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THE EFFECT OF FOLIAR FERTILIZATION OF FRENCH BEAN WITH IRON SALTS AND UREA ON SOME PHYSIOLOGICAL PROCESSES IN PLANTS RELATIVE TO IRON UPTAKE AND TRANSLOCATION IN LEAVES

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Abstract. Iron chlorosis is a wide-spread limiting factor of production in agriculture. Fe deficiencies is done mainly by foliar sprays because soil application generally are ineffective, especially for annual crops. A pot experiment, conducted in a phytotron, investigated the effectiveness of foliar fertilization of French bean with 3 inorganic iron salts [$FeSO_4 \cdot 7H_2O$, $FeCl_3 \cdot 6H_2O$, $Fe(NO_3)_3 \cdot 9H_2O$] and 2 organic iron salts [Fe-Citr., Fe-EDTA], applied with and without the addition of 0.5% $CO(NH_2)_2$. Iron was applied 3 times only on simple leaves in the form of aqueous solutions containing 0.2 mg Fe in 1 cm^3, compared to water as the control treatment. The obtained results show that the application of iron salt solutions resulted in a distinct increase of Fe content in simple leaves and in the next trifoliate leaves. Foliar fertilization of the plants with $Fe(NO_3)_3$ was the most effective, while feeding with Fe-EDTA was the least effective. Iron given in the form of chelates showed greater mobility in the plants than that applied in the form of inorganic salts. Foliar fertilization of the plants with inorganic iron salts significantly increased chlorophyll a+b and carotenoid content in the leaves as well as their stomatal conductance and the photosynthesis and transpiration rates. But the impact of Fe chelates, in particular Fe-EDTA, on the abovementioned plant traits was not clear. The leaves of the plants treated with $Fe(NO_3)_3$ showed the highest content of photosynthetic pigments and the most intense gas exchange. The application of inorganic iron salts and Fe-Citr. resulted in a significant increase in the number of nodules formed on the bean roots and their weight. The plants treated with Fe-Citr. produced the largest number of nodules, while those treated with $Fe(NO_3)_3$ developed nodules with the highest weight. The addition of urea to the iron solutions had an effect on the increase in the value of the iron transport rate and on the decrease in iron and carotenoid content, the leaf gas exchange rate as well as the number of root nodules and their weight.

Key words: foliar nutrition, iron chelates, inorganic iron, gas exchange, chlorophyll, nodules

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INTRODUCTION

Iron deficit in plants decreases the content of photosynthetic pigments [Terry 1980], which is termed as iron chlorosis. Under such conditions, leaf stomatal conductance is reduced which, as a consequence, causes a decline in the photosynthesis and transpiration rates as well as an increase in substomatal CO_2 concentration [Larbi et al. 2006]. In order to eliminate diseases, new plant growing technologies recommend spraying plants with compounds of iron, since its supply to the soil is usually not very effective. $FeSO_4$ [Abadia et al. 2002; Fernandez et al. 2006] or $FeCl_3$ [Brüggemann et al. 1993; Schőnherr et al. 2005] are used most frequently for this purpose. However, because inorganic Fe salts are not very stable and they are phytotoxic due to a low pH [Schőnherr et al. 2005], but also due to the fact that the uptake of Fe^{+3} iron must be preceded by its reduction to Fe^{+2} [Brüggemann et al. 1993], organic iron compounds in the form of chelates are used more and more frequently to save plants [Kannan and Wittwer 1965; Wittwer et al. 1967; Reed et al. 1988; Brüggemann et al. 1993; Rombola et al. 2000; Abadia et al. 2002; Schőnherr et al. 2005; Fernandez et al. 2006; Tyksiński and Komosa 2008]. But the effectiveness of iron supplied to plants in the form of chelates is not clearly demonstrated in the literature. Reed et al. [1988] report that the effectiveness of Fe chelates depends on such properties as the molecular mass of a complex, the dissociation constant and stability at varying pH of a solution. On the other hand, Schőnherr et al. [2005] found that there was no correlation between the molecular mass and the penetration rate of the complex. It also turned out that the permeability of the cuticular membrane decreased with an increasing concentration of Fe chelates; according to some authors, it may be attributed to the fact that these compounds, as the penetration proceeds, reduce the dimensions of water pores in the membrane. Therefore, in order to enhance the effectiveness of penetration of iron contained in Fe chelates, they suggest that 100% of air relative humidity should be maintained for a long time after the spraying. The penetration of Fe, in particular that supplied in the form of $FeSO_4$, was also significantly increased by the addition of urea [Kannan and Wittwer 1965; Wittwer et al. 1967], but Borowski and Michałek [2010] did not find a similar correlation with respect to the penetration of Mg from $MgSO_4$ and from other inorganic salts of this metal.

