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# TOMATO REACTION ON EXCESSIVE MANGANESE NUTRITION

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## Abstract

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Manganese (Mn) is a heavy metal essential for plant growth. Most of the groundwater to a depth of 100 m used to nutrient solution prepare contain up to 0.5 mg Mn $dm^{-3}$. However, sometimes water may contain up to 10 mg Mn·$dm^{-3}$. About 5% of waters contain from 1 to 4.5 mg Mn·$dm^{-3}$. The optimal manganese concentration for tomato cultivation ranges between 0.3 to 0.6 mg·$dm^{-3}$ of nutrient solution The aim of the conducted study was to assess the effect of Mn stress (concentration in nutrient solution at 2.4 to 24.0 mg·$dm^{-3}$) on plant growth, nutrient uptake and yielding. The model plant was tomato (*Lycopersicon esculentum* Mill.cv. 'Alboney $F_1$'). Mn stress did not significantly affect the number leaves, flowers per inflorescence and visible clusters on the plants. Increasing Mn stress had a significant effect on the fresh weight of leaves, stalks, shoots and fruits. The biomass production was reduced by about 17.6%. Mn application resulted in the deterioration of qualitative and quantitative yield of tomato (total yield reduction by about 25.7%). In spite of the significant decrease of biomass production in all the studied combinations there were no other visual symptoms of Mn toxicity to plants. Mn stress significantly affected nutrient uptake, both macro- like micronutrients, especially on increasing content of microelement ion in plants parts. The highest content of Mn in plants was found in leaves, later in stalks and the lowest in case of fruits. It was found almost complete correlation between increasing Mn nutrition and between Mn content in leaves, stalks, fruits and whole plants (R. Pearson ranged from 0.94 to 0.99). Almost complete correlations were also found between Mn content in leaves in relation to fruits, stalks: leaves as well as a very high correlation for Mn concentration in stalks: fruits.

*Key words*: manganese, toxicity, growth, stress, nutrient uptake

## Introduction

In spite of its high atomic mass (54.93) manganese (Mn) is a metallic microelement. This nutrient influences many physiological process in plants, such as building blocks of photosynthetic proteins, lignins, flavonoids as well as several enzymes, ie. Mn-catalase, dehydrogenases, decarboxylases, hydroxylases and transferases (Ducic and Polle, 2005; Humphries et al., 2007; Lidon et al., 2004). Excessive Mn nutrition may damage the photosynthetic apparatus, significantly reducing chlorophyll content in leaves (Lee et al., 2011; Millaleo et al., 2010). Mn also affects absorption and translocation of other nutrients or elements (P, K, Ca, Mg, Fe, Zn, Cu, Si), as well as causes oxidative stress (Ducic and Polle, 2005; Foy et al., 1981; Galvez et al., 1989; Horst and Marschner, 1978; Kasraei et al., 1996; Lei et al., 2007; Lee et al., 2011; Shenker et al., 2004). Mn could also improve the phosphorus uptake (Clark, 1982; Galvez et al., 1989). The ion of Mn (II) is 0.075 nm in diameter and ranks between Fe (0.065 nm) and Ca (0.099 nm), thus at a quantitative predominance of Mn ions competence or substitution of the above mentioned ions occurs (Marschner, 1998). Lee et al. (2011) reported that the total content of free amino acids is reduced with an increase in Mn nutrition. A significant effect on the toxicity threshold on Mn was found for genotype, the environment, temperature, as well as nutrient status, e.g. for Ca, Mg or Fe (El-Jaoual and Cox, 1998; Alam et al., 2001; Le Bot et al., 1990a). Excessive Mn nutrition may lead to reduced plant yielding (Savvas et al., 2009). The optimal Mn concentration for tomato cultivation ranges between 0.3 and 0.6 mg·$dm^{-3}$ of nutrient solution (Kleiber, 2014).

In view of the above, the present experiment was designed to study the effects of Mn stress within a wide range of concentrations applied to the root zone (from 2.4 to 24.0 mg·$dm^{-3}$

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of nutrient solution) on growth, yielding and uptake of selected nutrients in tomato.

