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# NICKEL TRANSLOCATION FROM SEED DURING GERMINATION AND GROWTH OF YOUNG MAIZE PLANTS

ABSTRACT: Effect of different concentrations of nickel (0, $10^{-5}$, $10^{-4}$, $10^{-3}$ and $10^{-2}$ mol Ni/dm$^3$) present at the time of maize seed imbibition, on concentration, distribution and nickel accumulation coefficient in the root and the shoot, biological value of the seed and growth of young plants was investigated. It was found that during germination the nickel from the seed is intensively translocated to the root and shoot of young plants. With increase of applied concentrations of nickel, its concentration in the root and shoot increased as well. Nickel concentration and accumulation coefficient were higher in the root than in the shoot except at the highest applied concentration when the result was opposite. The highest applied concentration of nickel increased percentage of atypical seedlings and non-germinated seeds and decreased percentage of typical seedlings, germination energy and seed germination ability. Nickel implementation did not affect the growth and mass of the shoot. Root mass and length of the primary root decreased at the highest concentration of nickel, which led to change in shoot and root mass ratio.

Based on the obtained results it can be concluded that only the highest applied nickel concentration affected the biological value of the seed and the growth of young maize plants, regardless of its intensive accumulation in the root and the shoot, which indicates a significant tolerance of maize in initial phases of growth to presence of high nickel concentration. Intensive translocation of nickel during germination into newly formed organs points to its good mobility and potential possibility to enter the food chain from a contaminated seed.

KEY WORDS: maize, nickel imbibition of grain, germination, growth, translocation, distribution

## INTRODUCTION

Nickel (Ni) is widespread in the biosphere (Kabata-Pendias and Pendias, 2000). Discovery of nickel being the metal component of the enzyme urease (Dixon et al., 1975) and widespread in the plant world (Welch, 1981, Polacco, 1997), that bacteria require Ni for the synthesis of nickel hydrogenase(s), of nickel-containing carbon monoxide dehygrogenase(s), of methyl-CoM reductase and urease (Ankel-Fuchs and Thauer, 1988, Maier et al., 1990), intensified research related to the role of Ni in life processes of higher plants. In the 1970's but also earlier, numerous papers were published that prove favorable effect of Ni on plant growth and metabolism (Kastori and Petrović, 1976). However, it was only recently that its necessity for numerous organisms was confirmed, which led to including Ni in essential, biogene microelements (Eskew et al., 1983, Checkai et al., 1986, Brown et al., 1987, Ankel-Fuchs and Thaure, 1988, Marschner, 1995).

Higher concentrations of Ni, like other heavy metals, have toxic effect on plants (Asher 1991, Seregin and Kozhevnikova, 2006). Nickel became a significant pollutant. In crop plants there is much more concern about Ni contamination and toxicity. The application of sewage sludge which is often high in Ni (Brown et al., 1989) and certain phosphate fertilizers also may by important sources of Ni. Anthropogenic sources of Ni, industrial activity (metal processing operations, combustion of coal and oil) in particular, have resulted in significant increase of Ni content in soils (Kabata-Pendias and Pendias, 2000). In natural conditions, Ni toxicity appears in soils rich in Ni that originated on serpentine as parent rock. Nickel is readily mobile in the xylem and phloem (Kochian, 1991) and some plant species significantly translocate Ni into the seed (Petrović and Kastori, 1979, Petrović and Kastori, 1994). Many environmental stresses (e.g. mineral toxicities, soil acidity, nutrient deficiencies etc.) can directly or indirectly influence seed development including seed vigor and viability (Welch, 1999). The aim of this research was to investigate the influence of Ni contamination of the seed on biological properties of the seed, as well as translocation of Ni from the seed during germination into the root and the shoot of young maize plants.

