

Helena Kubicka\*, Tomasz Kubel\*

THE INFLUENCE OF CHOSEN BIOGENIC ELEMENTS  
ON GROWTH OF RYE INBRED LINES

DZIAŁANIE WYBRANYCH PIERWIASTKÓW BIOGENNYCH  
NA WZROST LINII WSOBNYCH ŻYTA (*SECALE CEREALE* L.)

**Słowa kluczowe:** cynk, jony, linie wsobne, mangan, żyto.

**Key words:** inbred lines, ions, manganese, rye, zinc.

*Cynk i mangan to biogenne pierwiastki występujące powszechnie w środowisku. Ich niedobór niekorzystnie wpływa na wzrost i rozwój roślin. W niniejszej pracy badano wrażliwość siewek pięciu linii wsobnych żyta na zwiększoną zawartość jonów cynku i manganu w podłożu. Użyte wyniki wskazują, że stężenie  $10^{-1}M$  obu pierwiastków powoduje znaczne zahamowanie wzrostu roślin, zarówno korzeni, jak i części nadziemnych. Natomiast niższe stężenie  $10^{-2}M$  jonów cynku lub manganu stymuluje wzrost u niektórych linii. Najwyższa tolerancja obu pierwiastków cechowała linię 154. Oceniane linie wsobne żyta zróżnicowanie reagowały na nadmiar jonów cynku lub manganu w podłożu. Bardziej wrażliwe na działanie obu pierwiastków, aniżeli korzenie, były części nadziemne roślin. Zastosowany w doświadczeniu kwas askorbinowy w kombinacji z cynkiem lub manganem w stężeniu  $10^{-1}M$  powodował zmniejszenie stresu spowodowanego nadmiarem tych pierwiastków w podłożu u niektórych linii.*

## 1. INTRODUCTION

Biogenic elements such as manganese and zinc are parts of many enzymes and are necessary for development and growth of plants. They are abundant in soil, and their main natural source is bedrock [CICAD 2004, Broadley et al. 2007].

Levels of anthropogenic zinc and manganese in environment have been rising along with increasing industrialization. In the nineties emission levels of zinc were close to 2700

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\* **Doc. dr hab. Helena Kubicka, mgr Tomasz Kubel – Ogród Botaniczny – Centrum Zachowania Różnorodności Biologicznej PAN, ul. Prawdziwka 2, 02-973 Warszawa; e-mail: helenakubicka@wp.pl**

kilotons \* year<sup>-1</sup> while two millennia ago it was only 10 kilotons \* year<sup>-1</sup>. The main areas of zinc contamination are around zinc mining sites [Pugh et al. 2002]. Zinc is also released into environment by burning fossil fuels and tyres, using pesticides as well as dumping sewage sludge and metals coated with anticorrosive zinc layers [Broadley et al. 2007].

The main sources of atmospheric manganese contamination are ore mills and burning fossil fuels [Hagelstein 2009]. Manganese soil contamination is mainly caused by penetration of manganese compounds from waste yards and through contaminated rainfalls [CI-CAD 2004]. A source of manganese contamination is also the gasoline additive metylocyclopentadienyl manganese tricarbonyl (MMT) that is used in some countries to increase fuel octane rating [Kitazawa et al. 2002].

Plant tolerance to deficiency of biogenic compounds differs not only between species but even between lines. One of zinc hyperaccumulators are plants from the *Thlaspi* genus and an example of a manganese hyperaccumulator is *Phytolacca americana*. Inhibition of growth and disturbance in the mineral uptake and distribution are the main symptoms of excess Mn and Zn in soil [Lidon 2001, Broadley et al. 2007, Mina et al. 2007].

The objective of this research was to evaluate the influence of zinc and manganese in the growth medium on growth of rye seedlings.