## Material and Methods

The experiment was conducted in a specialist culture greenhouse equipped with the modern climate control system. Climate parameters (temperature, $CO_2$ content, % RH) were recorded using the Synopta software. The effect of nutrient solution (NS) with varied concentrations of Mn on growth, development, nutrient uptake and yielding was studied on tomato (*Lycopersicon esculentum* Mill.cv. 'Alboney $F_1$') grown in rockwool. Plants were grown to have 1 cluster. Three mats (for each combination analysis) of 100x15x7.5 cm were placed in growing containers. Two plants were planted in each mat. Plants were grown in the hydroponics stagnant system, complementing every day to level of ½ the height of the mat. Each Mn-treatment was replicated tree times and comprised two plants. Seeds were sown to cultivation plugs in the 1$^{st}$ decade of March. After 3 weeks seedlings were transplanted to rockwool cubes (10×10×10 cm). Plants were transplanted to permanent beds in the middle of April. The experiment was concluded at the end of June.

Plants were grown in rockwool using a NS with the following Mn contents (in mg·dm$^{-3}$): 2.4, 4.8, 9.6, 14.4, 19.2, 24.0 denoted as Mn-2.4; Mn-4.8; Mn-9.6; Mn-14.4; Mn-19.2; Mn-24.0. Manganese sulfate ($MnSO_4 \cdot H_2O$, 32.3% Mn) was the source of Mn in the tested combinations. Plants, following their transplantation to their permanent site, were fertigated with a standard NS containing (in mg·dm$^{-3}$): N-$NH_4$ 2.2; N-$NO_3$ 230; P 50, K 430, Ca 145, Mg 65, Cl 35, S-$SO_4$ 120, Fe 2.48, Zn 0.50, Cu 0.07, pH 5.50, EC 3.00 mS·cm$^{-1}$. Fruit setting was facilitated by vibration of the trusses at approximately once a week. Fruits were harvested once at end the of the vegetation experiment. Fruits were divided into grades according to their diameter (in cm): I >10.2, II 10.1-8.2, III 8.1-6.7, IV 6.6-5.6, V 5.6-4.7 and VI<4.7.

In the end of vegetation experiments samples of stalks, leaves and fruits were collected in order to determine macro- and microelement contents. Plant material was dried at 45–50ºC and then ground. For assays of total N, P, K, Ca and Mg the plant material was mineralized in concentrated sulfuric acid. After mineralization of the plant samples, chemical analyses were performed using the following methods: total N according to Kjeldahl in a Parnas-Wagner distillation apparatus, P – by colorimetry with ammonium molybdate, and K, Ca, Mg by ASA (in a Carl Zeiss Jena apparatus). In the determinations of total Fe, Mn, Zn and Cu the plant material was mineralized in a mixture of dioxonitric and tetraoxochloric acids (3:1 v/v). After mineralization Fe, Mn, Zn and Cu were determined according to ASA.

Results of plant yielding and chemical analyses of all the plant parts for their contents of macro- and microelements were analyzed statistically using the Duncan test, with inference at the significance level $\alpha = 0.05$ (values described with identical letters do not differ significantly). Pearson correlations were also determined.

## Results and Discussion

Mn stress did not significantly affect the numbers of leaves, flowers per inflorescence and visible clusters on the plants (Figure 1a).

In spite of a lack of differences in the number of leaves per 1 plant a significant effect of Mn on the fresh weight of leaves, stalks, shoots and fruits was observed (Figure 1b).