## MATERIALS AND METHODS

The experiment was carried out on a maize hybrid NS 7016 seed. Nickel treatment was performed by soaking the maize seed in $NiSO_4$ solution. It was previously determined that at the temperature of 22°C a seed soaked during 24 h contains 37% of water and that after that period there is no statistically significant increase of water content in the seed. In order to germinate, the seed needs about 30% of water. In the course of the experiment, effect of five treatments was investigated, the seed was soaked in: 0 (control, deionized water), $10^{-5}$, $10^{-4}$, $10^{-3}$ and $10^{-2}$ mol Ni/dm$^3$. The experiment was set up in five replications. After the treatment, the seed was rinsed with deionized water. Than it was determined the biological value of the seed according to standard procedure published in Službeni list SFRJ 47/87 (Official Gazette of SFRY). It was investigated: germination energy, seed germination ability, representation of typical and atypical seedlings and the percentage of non-germinated seeds. After seven days it was measured the length of the shoot, of the primary root and the mesocotile root at 20 plants. It was determined the mass of the dry matter of the root and the shoot after drying the plant material to constant mass.

In the homogenized dry plant material of the shoot and the root it was determined the concentration of Ni and other mineral matter after digestion in cc $HNO_3$ + cc $H_2O_2$. Element concentration was determined using ICP. Based on results of chemical analysis there were determined the concentration, distribution and accumulation coefficient of Ni, as well as concentration of some macro- and microelements in the shoot and the root. Accumulation coefficient of Ni was calculated from the difference in Ni concentration between the treatment and the control.

The results were statistically processed by calculating the smallest significant difference between arithmetic means.

## RESULTS AND DISCUSSION

Results on Ni concentration in the seed indicate that Ni ions pass with, more or less, no obstruction through the testa and the semipermeable cell membrane of the seed and that in relatively short time they are in all parts of the seed, to which points their intensive accumulation in young plants (Tab. 1). Certain parts of the seed do not absorb water, which is probably why they do not absorb Ni at the same intensity. The germ absorbs water 2—3 times more intensively than the endosperm (Kastori, 1984), which probably contributed to significant Ni accumulation in the root and the shoot of young plants. At lower applied concentrations, the Ni concentration was higher in the root than in the shoot, while at the highest applied concentration it was vice versa. During germination, products of organic matter decomposition of the seed, as well as mineral matter, partly defund into the environment. Probably a part of Ni taken up by the seed got into the environment which enabled the root to uptake it, which reflected on its higher concentration at lower applied concentrations of Ni. With appearance of shoot and transpiration with it, the ascending transport of Ni was probably induced, especially at the highest applied concentration of Ni, which partially explains its higher concentration in the shoot than in the root. Accumulation coefficient which indicates the intensity of increase of Ni concentration with the increase of applied Ni concentration was higher in the shoot than in the root only when applying the highest concentration of Ni. According to Cieslinski et al. (1996) at a higher concentration of cadmium in the soil, out of the total uptaken quantity of cadmium a relatively smaller quantity is being translocated into above-ground organs than at lower concentrations, which they explain as a certain form of protection mechanism. On the contrary, our research results point to a relatively more intensive translocation of Ni to the shoot at a higher applied concentration. Intensive mobilization of Ni from the seed during germination and development of a young plant into newly-formed organs confirms earlier findings about good mobility of Ni in the plant phloem and xylem (Petrović and Kastori, 1979, Neumann and Chamel, 1986, Yang et al., 1996, Page and Feller, 2005).

Tab. 1 — Concentration, distribution and accumulation coefficient of Ni in young maize plants treated with Ni

| Organ | Treatment (mol Ni/dm$^3$) | | | | |
|---|---|---|---|---|---|
| | 0 | $10^{-5}$ | $10^{-4}$ | $10^{-3}$ | $10^{-2}$ |
| | Concentration (μg/g DM) | | | | |
| Shoot | 4.89 | 6.90** | 10.09** | 41.36** | 148.85** |
| Root | 4.17 | 46.35** | 52.77** | 70.74** | 110.29** |
| | Distribution (%) | | | | |
| Shoot | 46.88 | 10.43 | 12.71 | 32.04 | 54.83 |
| Root | 53.12 | 89.57 | 87.29 | 67.96 | 46.17 |
| | Accumulation coefficient | | | | |
| Shoot | 0.00 | 1.41 | 2.06 | 8.45** | 30.43** |
| Root | 0.00 | 11.11** | 12.65** | 16.69** | 26.45** |