## 2. MATERIALS AND METHODS

Five, genetically different inbred lines (L176, 154, L29, CH7/99, L230, M353) of rye were used in the experiment [Kubicka et al. 2006]. Four-day-old rye seedlings were transferred to the Hoagland's medium with addition of zinc or manganese: the medium without zinc or manganese addition was used as the control: 10<sup>-1</sup>M and 10<sup>-2</sup>M Zn or Mn. Two combinations were prepared with addition of ascorbic acid (10<sup>-2</sup>M) and zinc or manganese at a concentration 10<sup>-1</sup>M. The length of roots and shoots was measured after seven and fourteen days. The tolerance index was calculated according to the equation:

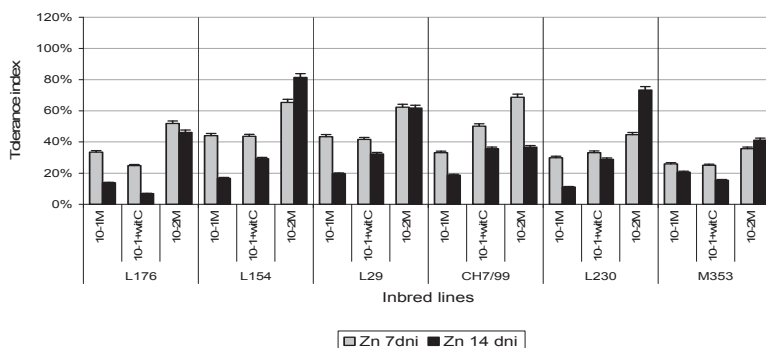
$$\text{Tolerance index} = \frac{\text{value before metal treatment}}{\text{value after metal treatment}} \times 100\%$$

## 3. RESULTS AND DISCUSSION

Although Zn and Mn are biogenic elements, exposition of rye inbred lines seedlings to excess Zn or Mn resulted mainly in inhibition of seedlings growth (Fig. 1–4).

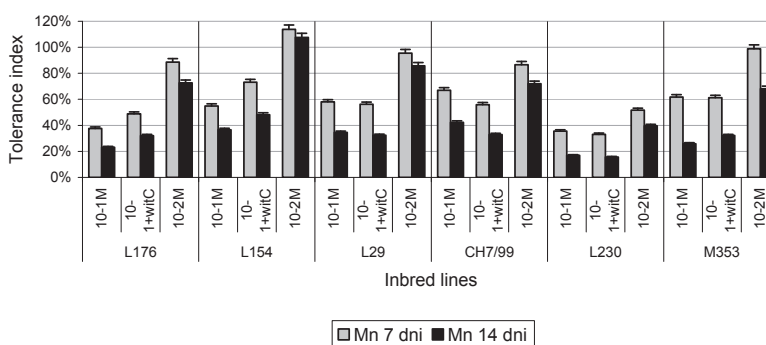
The shoots of tested rye inbred lines were more susceptible to Zn or Mn than the roots. Distinguished growth inhibition was observed especially on the growth mediums with higher metal concentration (10<sup>-1</sup>M). The plants exposed to lower metal concentration (10<sup>-2</sup>M) developed better. Manganese (10<sup>-2</sup>M) stimulated growth of 7 and 14 days old seedlings of inbred

line 154. Lines L29 and M353 showed high tolerance to manganese at a  $10^{-2}\text{M}$  concentration – 7 days old seedlings were almost of the same length as the control seedlings. The biggest inhibition of growth, on the medium with excess Mn and Zn, was observed in line L230 (Fig. 1–2). Shoots of tested lines were more sensitive to Zn: they grew much shorter than in the lines with Mn added to the medium. Zinc concentration of  $10^{-2}\text{M}$  had less influence on rye seedling growth. Lines 154, L230 and L29 had the highest tolerance index, ranging from 60% (line L29) to 80% (line 154). Some lines growing on the medium with ascorbic acid showed higher tolerance to Zn and Mn. Shoot increment was noted in lines: CH7/99 and L230 (7 days, Zn), L154 and L176 (7 and 14 days, Mn).