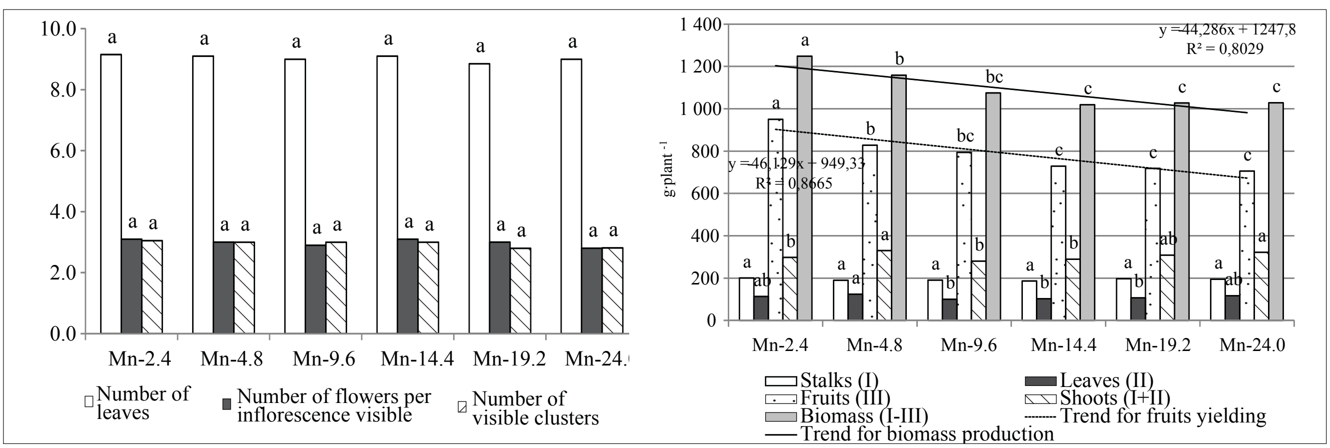


**Fig. 1. The effect of Mn stress on: (a) plant growth (numbers per 1 plant) and (b) fresh weight of stalks, leaves, fruits, shoots and biomass production of tomato (in g·plant$^{-1}$)**

The biomass production was reduced by about 17.6%, while fruit production by about 25.7% when comparing Mn-24.0 with Mn 2.4. In spite of the significant decrease of biomass production in all the studied combinations there were no symptoms of Mn toxicity on the plants. Savvas et al. (2009) claimed that both the excessively high and low Mn levels in the root environment of tomato reduced the yield by restricting the number of fruits per plant, but had no effect on the mean fruit weight. The limiting effect of Mn deficit or excessive Mn concentration in the root zone on tomato yielding was confirmed by Kleiber (2014). In our study fruit production was found to decrease – the most possible reason of that fact could be a reduction of the photosynthetic apparatus (weight of leaves) with increasing Mn stress. Savvas et al. (2009) reported that decreased yielding could be the reason for the reduced leaf area indicated by the significantly lower dry leaf weight per plant in comparison with plants grown with a standard Mn supply.

Increasing Mn stress significantly modified the fruit production of different classes, as it significantly decreased production of fruit with the highest diameter (Figure 2). Generally a significant increase was found in the production of fruit with smaller diameters (mainly classes IV and V) with an increase in Mn doses.

Excessive/toxic manganese concentration applied in NS significantly changed the nutrient uptake by the aboveground parts of plants: stalks, leaves and fruits. Details are shown in Figures 3 and 4.

In the literature there are no data on the nutrient content in the stems of tomato, while available sources cite contents for leaves or fruits (Tables 1 and 2). The content of N in leaves

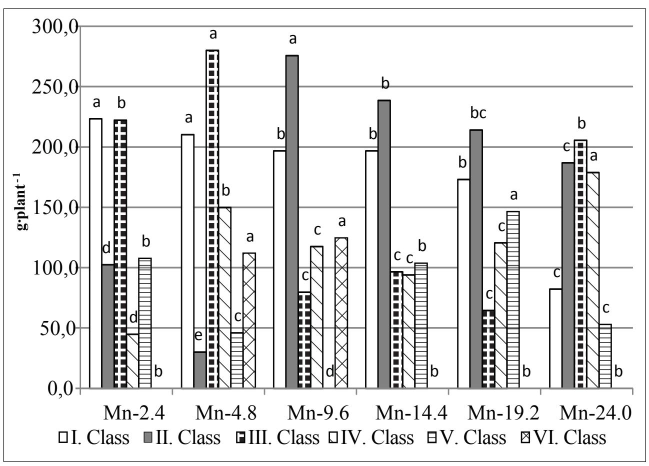


**Fig. 2. The effect of Mn stress on different class fruit production**

determined in our studies ranged from 5.11 to 8.05% N. Most other researchers reported lower contents of that nutrient (Table 1). In our study the content of P was comparable to that cited in literature. A higher content of that nutrient was found only by Kowalska (2004), Pawlińska and Komosa (2006) and (Plank, 1999). The content of K in tomato leaves was lower than that found by most other authors. A majority sources in literature showed a higher content of Ca in leaves (Table 1) – except Agric. Service (2001), Campbel (2000) and Plank (1999). Determined contents of Mg were similar to found in literature (Table 1).