As the literature does not provide clear data on the effectiveness of inorganic and organic iron salts in foliar fertilization of plants, a study was undertaken that was designed to determine the effect of foliar fertilization of French bean with 3 inorganic iron salts with the participation of Fe^{+3} and Fe^{+2} ($FeSO_4$, $FeCl_3$, $Fe(NO_3)_3$) and with 2 Fe chelates of distinctly varying molecular mass (Fe-Citr., Fe-EDTA) on leaf iron content, translocation of iron in plants and its effect on photosynthetic pigment content in leaves, gas exchange, and root nodulation. All iron solutions were applied with or without the addition of urea.

MATERIALS AND METHODS

The experiments were conducted in a phytotron of the University of Life Sciences in Lublin in 2010 during the period from 14 June to 15 July (experiment I) and from 19

July to 20 August (experiment II). Plants of self-terminating French bean, cv. 'Korona', were grown in 2.0 dm^3 pots filled with quartz sand, without fine particles. The experiments were carried out according two-factorial scheme using fluorescent light with the far-field flux density of ca. 200 µmol × m^{-2} × s^{-1}, day length of 14 hours, air temperature of 24°C/18°C (day/night), and relative humidity of 80%.

Each experiment comprised 48 pots, with 2 plants growing in one pot. After emergence, the plants were fertilized by supplying, per pot, 210 mg N-NO_3, 236 mg K, 32 mg P, 200 mg Ca, 64 mg S, and 48 mg Mg in the form of a single concentration of Hoagland's medium as well as 2 cm^3 of the micronutrient solution (A-Z), without iron.

During the growing period, the plants were watered with distilled water to constant weight, maintaining substrate moisture content at a level of 70% of field water capacity (FWC). In the third week of growth, when the plants had fully-developed simple leaves, 12 experimental variants were set up with four statistical replicants (4 pots), differentiated in terms of the used foliar-applied iron salts and the addition of urea, or not. The respective experimental series were sprayed with aqueous solutions of the following iron salts: 1) H_2O – control; 2) $FeSO_4 \cdot 7H_2O$; 3) $FeCl_3 \cdot 6H_2O$; 4) $Fe(NO_3)_3 \cdot 9H_2O$; 5) Fe-Citr.($C_6H_5FeO_7 \cdot H_2O$); 6) Fe-EDTA ($C_{10}H_{12}FeN_2NaO_8 \cdot 3H_2O$) in pure form or a mixture of the particular salts with 0.5% $CO(NH_2)_2$, each time prepared just before spraying (experimental design – Table 1). The aqueous solutions of the abovementioned iron salts contained 0.2 mg of pure Fe in 1 cm^3. Foliar feeding of the plants with the abovementioned forms of iron salts was repeated in the fourth and fifth week of growth, spraying only simple leaves. During spraying, the remaining portion of each plant, which was above the simple leaves (including trifoliate leaves), was tightly isolated with plastic bags that remained on the plants until the solution dried up. The solutions were applied at an air temperature of 24°C, just before nightfall. They were sprayed using a small manual sprayer, each time until complete moisturization of the upper and lower leaf epidermis was obtained.

