. The potential roles of lime and molybdenum on the growth, nitrogen fixation and assimilation of metabolites in nodulated legume: A special reference to Phaseolus vulgaris L., African Journal of Biotechnology, 8:2482-2489.

Bankhofer H.: (1994) Bio-szelén. Golden Book Kiadó, Budapest.

Berger K. C., Pratt P F.: 1965. Advances in secondary and micronutrient fertilization. In: McVicka M. M. Bridger G. L. Nelson L. B. (eds), Fertilizer technology and usage. Soil Science Society of America, Madison, Wisconsin, USA, 287-340.

Berks B. C., Ferguson S. J., Moir J. W. B., Richardson D. J.: 1995. Biochim.Biophys.Acta - Bioenergetics, 1232:97.

Chengyi Z., Ren J., Xue C., Lin E.: (2005) Study on the relationship between soil selenium and plant selenium uptake. Plant and Soil. 277:197-206.

Duval, L., More E., Sicot: A.: 1991. Observations on molybdenum deficiency in cauliflower in Brittany.ComtesRendus de I Academic d Agriculture de France, 78:27-34.

Fontes R. L. F., Coelho H. A.: 2005. Commun. Soil Sci. Plant Anal., 36:2367–2381.

Gupta U. C., Srivastava P. C., Gupta S. C.: 2011. Role of Micronutrients: Boron and Molybdenum in Crops and in Human Health and Nutrition. Curr.Nutr. Food Sci., 7:126-136.

Kádár I., Koncz J., Gulyás F.: 2000. Mikroelem-terheléshatása a kukoricaösszetételeréés a talajkönnyenoldhatóelemtartalmárakarbonátoscsernozjo mon.AgrokémiaésTalajtan, 49:205-220.

Kádár I.: 1995. A talaj-növény-állat-ember táplálékláncszennyeződésekémiaielemekkelMagyarországon.KörnyezetvédelmiésTerületfejlesztésiMinisztérium MTA TalajtaniésAgrokémiaiKutatóIntézete, Budapest.

Kádár I.: 1998. Szelénforgalma a talaj-növényrendszerben. [In: Cser M. Á. ésSziklai-László I. (szerk.) A szelénszerepe a környezetbenésegészségvédelemben.] Frag Bt., Budapest.

Katyal J. C., Randhawa N. S.: 1983. Micronutrients (molybdenum).FAO Fert.Plant Nutr. Bull., 7:69-76.

Knowles R. G., Palacios M., Palmer R. J. M., Moncada S.: 1989. Formation of nitric oxide from L-arginine in the central nervous system: a transduction mechanism for stimulation of the soluble guanylatecyclase. Proc. Natl. Acad. Sci., 86:5159-62.

Kovács B., Győri Z., Prokisch J., Loch J. and Dániel P.: 1996. A study of plant sample preparation and inductively coupled plasma emission spectrometry parameters. Communications in Soil Science and Plant Analysis, 27(5-8):1177-1198.

Kovács M., Turcsányi G., Siller I., Engloner A.: 1998. A Se elöfordulásaMagyarországnövénytársulásaiban. 85. [In: Cser M. Á. ésSziklai-László I. A szelénszerepe a környezetbenésegészségvédelemben.] Frag Bt., Budapest.

Loch, J., Nosticzius, Á.: 1992. Agrokémiaésnövényvédelmikémia.MezőgazdaKiadó, Budapest.

Lteif A., Bashour I., Nimah M. and Darwish R.: 2005. Selenium Levels in Beqaa Valley in Lebanon. In: Lee et al., Plant Nutrition for Food Security, Human Health and environmental Protection. Tsinghua University Press, Beijing, China.

Nagy K., Lévai L., Kovács B.: 2010. A szelénellátáshatása a kukoricaésnapraforgónövényekre.Növénytermelés, 59:61-84.

Puskás-Preszner A., Kovács B.: 2009. Molibdén-kezeléshatásaszabadföldikísérletben a növényifelvételreés a talajmolibdénfrakcióira.AgrártudományiKözlemények. Debrecen, 36:117-122.

Reilly C.: 1991. Metal contamination of food. Elsevier Science Publisher Ltd., 225-229.

Robb D. A., Pierpoint W. S.: (1983) Metals and micronutrients: uptake and utilization by plants. Academic Press. London.

Schulte E. E.: 2004. Soil and Applied Molybdenum. Understanding Plant Nutritients.Produced by Cooperative Extension Publications, University of Wisconsin -Extension.

Szalai I.: 2006. A növényekélete.Ahogyan ma látjuk I. NemzetiTankönyvkiadóZrt., Budapest.

Szebeni A., VargaI., Kovács B., TolvajGy. And Zalatnai A.: (2011) Microanalytical Determination of Trace Elements from Liver Biopsy Materials of Patients with Chronic Diffuse Liver Diseases with Different Ultrasound Attenuation, Liver Biopsy, Hirokazu Takahashi (Ed.). ISBN: 978-953-307-644-7, InTech. http://www.intechopen.com/articles/show/title/microanalytical-determination-of-trace-elements-from-liver-biopsy-materials-of-patients-with-chronic

Terry N., Zayed A. M., Desouza M. P., Tarun A. S.: (2000) Selenium in higher plants. Annual Review of Plant Physiology and Plant Molecular Biology. 51:401-432.

Tunçeli A, Türker A. R.: 2004. Solid-Phase Extraction and Spectrophotometric Determination of Molybdenum(VI) in Soil and Plant Samples as a Mo(V)-Thiocyanate Complex Microchim. Acta 144:69-74.

Wang Z. Y., Tang Y. L., Zhang F. S.: 1999. Effect of molybdenum on growth and nitrate reductase activity of winter wheat seedlings as influenced by temperature and nitrogen treatments. J. Plant Nutr. 22(2):387-395.

Williams R. J. P., da Silva J. J. R. F.: 2002. The involvement of molybdenum in life. Biochemical and Biophysical Research Communications, 292:293-299.

Yaneva I., Mack G., VunkovaRadeva R, Tischner R.: 1996. Changes in nitrate reductase activity and the protective effect of molybdenum during cold stress in winter wheat grown on acid soil. Journal of Pant Physiology 149:211-216.

Zaijun L., Yuling Y., Jian T., Jiaomai P.: 2005. 9-(2,4-dihydroxyphenyl)-2,3,7-trihydroxyl-6-fluorone as analytical reagent for spectrophotometric determination of molybdenum in plant tissues, Journal of Food Composition and Analysis, 16:561-569.

Zimmer W., Mendel R.: 1999. Molybdenum metabolism in plants. Plant Biol. 1:160-168.