The determined N content in fruits ranged from 4.76 to 6.09% in d.m. and varied depending on Mn concentration. The N contents recorded by other authors were lower (Table 1). In our study P content in fruits ranged from 0.54 to 0.59%. Most of the cited authors showed a similar range of contents. The content of K was lower than that determined by other researchers (except for Olaniyi et al., 2010). Important factors influencing K content include growing medium (Kleiber et al., 2012) and nutrition level (Jarosz and Dzida, 2011; Fanasca et al., 2006). Data in literature showed a higher fruit content of Ca (Kleiber et al., 2012, Fanasca et al., 2006; Olaniyi et al., 2010) and Mg (Kleiber et al., 2012, Olaniyi et al., 2010).

In the case of microelements most cited sources showed similar contents of Fe, Zn and Cu in leaves to those determined in our study. A drastically higher content of Cu was determined by Kowalska (2004). Mn-content up to Mn-19.2 generally was similar to that shown by other authors. Different sources gave highly varied Fe contents in tomato fruits. Important factors influencing contents of that nutrient in fruit include the growing medium (Kleiber et al., 2012; Premuzic et al., 1998) and nutrition (Gad and Kandil, 2010). The determined Fe content was similar to that presented by Gad and Kandil (2010), but it was drastically higher than cited by Kleiber et al. (2012) and Olaniyi et al. (2010). Increasing Mn concentration positively influenced the content of that microelement in fruits. The determined contents were absolutely higher than cited by other authors. A natural defence process in plants against Mn-stress is to accumulate it in physiologically relatively inactive cell sites (Horst, 1988).

Increasing Mn stress had a negative effect on the Zn content in fruit. Similarly as in the case of Fe, important factors affecting Zn contents include nutrition (Gad and Kandil, 2010) and growing medium (Kleiber et al., 2012). The content of that microelement determined by other authors varied. The content of Cu in tomato fruits was lower than found in other studies.

Mn also affects absorption and translocation of other nutrients or elements, such as P, K, Ca, Mg, Fe, Zn, Cu and Si (Ducic and Polle, 2005; Foy et al., 1981; Galvez et al., 1989;

**Table 1**

**Content of macroelements according to other authors (in % in d.m.; * in mg·kg$^{-1}$ d.m.)**

| Author/s | Nutrient content | | | | |
|---|---|---|---|---|---|
| | N | P | K | Ca | Mg |
| **Leaves** | | | | | |
| Agric. Service... (2001) | 3.5-5.0 | 0.3-0.65 | 3.5-4.5 | 1.0-3.0 | 0.35-1.00 |
| Campbel (2000) | 3.5-5.0 | 0.3-0.7 | 3.0-4.5 | 1.0-2.0 | 0.3-0.8 |
| Chohura and Komosa (2003a) | 3.31-3.89 | 0.36-0.47 | 5.02-5.54 | 7.08-7.47 | 0.45-0.69 |
| Jarosz and Dzida (2011) | 3.97-4.27 | 0.47-0.51 | 4.30-5.11 | 2.12-2.98 | 0.27-0.36 |
| Kowalska (2004) | 4.83-4.99 | 0.51-0.74 | 3.99-4.08 | 3.13-3.36 | 0.65-0.69 |
| Kreij et al. (1990) | 2.8-4.2 | 0.30-0.46 | 3.5-5.1 | 1.6-3.2 | 0.36-0.50 |
| Pawlińska and Komosa (2006) | 4.22-4.27 | 0.74-0.78 | 6.15-6.27 | - | - |
| Plank (1999) | 3.5-5.0 | 0.5-1.0 | 3.5-5.0 | 0.9-1.8 | 0.5-1.0 |
| **Fruits** | | | | | |
| Fanasca et al. (2006) | 1.98-2.08 | 0.49-0.52 | 2.97-3.59 | 0.97-3.59 | - |
| Jarosz and Dzida (2011) | 2.27-2.41 | 0.27-0.32 | 3.88-4.23 | 850-1096* | 669-804* |
| Kleiber et al. 2012 | 1.47-2.59 | 0.47-0.54 | 4.61-5.09 | 0.15-0.20 | 0.68-0.86 |
| Olaniyi et al. (2010) | - | 0.38-0.57 | 0.07-0.17 | 0.25-0.38 | 0.12-0.22 |