Seed quality, vigor and viability are important characteristics influencing seedling establishment, crop growth, and productivity (TeKrony and Egli, 1991). Only the highest applied concentration of Ni affected biological value of the seed (Tab. 2), which confirms findings of Seregin and Kozhevnikova (2005) of great plant tolerance to high Ni concentration in the germination phase. Appliance of the highest Ni concentration has significantly decreased germination ability, germination energy and percentage of typical seedlings and increased percentage of atypical seedlings and non-germinated seed. Niethammer (1930) was one of the first to observe that Ni affects the seed germination. He established that lower concentrations (0.1% solutions) had stimulant effect, and higher (0.5 to 1.0%) had inhibitory effect. Among the investigated salts, the lowest toxicity was shown by $NiSO_4$ and the highest by Ni $(NO_3)_2$. Effect of Ni on germination thus depended on concentration and type of the salt. It was also observed that a young seed, with well preserved viability, reacts differently to Ni than an old seed having partially damaged germination ability. Namely, Ni concentrations that induced inhibition at a young, vital seed had no effect on an old seed or they even increased germination ability. Also later, a large number of authors investigated the influence of Ni on germination and seed germination ability, and the obtained results were often contradictory (Kastori and Petrović, 1976). Brown et al. (1987) state that imbibition with 1 μM $NiSO_4$ did not improve the percent germination of barley grain, suggesting that the availability of Ni for essential processes in the grain is not limiting germination. However, there is the possibility that Ni in the imbibition treatment was not available for uptake by the embryo. The mentioned authors have concluded, based on results of their own research which showed that plants grown with 1 mM Ni and nutrient solution produced grain with better germination, vigor and viability, and other authors' results, concluded that Ni is required for normal grain development, maturation and plant senescence. Brown et al. (1987) state that without adequate Ni supply, maternal barley plants developed nonviable grain that did not germinate upon imbibition. The exact metabolic role of Ni in seed germination is not known. It can be assumed that Ni affects the utilization of nitrogen stored in compounds in the seed during their catabolism upon germination because Ni is an essential component of the enzyme urease (Welch, 1999). Consequently a question arises at which extent is the nitrogen in the seed of small grains present in the form of ureides. It is obvious that the effect of Ni on seed germination depends on a larger number of factors: concentration, method of use and Ni enrichment of the seed, plant species, seed maturity etc. which could explain often contrary results about this element's effect on biological properties of the seed.

Tab. 2 — Effect of different Ni treatments on biological properties of maize seeds and seedlings

| Parameters | Treatment (mol Ni/dm$^3$) | | | | |
|---|---|---|---|---|---|
| | 0 | $10^{-5}$ | $10^{-4}$ | $10^{-3}$ | $10^{-2}$ |
| Typical seedlings (%) | 83.50 | 83.75 | 84.00 | 83.25 | 27.50** |
| Atypical seedlings (%) | 10.75 | 9.50 | 10.75 | 13.00 | 25.25** |
| Non-germinated seed (%) | 5.75 | 6.75 | 5.25 | 3.75 | 47.25** |
| Germination energy (%) | 81.75 | 82.25 | 82.50 | 80.75 | 20.00** |
| Germination ability (%) | 83.50 | 83.75 | 84.00 | 83.25 | 27.50** |

Usage of different Ni concentrations did not significantly affect the growth and mass of the shoot (Tab. 3). Higher Ni concentrations decreased growth of the primary root, root mass and increased shoot/root dry mass ratio, which suggests that the unfavorable effect of higher Ni concentrations affected more growth and development of the root system than of the shoot, even the Ni concentration being higher in the shoot than in the root at the highest applied Ni concentration. This contradicts the statement of Seregin et al. (2003) who point out that the growth of organs where Ni is more accumulated is being more intensively inhibited than the growth of organs where the accumulation of Ni is lower. It is reasonable to assume that the effect of toxic concentrations of an element on growth depends also on the length of the treatment. In our case it was short, which can probably partially explain no inhibition of the shoot growth. Significant decrease in the root growth at young maize plants in the presence of Ni was also established by Maksimović et al. (2007).