After 4 days from the last spraying, measurements were made of leaf stomatal conductance for water vapour as well as of the transpiration and photosynthesis rates. The measurements were made in 10 replications on the middle leaflet of fully-developed trifoliate leaves, using a leaf microclimate control system LCA-4. During measurement recording, temperature in the measurement chamber was approx. 25°C, while the far-field flux density approx. 200 µmol × m^{-2} × s^{-1}. Concurrently, leaf samples were collected to determine the content of chlorophyll a+b according to Arnon [1949] and the content of carotenoids according to Britton [1985]. Next, the above-ground parts of the bean plants were separated from their roots and the total fresh weight of aerial parts was determined, but the simple leaves earlier treated with the iron salt solutions were separated from the other leaves on the plant and subjected to drying. Before drying, the simple leaves were washed three times in distilled water. The number of root nodules and their total fresh weight were determined for the bean roots, after removing sand from them using water, whereas dry leaf material, after mineralization of leaves, was used to determine iron content with atomic absorption spectrometry (AAS). Based on the content of the element in question in the leaves of the plants treated with the Fe solutions and in the other leaves, their dry weight and translocation time, the rate of iron transport in the plants was calculated using the formula given in the paper of Baligar et al. [1993]. This paper presents average results obtained in two experiments. These data were subjected to statistical analysis using double cross-classification, determining the significance of differences by Tukey's test at the probability level $\alpha = 0.05$.

RESULTS AND DISCUSSION

In both experiments conducted, the control plants did not show any chlorosis symptoms in spite of the elimination of Hoagland's medium that had been used to feed the plants with nutrients at the beginning of the experiment as well as the spraying of the plants with distilled water during growth. This is also confirmed by the data contained in table 1, which show the sufficient content of Fe in simple and trifoliate leaves that did not significantly differ from the content of this component in the leaves of, for example, lettuce root-fed with iron chelates [Tyksiński and Komosa 2008]. In this case, the source of iron was probably the sand used in this experiment; in spite of the fact that this sand did not contain fine particles (it had been earlier rinsed with mains water and dried), but during the watering of the plants with distilled water at a pH of 5.5, it released sufficient amounts of this element for the plants.

Table 1. Effect of iron salts and addition of urea on iron content in simple and trifoliate leaves of French bean ($mg \cdot kg^{-1}$ d.m.)

Tabela 1. Wpływ soli żelaza i dodatku mocznika na zawartość żelaza w pojedynczych i trójlistkowych liściach fasoli szparagowej ($mg \cdot kg^{-1}$ s.m.)

Iron salts Sole żelaza	Solution – Roztwór		Mean Średnia	Solution – Roztwór		Mean Średnia
	without – bez $CO(NH_2)_2$	with – z $CO(NH_2)_2$		without – bez $CO(NH_2)_2$	with – z $CO(NH_2)_2$	
	simple – pojedyncze			trifoliate – trójlistkowe		
Control – Kontrola	220.2	120.4	170.3	110.5	80.4	95.4
$FeSO_4 \cdot 7H_2O$	750.5	300.9	525.7	180.6	200.2	190.4
$FeCl_3 \cdot 6H_2O$	600.4	280.8	440.6	180.7	180.8	180.7
$Fe(NO_3)_3 \cdot 9H_2O$	1000.8	450.6	725.7	310.5	205.3	257.9
Fe-Citr. – Fe-Cytr.	560.3	250.1	405.2	300.8	180.9	240.8
Fe-EDTA	560.6	170.7	365.6	230.4	120.3	175.3
Mean – Średnia	615.5	262.2		218.9	161.3	
$LSD_{0.05}$ for salt – $NIR_{0,05}$ dla soli			268.2		33.1	
$LSD_{0.05}$ for urea – $NIR_{0,05}$ dla mocznika			103.5		12.8	
$LSD_{0.05}$ for salt × urea – $NIR_{0,05}$ dla sól × mocznik			n.s – r.n.		54.6	