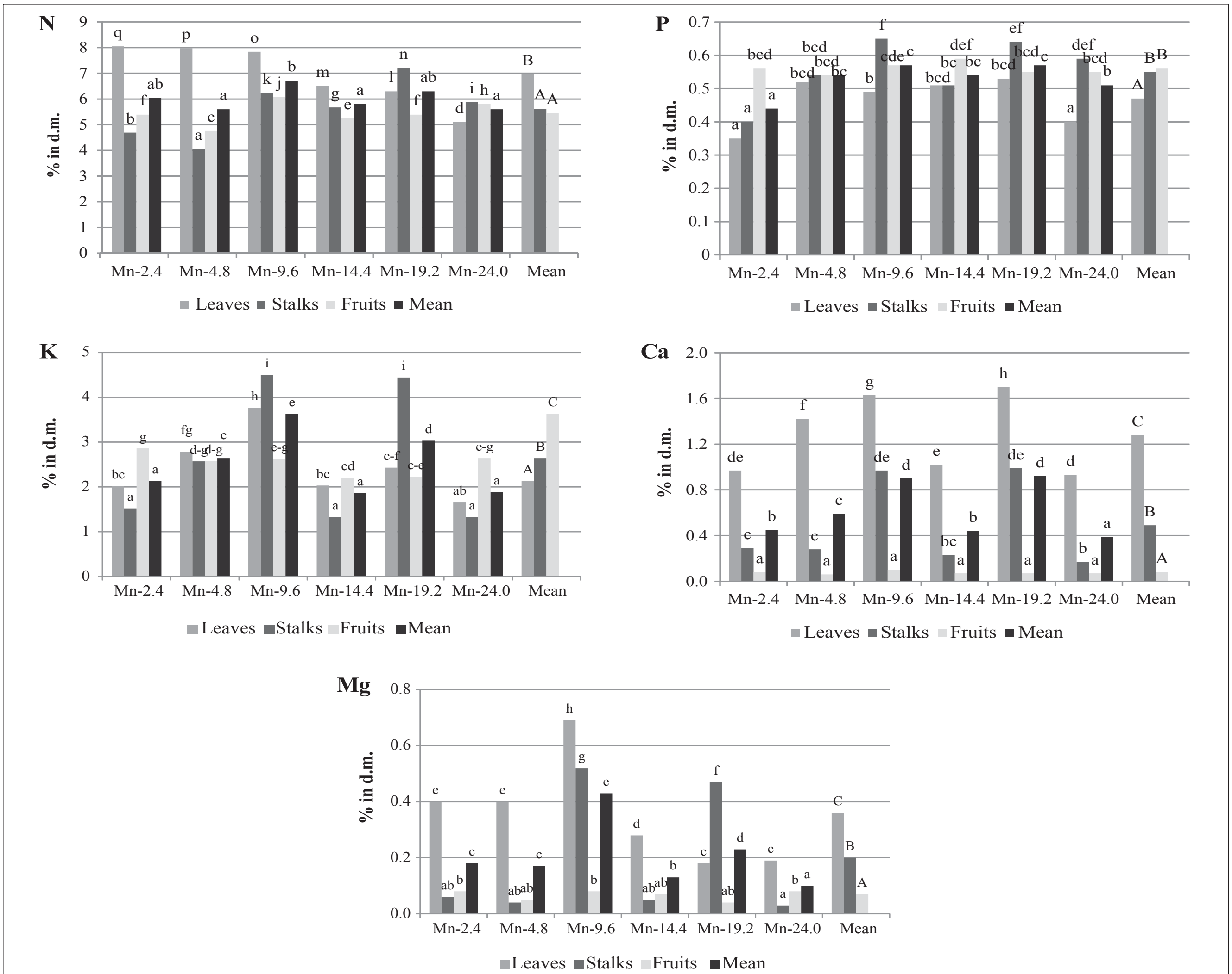


**Fig. 3. The effect of Mn stress on the macroelement content in different parts of plant, % in d.m.**

Horst and Marschner, 1978; Kasraei et al., 1996; Lei et al., 2007; Lee et al., 2011; Shenker et al., 2004). Based to the classification of correlations depending on the values of the coefficient of correlation an almost complete correlation was found between increasing Mn stress and N content in leaves (negative, r=-0.96) and between Mn content in NS and content of that ion in leaves, stalks, fruits and whole plants (positive, r ranged from 0.94 to 0.99) (Table 3). Almost complete correlations were also found between Mn content in leaves in relation to fruits, stalks: leaves as well as a very high correlation for Mn concentration in stalks: fruits.

In our studies for the Mn-24.0 combination we recorded manganese concentration of 259.4 mg·kg$^{-1}$ d.m. in leaves – with no visual symptoms of toxicity. Shenker et al. (2004) reported toxicity of that microelement with leaf Mn concentration of 207.4 mg·kg$^{-1}$ d.m. Savvas et al. (2009) in their experiment on the interactive effects of grafting and Mn supply on growth, yield, and nutrient uptake by tomato recorded higher concentrations of that ion in leaves (290 mg·kg$^{-1}$d.m.). Similarly to our study, both the above mentioned authors