The critical toxic level of Ni in sensitive cultivated species is 10 μg/g of dry matter and 50 μg/g of dry matter in moderately sensitive species (Asher, 1991). Ni concentration in the shoot and the root when applying the highest investigated concentration of Ni was significantly above mentioned values, which confirm the findings of Seregin (2005) about increased tolerance of plants to surplus of Ni during germination. Toxic concentrations of Ni affect photosynthesis (Singh et al., 1989), as well as the function of cell membranes and the water regime (Llamas et al., 2008), they decrease the chlorophyll content, water potential and transpiration (Pandey and Sharma, 2002) and they affect the concentration and distribution of elements (Petrović et al., 1998, Ilin and Kastori, 1999). Kao and Lin (2005) state that the toxic concentration of Ni increases peroxidase activity, which causes inhibition of synthesis of cellulose and lignin, components of the cell wall, and thus decreases its thickness or even disables normal forming of the cell wall which prevents cell elongation. L'Huiller et al. (1996) state that unfavorable effect of higher Ni concentrations on the plant growth can, among other, be attributed to its influence on cell division. Findings of these authors can explain inhibition of the root growth and, partially, of the shoot at the highest concentration Ni treatment.

Tab. 3 — Effect of different Ni treatments on growth and dry mass of young maize plants

| Parameters | Treatment Ni (mol/dm$^3$) | | | | |
|---|---|---|---|---|---|
| | 0 | $10^{-5}$ | $10^{-4}$ | $10^{-3}$ | $10^{-2}$ |
| Shoot length (cm) | 6.89 | 6.42 | 6.75 | 7.08 | 6.19 |
| Primary root length (cm) | 11.84 | 10.88 | 11.63 | 10.29* | 10.42* |
| Mesocotile root length (cm) | 8.97 | 8.86 | 9.08 | 9.13 | 8.46 |
| Shoot dry mass (mg/plant) | 24.00 | 22.70 | 22.90 | 25.80 | 23.96 |
| Root dry mass (mg/plant) | 31.70 | 30.10 | 29.70 | 32.10 | 26.63 |
| Shoot dry mass / root dry mass | 0.75 | 0.75 | 0.77 | 0.80 | 0.90 |

Translocation of some macro- and micro elements was investigated, from the seed to the shoot and the root, during the growth of young plants at a non-treated seed (Tab. 4). Translocation of the investigated elements from the seed during germination into the shoot and the root varied. In the shoots, P, K and B accumulated more intensively, S, Mg, Ca, Fe, Mn, Mo and Al in the root, while accumulation of Zn and Cu was approximately even. Generally, it can be said that translocation of mineral matter during maize seed germination is more intense into the root than into the shoot.

Tab. 4 — Translocation of macro- and microelements during maize seed germination

| Organ | Macronutrients (mg/100 g DM) | | | | |
|---|---|---|---|---|---|
| | **P** | **K** | **S** | **Mg** | **Ca** |
| Shoot | 672.25 | 1062.50 | 47.22 | 159.10 | 176.50 |
| Root | 451.31 | 885.28 | 112.83 | 245.23 | 987.95 |

| | Micronutrients (µg/g DM) | | | | | | |
|---|---|---|---|---|---|---|---|
| | **Fe** | **Zn** | **Cu** | **Mn** | **Mo** | **Al** | **B** |
| Shoot | 29.33 | 7.27 | 0.50 | 2.54 | 0.05 | 9.19 | 1.05 |
| Root | 247.30 | 7.24 | 0.46 | 6.84 | 0.10 | 99.49 | 0.64 |

Obtained results show that due to intensive mobilization of Ni in the seed during germination and its translocation into the root and the shoot, there is a great possibility of Ni entering the food chain from the contaminated seed, which especially has to be taken into account when producing organic food.

## REFERENCES

Ankel-Fuchs, D., Thauer, R. K. (1988): *Nickel in Biology: Nickel as an Essential Trace Element*. In: Jack, R., Lancaster, Jr. (ed.) *The Bioneorganic Chemistry of Nickel*. VCH Publishers, New York.

Asher, C. J. (1991): *Beneficial elements, functional nutrients, and possible new essential elements*. In: Mortvedt, J. J., Cox, F. R., Shuman, L. M., Welch, R. M. (eds.) *Micronutrients in Agriculture* 2nd edition, Soil Sci. Soc. Amer. Book Series No. 4, Madison, WI, USA, 703—723.