**Fig. 1.** The influence of zinc ions on the length of shoots of 7 and 14 day seedlings of rye inbred lines in comparison to control [%]

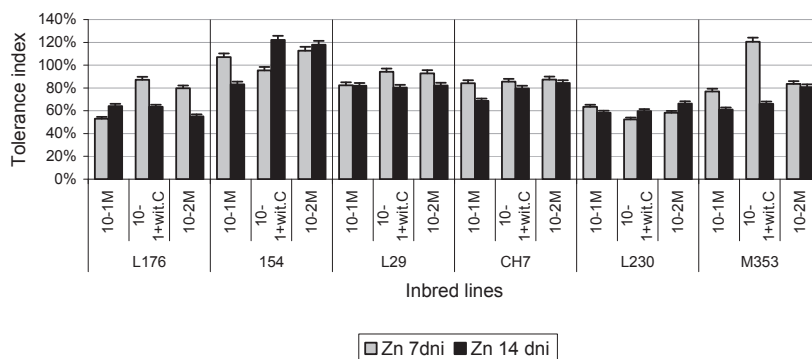
**Rys. 1.** Wpływ jonów cynku na długość kielków 7 i 14 dniowych siewek linii wsobnych żyta w porównaniu do kontroli [%]



**Fig. 2.** The influence of manganese ions on the length of shoots of 7 and 14 day seedlings of rye inbred lines in comparison to control [%]

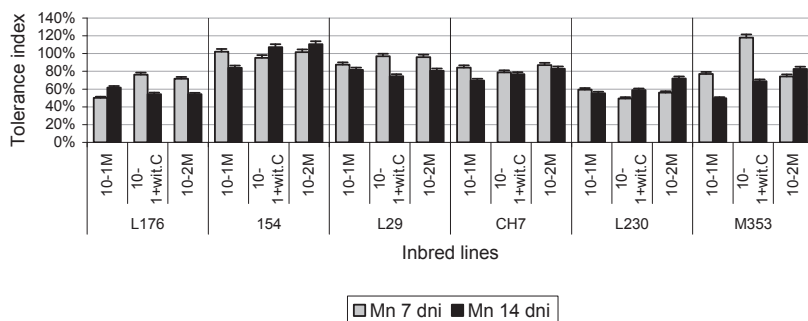
**Rys. 2.** Wpływ jonów manganu na długość kielków 7 i 14 dniowych siewek linii wsobnych żyta w porównaniu do kontroli [%]

The excess of both biogenic molecules had less influence on growth of roots than shoots (Fig. 3–4). The biggest growth inhibition for Zn and Mn was observed at a concentration of  $10^{-1}M$ . The rye inbred lines differed in their reaction to tested metals. Lines L176 and L230 appeared to be most susceptible and line 154 most tolerant. The root tolerance index of inbred line 154 exceeded 100%: at a concentration of  $10^{-2}M$  Zn or Mn, after 7 days at a concentration  $10^{-1}M$  (Zn or Mn) and after 14 days at a concentration  $10^{-1}M$  with addition of ascorbic acid. After 7 days of observation ascorbic acid lowered the stress caused by excess Zn or Mn in lines M353, L176 and 29.



**Fig. 3.** The influence of zinc ions on the length of roots of 7 and 14 day seedlings of rye inbred lines in comparison to control [%]

**Rys. 3.** Wpływ jonów cynku na długość korzeni 7 i 14 dniowych siewek linii wsobnych żyta w porównaniu do kontroli [%]



**Fig. 4.** The influence of manganese ions on the length of roots of 7 and 14 day seedlings of rye inbred lines in comparison to control [%]

**Rys. 4.** Wpływ jonów manganu na długość korzeni 7 i 14 dniowych siewek linii wsobnych żyta w porównaniu do kontroli [%]