recorded higher Mn content than that reported by Mills and Jones (1996) at 250 mg·kg$^{-1}$ in leaf petioles to be the maximum safe level for tomato. Savvas et al. (2009) reported a reduced fruit yield in NS application with Mn concentration at 2.65 mg (=50 µM) (in the case of plants grafted onto 'He-Man'). In our studies the application of Mn above 2.4 mg per 1 dm$^3$ caused a significantly decrease in fruit yielding. Kleiber (2014) reported that in the case of tomato grown in rockwool a significant decrease in tomato production was recorded at 1.2 mg Mn per 1 dm$^3$ comparing with 0.3 and 0.6 mg·dm$^{-3}$. The first visual symptoms of Mn toxicity on the plants were reported after 1.5-month application of NS with 19.2 mg·dm$^{-3}$ Mn, but it was after 3 months in the case of 4.8 mg·dm$^{-3}$. Manganese is absorbed in the $Mn^{+2}$ forms and next it is quickly transported to the aboveground parts, thus the symptoms of deficit or toxicity of this nutrient are first manifested on the aboveground parts of plants (Marschner, 1998). Visual symptoms of Mn toxicity are shown in the form of brown spots on older leaf surfaces, chlorosis and necrosis, and browning of roots, leaf drying and defoliation (Foy et al., 1978; Horst and Marschner, 1978; Horst, 1988). Symptoms of toxicity of the nutrient lead to a reduction in the intensity of