Brown, P. H., Dunemann, L., Schulz, R., Marschner, H. (1989): *Influence of redox potential and plant species on the uptake of nickel and cadmium from soils*. Z. Pflanzenähr. Bodenk. 152: 85—91.

Brown, P. H., Welch, R. M., Cary, E. E. (1987): *Nickel: a micronutrient essential for higher plants*. Plant Physiol. 85: 801—803.

Checkai, R. T., Norvell, R. M., Welch, R. M. (1986): *Investigation of nickel essentiality in higher plants using a recirculating resin-buffered hydroponic system*. Agron. Abst. 195.

Cieslinski, G., Neilsen, G. H., Hogue, E. J. (1996): *Effect od soil cadmium application and pH on growth and cadmium in roots, leaves and fruits of strawberry plants (Fragaria x ananassa Duch)*. Plant Soil 180: 267—276.

Dixon, N. E., Evans, F. J., Hanus, F. J. (1975): *Jack-Bean urease (E.C.3.5.1.5.3). A metalloenzyme. A simple biological role for nickel*. J. Am. Chem. Soc. 97: 4131—4133.

Eskew, D. L., Welch, R. M., Cary, E. E. (1983): *Nickel: an essential micronutrient for legumes and possibly all higher plants*. Science 222: 621—623.

Ilin, S., Kastori, R. (1999): *Uticaj nikla na sadržaj neophodnih makroelemenata u mladim biljkama pšenice i kukuruza*. Zbornik radova Naučnog instituta za ratarstvo i povrtarstvo, Novi Sad. 31:603—610.

Kabata-Pendias, A., Pendias, H. (2000): *Trace Elements in Soils and Plants*. CRC Press, Boca Raton.

Kao, C. H., Lin, C. (2005): *Nickel toxicity of rice seedlings: Cell wall peroxidase, lignin and* $NiSO_4$*-inhibited root growth*. Crop, Environment & Bioinformatics. 2: 131—136.

Kastori, R. (1984): *Fiziologija semena*. Matica srpska, Novi Sad.

Kastori, R., Petrović, N. (1976): *Uloga nikla u životnim procesima biljaka*. Savremena poljoprivreda 9—10: 73—83.

Kochian, L. V. (1991): *Mechanism of micronutrient uptake and translocation i plants*. In: Mortvedt, J. J. (ed.) Soil Sci. Soc. Am. Book Series No. 4, Medison, WI, 229—296.

L'Huillier, L., D'Auzac. J., Durand, M., Michaud-Ferriere, N. (1996): *Nickel effect on two maize (Zea mays) cultivars: growth, structure, Ni concentration, and localization*. Can. J. Bot. 74: 1547—1554.

Llamas, A., Ullrich, C. I., Sanz, A. (2008): $Ni^{2+}$ *toxicity in rice: Effect on membrane functionality and plant water content*. Plant Physiol. Biochem. 46: 905—910.

Maier, R. J., Phil, T. D., Stults, L., Sray, W. (1990): *Nickel accumulation and storage in Bradyrhizobium japonicum*. Appl. Environ. Microbiol. 56: 1905—1911.

Maksimović, I., Kastori, R., Krstić, L., Luković, J. (2007): *Steady presence of cadmium and nickel affects root anatomy, accumulation and distribution of essential ions in maize seedlings*. Biol. Plant. 51: 589—592.

Marschner, H. (1995): *Mineral Nutrition of Higher Pants*. Academic Press, London.

Neumann, P. M., Chamel, A. (1986): *Comparative phloem mobility of nickel in no senecent plants*. Plant Physiol. 81: 689—691.

Niethammer, A. (1930): *Landwirtschaftlich-biologische Studien mit Nickel- und Cyanverbindungen*. Arch. Pflanzenbau 4: 607—634.

Padney, N., Sharma, Ch. P. (2002): *Effect of heavy metals $Co^{2+}$, $Ni^{2+}$ and $Cd^{2+}$ on growth and metabolism of cabbage*. Plant Sci. 163: 753—758.

Page, V., Feller, U. (2005): *Selective transport of zinc, manganese, nickel, cobalt and cadmium in the root system and transfer to the leaves in young wheat plants*. Ann. Bot. 96:425—434.