The results show that line 154 is the most tolerant to the excess Zn and Mn. However, it has a higher tolerance to Mn than Zn. Depending on plant organs, the difference in tolerance was observed. Shoots were more susceptible to excess Zn or Mn than roots, though Zn had a greater inhibiting effect. The roots of line 154 expressed especially high tolerance to Zn ions. The tolerance index for the lower concentration of Zn reached 120% and was greater than 100% for Mn. Growth of line 154 roots was stimulated by the lower concentration ( $10^{-2}$ M) of manganese or zinc and shoot growth was stimulated by the lower concentration of manganese. This can be caused by an important role of Mn ions in the PS II complex, an increased efficiency of photosynthesis and better distribution of minerals. According to Monnet et al. [2001] an elevated concentration of manganese caused a disorder in the uptake of important minerals – including iron, which is necessary for plant growth and development. It is important to notice that the tolerance of roots is higher than shoots. This is in accordance with suggestions of Lidon [2001] about storing manganese in different plant organs. Lower Zn tolerance is probably caused by oxidative stress induction and mineral uptake disorders of Mn, Mg, or Fe, what is proved by Broadley et al. [2007].

To test whether Zn and Mn do induce oxidative stress, an experiment was conducted where the growth medium was enriched with ascorbic acid and Zn or Mn at a concentration of  $10^{-1}$ M. It is well known that ascorbic acid is an important dietary ingredient. Plants are rich in this compound and it is especially abundant in chloroplasts. It mainly works as an antioxidant [Smirnoff 2000]. Because heavy metals induce oxidative stress, the higher tolerance index was expected when plants grew on the medium enriched with ascorbic acid. The results show slightly better growth of some lines on the medium with ascorbic acid. It is true for both 7 and 14 day-old seedlings.

Tested rye inbred lines differed in their reaction to excess Zn or Mn in the growth medium. Line 154 showed to have the highest tolerance among all tested lines. The most susceptible was line L230.

#### 4. CONCLUSIONS

1. Inbred lines of rye responded differently to excess of Zn or Mn in the medium.
2. Ascorbic acid in combination with Zn or Mn at a concentration  $10^{-1}$ M in the medium reduced the stress caused by the excess of analyzed heavy metal ions.

#### REFERENCES

BROADLEY M.R., WHITE P.J., HAMMOND J.P., ZELKO I., LUX A. 2007. Zinc in plants. *New Phytol.* 173: 677–702.

**World Health Organization, Manganese and its Compounds: Environmental Aspects.** 2004. CICAD.

- HAGELSTEIN K. 2009. Globally sustainable manganese metal production and use. *Environ. Manage.* 90: 3736–3740.
- KITAZAWA M., WAGNER J.R., KIRBY M.L., ANANTHARAM V., KANTHASAMY A.G. 2002. Oxidative Stress and Mitochondrial-Mediated Apoptosis in Dopaminergic Cells Exposed to Methylcyclopentadienyl Manganese Tricarbonyl. *JPET.* 302: 26–35.
- LIDON F.C. 2001. Tolerance of rice to excess manganese in the early stages of vegetative growth. Characterisation of manganese accumulation. *J. Plant Physiol.* 158: 1341–1348.
- MINA Y., BOQINGB T., MEIZHENC T., AOYAMA I. 2007. Accumulation and uptake of manganese in a hyperaccumulator *Phytolacca americana*. *Minerals Engineering* 20: 188–190.
- MONNET F., VAILLANT N., VERNAY P., COUDRET A., SALLANON H., HITMI A. 2001. Relationship between PSII activity, CO<sub>2</sub> fixation, and Zn, Mn and Mg contents of *Lolium perenne* under zinc stress. *J. Plant Physiol.* 158: 1137–1144.
- PUGH R.E., DICK D.G., FREDEEN A.L. 2002. Heavy Metal (Pb, Zn, Cd, Fe, and Cu) Contents of Plant Foliage near the Anvil Range Lead/Zinc Mine, Faro, Yukon Territory. *Ecotoxicol. Environ. Saf.* 52: 273–279.
- SMIRNOFF N. 2000. Ascorbate biosynthesis and function in photoprotection. *Phil. Trans. R. Soc. Lond. B.* 355:1455–1464.