Despite the sufficient amount of Fe in the leaves of the control plants, triple foliar spraying of the youngest leaves on the plants distinctly increased iron content in these leaves as well as in the next developing leaves until the onset of flowering of the plants. The simple leaves treated with the solutions of the used iron salts contained, on average, 2–3 times more iron than the remaining trifoliate leaves, which seems to be fully understandable. However, the leaves of the plants treated with the solution of $Fe(NO_3)_3$ showed the highest Fe content (both simple and trifoliate leaves), while a significantly lower iron content was found in the simple and trifoliate leaves sprayed with the other Fe salts, except for $FeSO_4$ in the case of simple leaves and Fe-Citr. in the case of trifoliate leaves. Higher Fe uptake from $FeSO_4$ than from chelates or the quicker re-greening in chlorotic plants under the influence of this salt has also been found by Kannan and Wittwer [1965], Rombola et al. [2000], Fernandez et al. [2006]. It is difficult to explain what the highest content of iron supplied in the form of $Fe(NO_3)_3$ can be attributed to, in particular that in the available literature there are no studies that would investigate the application of this salt, but it seems to be related to a high tendency of leaves to absorb NO_3^- ions in this way. In these conditions for, balancing the increased amount of negative charges inserted with NO_3^- anions, the leaves took the additional amount of iron cations. Using foliar application of various nitrogen salts, Borowski and Michałek [2008] found that spinach leaves treated with $Ca(NO_3)_2$ contained the highest amount of total N. However, similar iron absorption from the other inorganic and organic Fe salts finds confirmation in the studies of Reed et al. [1988] and Abadia et al. [2002]. The addition of urea to all iron salts reduced 2–3 times, on average, iron content in simple leaves, which evidently translated into lower iron content in trifoliate leaves (tab. 1). It is difficult to explain, in particular given that Kannan and Wittwer [1965] as well as Wittwer et al. [1967] found that the addition of $CO(NH_2)_2$ to the solution of $FeSO_4$ increased Fe absorption by leaves. In the case in question, this may have resulted from the fact that iron in the application solution occurred at a lower concentration than urea; hence, after the application of the solution on the leaves, NH_4^+ ions formed from the decomposition of $CO(NH_2)_2$ effectively competed with Fe^{+3} or Fe^{+2} ions in the penetration through the leaf epidermis. In a similar study on the application of various magnesium salts on spinach leaves, Borowski and Michałek [2010] also found that the addition of urea decreased Mg^{+2} absorption by leaves.

Table 2. Effect of iron salts and addition of urea on value transport index (I_{TR}) of iron beetwen leaves of French bean ($\mu mol \cdot g^{-1} \cdot h^{-1}$)

Tabela 2. Wpływ soli żelaza i dodatku mocznika na wartość indeksu transportu (I_{TR}) pomiędzy liśćmi fasoli szparagowej ($\mu mol \cdot g^{-1} \cdot h^{-1}$)

Iron salts Sole żelaza	Solution – Roztwór		Mean Średnia
	without – bez $CO(NH_2)_2$	with – z $CO(NH_2)_2$	
Control – Kontrola	3.53	2.88	3.20
$FeSO_4 \cdot 7H_2O$	5.59	8.80	7.19
$FeCl_3 \cdot 6H_2O$	8.35	9.32	8.83
$Fe(NO_3)_3 \cdot 9H_2O$	6.04	8.63	7.33
Fe-Citr. – Fe-Cytr.	9.60	11.61	10.60
Fe-EDTA	9.14	9.95	9.54
Mean – Średnia	7.04	8.53	

The value of the calculated rate of Fe transport between leaves to which iron had been supplied, compared to the other leaves, was described on the basis of comparison of means, and depended on both the type of iron salt and the urea addition. The value of the parameter in question was the lowest for the control plants; it was nearly 2.5 times higher for inorganic iron salts, and almost 3 times higher for Fe chelates. Kannan and Wittwer [1965], Wittwer et al. [1967], Fernandez and Ebert [2005] also confirm that the transport of iron supplied to leaves in the form of chelate compounds occurs more efficiently than the transport of Fe from inorganic salts. The addition of urea to the used foliar-applied Fe solutions also increased distinctly the value of the rate in question (tab. 2).