Petrović, N., Kastori, R. (1979): *Uptake and translocation of $^{63}Ni$ in young wheat plants*. Proceedings of the First International Symposium on Plant Nutrition. Mineral Nutrition of Plants. Vol. II, Varna, Bulgaria, 324—327.

Petrović, N., Kastori, R. (1994): *Uptake, distribution and translocation of nickel ($^{63}Ni$) in wheat*. J. Food Physics 1:71—73.

Petrović, N., Kastori, R., Sekulić, P., Kevrešan, S. (1998): *Effect of Cr and Ni on the accumulation and distribution of mineral elements in young sugar beet plants*. Zemljište i biljka. 47: 17—29.

Polacco, J. C. (1977): *Nitrogen metabolism in soybean tissue culture*. Plant Phyisiol. 59: 827—830.

Seregin, I. V., Kozhevnikova, A. D. (2005): *Distribution of cadmium, lead, nickel, and strontium in imbibing maize caryopses*. Russ J. Plant Physiol. 565—569.

Seregin, I. V., Kozhevnikova, A. D. (2006): *Physiological role of nickel and its toxic effect on higher plants*. Russ. J. Plant Physiol. 53: 257—277.

Seregin, I. V., Kozhevnikova, A. D., Kazyumina, E. M., Ivanov, V. B. (2003): *Nickel toxicity and distribution in maize roots*. Russ. J. Plant Physiol. 711—718.

Singh, D. P., Punam, K., Bisen, P. S. (1989): *Effect of $Ni^{2+}$, $Hg^{2+}$ and $Cu^{2+}$ on growth, oxygen evolution and photosynthetic electron transport on Cylindrospermium IU 942*. Plant Physiol. 134: 406—421.

TeKrony, D. M., Egli, D. B. (1991): *Relationship of seed vigor to crop yield: A review*. Crop Sci. 31: 816—822.

Welch, R. M. (1981): *The biological significance of nickel*. J. Plant Nutr. 31: 345—356.

Welch, M. R. (1999): *Importance of Seed Mineral Nutrient Reserves in Crop Growth and Development*. In: Rengel, Z. (ed.) Mineral Nutrition of Crops- Fundamental Mechanisms and Implications. Food Products Press, New York London Oxford. 205—226.

Yang, X., Baligar, V. C., Martens, D. C., Clark, R. B. (1996): *II Nickel effects on influx and transport of mineral nutrients in four plant species*. J. Plant Nutr. 19: 265—279.

# ТРАНСЛОКАЦИЈА НИКЛА ИЗ СЕМЕНА У ТОКУ КЛИЈАЊА И РАСТ МЛАДИХ БИЉАКА КУКУРУЗА

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## Резиме

Испитивано је дејство бубрења семена кукуруза у растворима различитих концентрација никла (0, $10^{-5}$, $10^{-4}$, $10^{-3}$ и $10^{-2}$ mol Ni/dm$^3$) на садржај, дистрибуцију и коефицијент акумулације никла у корену и изданку, биолошку вредност семена и раст младих биљака. Утврђено је да се никал из семена у току клијања интензивно транслоцира у корен и изданак младих биљака. Са повећањем примењене концентрације никла значајно се повећао његов садржај у изданку и корену. Садржај никла и коефицијент акумулације били су већи у корену него у изданку, изузев код највеће примењене концентрације, где је било обрнуто. Највећа примењена концентрација никла повећала је удео атипичних поника и непроклијалог семена и смањила удео типичних поника, енергију клијања и клијавост семена. На раст и масу изданка примена никла није утицала. Маса корена и дужина примарног корена се смањила при употреби највеће концентрације никла, што је довело до промене односа масе изданка и корена.

На основу добијених резултата може се закључити да је само највећа примењена концентрација никла утицала на билошку вредност семена и раст младих биљака кукуруза и поред његовог интензивног накупљања у корену и изданку, што указује на значајну толерантност кукуруза у почетним фазама раста на присуство високе концентрације никла. Интензивна транслокација никла у току клијања у новоформиране органе указује на његову добру покретљивост и потенцијалну могућност да из контаминираног семена улази у ланац исхране.