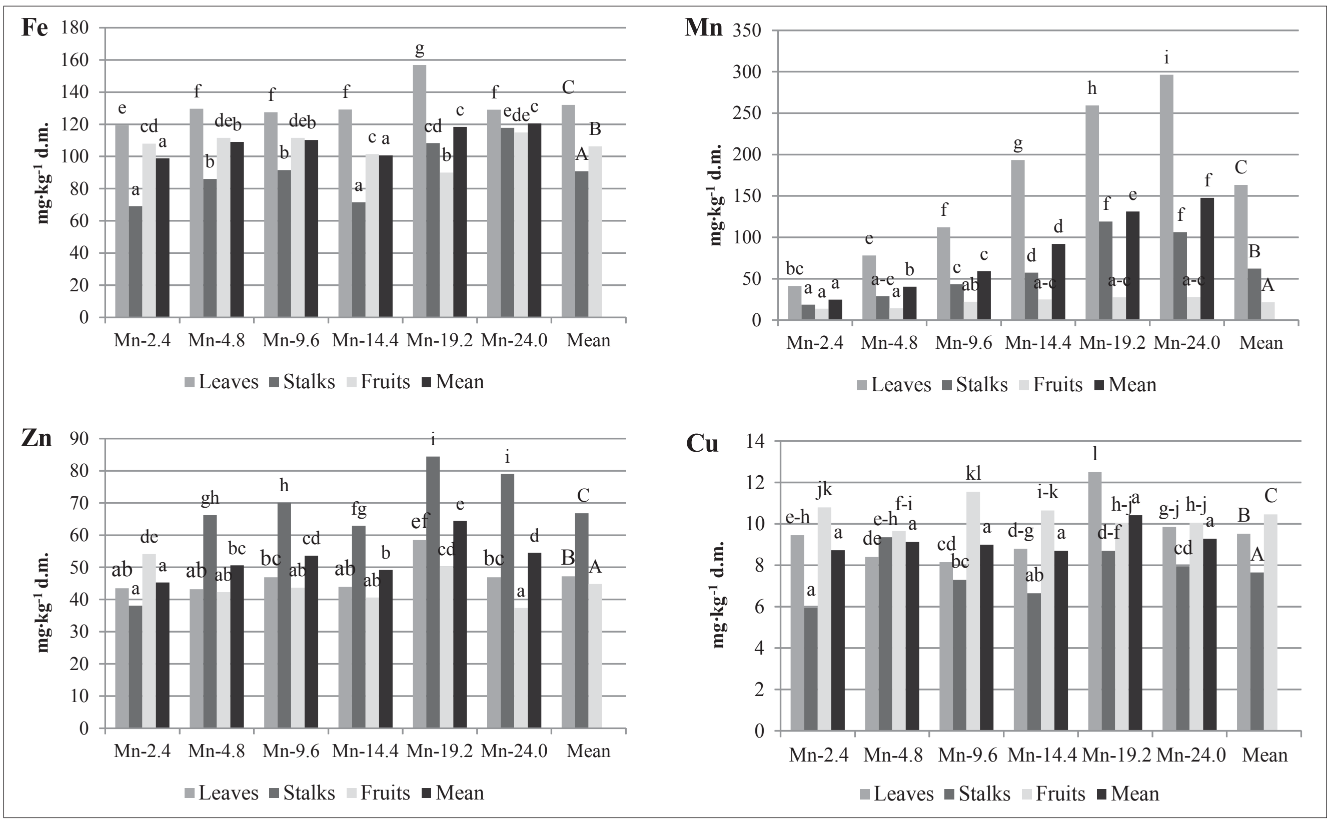


**Fig. 4. The effect of Mn stress on the microelement content in different parts of plant, in mg·kg$^{-1}$ d.m.**

photosynthesis and, consequently, to a significant reduction in crop yield (Horst, 1988; Marschner, 1998; Reichman et al., 2001; Kitao et al., 2001; Kleiber, 2014).

Tomato is considered a species relatively tolerant to manganese (Foy, 1973; Savvas et al., 2009). Based on our studies it may be stated that in the short term tomato exhibits a relatively high tolerance to Mn understood as a lack of visual symptoms on the plants with a simultaneous decrease in biomass production. Other plant species vary in terms of their sensitivity to high concentrations of manganese. In Chinese cabbage a significant reduction of plant yielding was observed at the application of NS containing 80 mg Mn·dm$^{-3}$ (Lee et al., 2011). Desmond (1993) in sweet potato growing indicated a significant deterioration of growth at the application of high Mn contents reaching up to 100 mg·dm$^{-3}$. Le Bot et al. (1990b) found a significant effect of magnesium as an ion causing an increase in tolerance to high Mn concentrations in the NS as a result of a significant reduction in its uptake. According to the same authors, a better indication of potential toxicity of Mn is the Mg:Mn ratio rather than Mn concentration alone. Another ion having an alleviating effect on symptoms of Mn toxicity is silicon (Horst and Marschner, 1978; Osawa and Ikeda, 1976). This effect was also found in the case of tomato and lettuce cultivation (Kleiber, unpublished data).