The increase in the content of iron in bean leaves associated with its application to the oldest leaves had a significant impact on the content of photosynthetic pigments in trifoliate leaves (tab. 3). Generally, foliar treatment of the plants with all iron salts, except for Fe-EDTA, significantly increased the content of chlorophyll a+b and carotenoids. This undoubtedly resulted from the fact that iron is a stimulator of the activity of chlorophyll synthesis enzymes, and even up to 80% of the total amount of this component in a plant is accumulated in chloroplasts. The obtained data are confirmed by similar studies conducted on beet plants [Terry 1980], and peach trees [Fernandez et al. 2006]. On the other hand, the insignificant increase in chlorophyll a+b and carotenoid content, relative to the control treatment, under the effect of Fe-EDTA could have resulted from a low iron content in the leaves treated with this chelate related to its relatively high molecular mass (421.1), whereas the molecular mass of Fe-Citr. is markedly lower (262.8). Reed et al. [1988] report that the ability of leaves to absorb nutrients contained in chelates depends, *inter alia*, on the molecular mass of a complex – the uptake of nutrients from a complex of high molecular mass is more difficult. The addition of urea to the application solutions significantly decreased leaf carotenoid content, but had no effect on chlorophyll a+b content (tab. 3).

Table 3. Effect of iron salts and addition of urea on content of chlorophyll a+b and corotenoids in leaves of French bean

Tabela 3. Wpływ soli żelaza i dodatku mocznika na zawartość chlorofilu a+b i karotenoidów w liściach fasoli szparagowej

Iron salts Sole żelaza	Solution – Roztwór		Mean Średnia	Solution – Roztwór		Mean Średnia
	without – bez $CO(NH_2)_2$	with – z $CO(NH_2)_2$		without – bez $CO(NH_2)_2$	with – z $CO(NH_2)_2$	
	chlorophyll (mg · g^{-1} f.m.) chlorofil (mg · g^{-1} ś.m.)			carotenoids (mg · g^{-1} f.m.) karotenoidy (mg · g^{-1} ś.m.)		
Control – Kontrola	2.83	2.56	2.69	0.25	0.18	0.21
$FeSO_4 \cdot 7H_2O$	3.04	3.03	3.03	0.30	0.25	0.27
$FeCl_3 \cdot 6H_2O$	2.88	2.93	2.90	0.29	0.24	0.26
$Fe(NO_3)_3 \cdot 9H_2O$	3.32	3.26	3.29	0.30	0.27	0.28
Fe-Citr. – Fe-Cytr.	3.06	3.08	3.07	0.28	0.24	0.26
Fe-EDTA	2.85	2.81	2.83	0.27	0.21	0.24
Mean – Średnia	3.00	2.94		0.28	0.23	0.25
$LSD_{0.05}$ for salt – $NIR_{0,05}$ dla soli			0.22		0.05	
$LSD_{0.05}$ for urea – $NIR_{0,05}$ dla mocznika			n.s – r.n.		0.02	
$LSD_{0.05}$ for salt × urea – $NIR_{0,05}$ dla sól × mocznik			n.s – r.n.		n.s – r.n.	

The varying iron content in bean leaves also had an effect on leaf stomatal conductance; relative to the control, the plants treated with inorganic iron salts showed a significant increase in the value of the parameter in question, whereas those treated with chelates, in particular Fe-EDTA, demonstrated a distinct reduction. The addition of urea to the solutions also significantly decreased leaf stomatal conductance (tab. 4). Transpiration and photosynthesis of bean leaves changed similarly to the changes of stomatal conductance under the conditions of the present experiments. It seems to be fully justified, since the degree of stomatal opening determines the rate of diffusion of water vapour from leaves and of CO_2 into leaves. All inorganic iron salts significantly increased the intensity of transpiration compared to the control; this increase was the highest in the case of $Fe(NO_3)_3$, and the lowest for $FeCl_3$, whereas Fe-Citr., in particular Fe-EDTA, significantly reduced the rate of discharge of water vapour from leaves.

Table 4. Effect of iron salts and addition of urea on stomatal conductance and intensity of transpiration of French bean leaves

Tabela 4. Wpływ soli żelaza i dodatku mocznika na przewodność szparkową i intensywność transpiracji fasoli szparagowej

Iron salts Sole żelaza	Solution – Roztwór without – bez $CO(NH_2)_2$	Solution – Roztwór with – z $CO(NH_2)_2$	Mean Średnia	Solution – Roztwór without – bez $CO(NH_2)_2$	Solution – Roztwór with – z $CO(NH_2)_2$	Mean Średnia
	conductance – przewodność ($mol\ H_2O \cdot m^{-2} \cdot s^{-1}$)			transpiration – transpiracja ($mmol\ H_2O \cdot m^{-2} \cdot s^{-1}$)		
Control – Kontrola	0.19	0.14	0.16	2.16	1.58	1.87
$FeSO_4 \cdot 7H_2O$	0.21	0.20	0.20	2.50	2.25	2.37
$FeCl_3 \cdot 6H_2O$	0.19	0.19	0.19	2.27	2.01	2.14
$Fe(NO_3)_3 \cdot 9H_2O$	0.23	0.21	0.22	2.76	2.82	2.79
Fe-Citr. – Fe-Cytr.	0.17	0.17	0.17	1.71	1.64	1.67
Fe-EDTA	0.14	0.14	0.14	1.22	1.36	1.29
Mean – Średnia	0.19	0.17		2.10	1.94	
$LSD_{0.05}$ for salt – $NIR_{0,05}$ dla soli			0.02		0.05	
$LSD_{0.05}$ for urea – $NIR_{0,05}$ dla mocznika			0.01		0.02	
$LSD_{0.05}$ for salt × urea – $NIR_{0,05}$ dla sól × mocznik			0.03		0.08	

The applied iron salts had a similar effect, to some extent, on leaf CO_2 assimilation as their effect on transpiration, since all inorganic Fe compounds as well as ferric citrate increased the intensity of photosynthesis. $Fe(NO_3)_3$ had the most beneficial effect (likewise its effect on transpiration), followed by $FeSO_4$, whereas the effect of $FeCl_3$ was the same as that of Fe-Citr. On the other hand, treatment of the plants with Fe-EDTA significantly decreased the intensity of the photosynthesis process. The addition of urea to the applied iron salts significantly decreased both transpiration and photosynthesis in the test plants (tab. 4, 5). It is difficult to confront the results obtained in this respect with literature data due to a lack of such data. It is only known from the study of Larbi et al. [2006] on beet, pear, and peach plants that iron deficit decreased stomatal conductance, photosynthesis, and transpiration. In the case of the present study, this seems to have resulted only from the effect of the applied salts on leaf stomatal conductance, as measurements of potential quantum yield of chlorophyll Fv/Fm (unpublished data) in the leaves of the bean plants did not show any significant differences.

An analysis of the amount of biomass produced by the above-ground parts of the plants shows a close connection with the intensity of the photosynthesis process in the leaves (tab. 5), as the biomass of the plants foliar treated with all iron salts, apart from Fe-EDTA, was higher than in the control treatment, but there was a significant increase only in the case of the application of $Fe(NO_3)_3$ and $FeSO_4$. Also, no effect was found of urea on the value of the trait in question (tab. 5).

Table 5. Effect of iron salts and addition of urea on intensity of photosynthesis and fresh mass of top parts French bean plants.
Tabela 5. Wpływ soli żelaza i dodatku mocznika na intensywność fotosyntezy i świeżą masę części nadziemnych roślin fasoli szparagowej