**Table 2**
**Content of microelements according to other authors (in mg kg$^{-1}$ d.m.)**

| Author/s | Nutrient content (average or range) | | | |
|---|---|---|---|---|
| | Fe | Mn | Zn | Cu |
| Leaves | | | | |
| Agric. Service…(2001) | 50-300 | 25-200 | 18-80 | 5-35 |
| Atherton et al. (1986) | >60 | 237.0 | 38.0 | 9.68 |
| Breś and Ruprik (2007) | 80.0-120.4 | 70.6-190.9 | 66.9-102.1 | 8.70-15.62 |
| Campbel (2000) | 45-300 | 30-300 | 18-75 | 41-789 |
| Chohura and Komosa (2003b) | 85.5-161.9 | 252.0-273.3 | 33.8-75.8 | 10.23-13.84 |
| Kowalska (2004) | 136.7-141.6 | 115.6-137.0 | 40.6-47.5 | 33.02-36.60 |
| Kreij et al. (1990) | 84-112 | 54-165 | 54-76 | 6 |
| Plank (1999) | 50-300 | 50-100 | 20-100 | 41-871 |
| Uchida (2000) | 60-300 | 50-250 | - | - |
| Fruits | | | | |
| Gad and Kandil (2010) | 133-145 | 44.5-53.3 | 32.6-42.6 | 29.2-37.7 |
| Kleiber et al. (2012 | 56.7-87.5 | 12.4-27.4 | 13.2-26.6 | 13.1-14.6 |
| Olaniyi et al. (2010) | 15.4-31.1 | - | - | - |
| Salam et al. (2010) | - | - | 34.3-45.7 | - |

**Table 3**
**The Pearson's correlation between content of Mn in NS and nutrient content in differ parts of plants and between different parts of plant**

| Nutrient | Leaves* | Stalks* | Fruits* | Mean | Leaves: fruits | Stalks: leaves | Stalks: fruits |
|---|---|---|---|---|---|---|---|
| N | **-0.96** | 0.71 | 0.1 | -0.13 | 0.06 | -0.54 | 0.17 |
| P | 0.14 | 0.59 | 0 | 0.38 | 0.08 | 0.6 | -0.16 |
| K | -0.38 | 0.02 | -0.5 | -0.18 | 0.07 | 0.79 | -0.23 |
| Ca | -0.05 | 0.09 | -0.19 | 0.02 | 0.25 | 0.89 | 0.55 |
| Mg | -0.63 | 0.13 | -0.09 | -0.27 | 0.39 | 0.39 | -0.24 |
| Fe | 0.53 | 0.79 | -0.24 | 0.75 | -0.81 | 0.55 | -0.02 |
| Mn | 0.95 | 0.94 | 0.95 | 0.99 | 0.93 | 0.98 | 0.89 |
| Zn | 0.56 | 0.79 | -0.49 | 0.66 | 0.3 | 0.67 | -0.49 |
| Cu | 0.54 | 0.26 | -0.26 | 0.53 | -0.39 | 0.26 | -0.71 |

## Conclusion

Mn stress did not significantly affect the number of leaves, flowers per inflorescence and visible clusters on the plants. Increasing Mn stress had a significant effect on the fresh weight of leaves, stalks, shoots and fruits. The biomass production was reduced by about 17.6%, while fruit production by about 25.7% when comparing Mn-24.0 with Mn 2.4. In spite of the significant reduction of biomass production in all the studied combinations there were no other visual symptoms of Mn toxicity on the plants. Mn stress significantly affected the nutrient uptake. A significant downward trend was shown for N content in leaves with a simultaneous positive correlation of Mn concentration in NS with the content of that microelement in leaves, stalks and fruits. Significant differences were found in the content of nutrients in different parts of plants. The highest mean content of N was found for leaves, P – in the case of stalks and fruits; K, Cu – in fruits, Ca, Mg, Fe and Mn – in leaves, while that of Zn in stalks. The lowest N content was determined in stalks and fruits, that P and K - in leaves, Ca, Mg, Mn and Zn – in fruits, while that of Fe and Cu – in stalks.

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