Iron salts Sole żelaza	Solution – Roztwór without – bez $CO(NH_2)_2$	Solution – Roztwór with – z $CO(NH_2)_2$	Mean Średnia	Solution – Roztwór without – bez $CO(NH_2)_2$	Solution – Roztwór with – z $CO(NH_2)_2$	Mean Średnia
	photosynthesis fotosynteza (μmol $CO_2 \cdot m^{-2} \cdot s^{-1}$)			fresh mass top parts (g · $plant^{-1}$) świeża masa części nadziemnych (g · $roślina^{-1}$)		
Control – Kontrola	5.70	4.83	5.26	22.48	20.97	21.72
$FeSO_4 \cdot 7H_2O$	6.63	6.28	6.45	24.96	25.72	25.34
$FeCl_3 \cdot 6H_2O$	5.75	5.74	5.74	22.06	22.20	22.13
$Fe(NO_3)_3 \cdot 9H_2O$	7.29	6.38	6,83	26.30	25.86	26.08
Fe-Citr. – Fe-Cytr.	5.65	5.80	5.72	23.01	23.12	23.06
Fe-EDTA	4.81	4.39	4.60	21.56	21.88	21.72
Mean – Średnia	5.97	5.57		23.39	23.29	
$LSD_{0.05}$ for salt – $NIR_{0,05}$ dla soli			0.11		3.55	
$LSD_{0.05}$ for urea – $NIR_{0,05}$ dla mocznika			0.04		n.s – r.n.	
$LSD_{0.05}$ for salt × urea –$NIR_{0,05}$ dla sól × mocznik			0.18		n.s – r.n.	

Table 6. Effect of iron salts and addition of urea on number and fresh mass of nodules on roots of French bean plants.
Tabela 6. Wpływ soli żelaza i dodatku mocznika na liczbę i świeżą masę brodawek na korzeniach roślin fasoli szparagowej

Iron salts Sole żelaza	Solution – Roztwór without – bez $CO(NH_2)_2$	Solution – Roztwór with – z $CO(NH_2)_2$	Mean Średnia	Solution –Roztwór without – bez $CO(NH_2)_2$	Solution –Roztwór with – z $CO(NH_2)_2$	Mean Średnia
	number of nodules (pieces · $plant^{-1}$) liczba brodawek (szt. · $roślina^{-1}$)			fresh mass of nodules (g · $plant^{-1}$) świeża masa brodawek (g · $roślina^{-1}$)		
Control – Kontrola	18.2	9.8	14.0	0.140	0.080	0.110
$FeSO_4 \cdot 7H_2O$	34.2	21.2	27.7	0.298	0.180	0.239
$FeCl_3 \cdot 6H_2O$	27.0	19.8	23.4	0.242	0.172	0.207
$Fe(NO_3)_3 \cdot 9H_2O$	42.5	25.0	33.7	0.787	0.275	0.531
Fe-Citr. – Fe-Cytr.	47.5	29.7	38.6	0.588	0.353	0.470
Fe-EDTA	12.2	3.0	7.6	0.155	0.018	0.086
Mean – Średnia	30.3	18.1		0.368	0.180	
$LSD_{0.05}$ for salt – $NIR_{0,05}$ dla soli			8.31		0.186	
$LSD_{0.05}$ for urea – $NIR_{0,05}$ dla mocznika			3.26		0.073	
$LSD_{0.05}$ for salt × urea – $NIR_{0,05}$ dla sól × mocznik			n.s – r.n.		0.305	

The effect of treatment of the plants with various iron salts and the addition of $CO(NH_2)_2$ on root nodulation was much clearer, since the plants treated with Fe-EDTA produced nearly 2 times fewer nodules on their roots than in the control treatment. The application of the other iron salts significantly increased the number of nodules. The highest number of nodules developed on the roots of the plants sprayed with Fe-Citr., followed by $Fe(NO_3)_3$, $FeSO_4$, $FeCl_3$. The addition of urea to the iron solutions significantly reduced the number of nodules as well as their total weight, which seems to be quite obvious, as the plants showed a lower requirement for symbiotic N under these conditions. But it is interesting that foliar treatment of the plants with $Fe(NO_3)_3$ did not result in a reduction in the number of nodules and even, as shown by their weight, they were larger (tab. 6). This may indicate that nitrates applied to the leaves together with the solution of $Fe(NO_3)_3$ were reduced and metabolized to amino acids and proteins to a small extent. The significantly beneficial effect of foliar fertilization of bean with the used iron salts on the number and weight of root nodules results from the participation of non-heme iron proteins in the processes of symbiotic N_2 fixation by bacteria.

CONCLUSIONS

1. The application of inorganic iron salts ($FeSO_4$, $FeCl_3$, $Fe(NO_3)_3$) and organic iron salts (Fe-Citr, Fe-EDTA) on the first simple bean leaves clearly increased iron content in these leaves and in the next trifoliate leaves, relative to the control treatment. Foliar fertilization of the plants with $Fe(NO_3)_3$ was the most effective, while feeding with Fe-EDTA was the least effective. It seems is necessary to link it in the first case with the necessity of maintaining ionic balance at intensive penetration of leaves with NO_3^- ions, but in the second case with high constant durability of the complex and its big ionic mass. Iron given in the form of chelates showed greater mobility (transport rate) in the plants than that applied in the form of inorganic salts.

2. Foliar fertilization of the plants with inorganic iron salts significantly increased chlorophyll a+b and carotenoid content in leaves as well as their stomatal conductance and the photosynthesis and transpiration rates. But the impact of Fe chelates, in particular Fe-EDTA, on the abovementioned plant traits was not clear. The leaves treated with $Fe(NO_3)_3$ showed the highest content of photosynthetic pigments and the most intense gas exchange.

3. The application of inorganic iron salts and Fe-Citr. resulted in a significant increase in the number of nodules formed on the bean roots and their weight. The presented relationship results from iron participation in the nitrogenase involved in the process of N_2 fixation. The plants treated with Fe-Citr. produced the largest number of nodules, while those treated with $Fe(NO_3)_3$ developed nodules with the highest weight.

4. The addition of urea to the iron solutions had an effect on the increase in the value of the iron transport rate and on the decrease in iron and carotenoid content, the leaf gas exchange rate as well as the number of root nodules and their weight.

5. Foliar fertilization of plants that do not show iron deficit symptoms with inorganic salts of this metal and Fe-Citr. has a beneficial effect on the important physiological processes in plants.

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WPŁYW DOLISTNEGO NAWOŻENIA FASOLI SZPARAGOWEJ SOLAMI ŻELAZA I MOCZNIKIEM NA PRZEBIEG NIEKTÓRYCH PROCESÓW FIZJOLOGICZNYCH W ROŚLINACH NA TLE JEGO POBIERANIA I TRANSLOKACJI W LIŚCIACH

Streszczenie. Chloroza żelazowa jest czynnikiem w szerokim zakresie ograniczającym produkcję rolniczą. Niedobór żelaza jest uzupełniany głównie poprzez dokarmianie dolistne, ponieważ aplikacja doglebowa jest generalnie nieefektywna, szczególnie w uprawie roślin jednorocznych. W doświadczeniu wazonowym prowadzonym w fitotronie badano efektywność dolistnego nawożenia fasoli szparagowej trzema nieorganicznymi [$FeSO_4 \cdot 7H_2O$, $FeCl_3 \cdot 6H_2O$, $Fe(NO_3)_3 \cdot 9H_2O$] i dwoma organicznymi [Fe-Cytr., Fe-EDTA] solami żelaza podanymi bez dodatku i z dodatkiem 0.5% $CO(NH_2)_2$. Żelazo zastosowano 3-krotnie tylko na liście pojedyncze w formie roztworów wodnych zawierających w 1 cm^3 – 0,2 mg Fe wobec wody jako kontroli. Uzyskane wyniki wykazały, że aplikacja roztworów soli żelaza wpłynęła na wyraźne zwiększenie zawartości Fe w liściach pojedynczych i kolejnych liściach 3-listkowych. Najbardziej efektywne było dolistne nawożenie roślin $Fe(NO_3)_3$, a najmniej Fe-EDTA. Żelazo podane w formie chelatów wykazywało w roślinach większy stopień mobilności niż w formie soli nieorganicznych. Dolistne nawożenie roślin nieorganicznymi solami żelaza w istotnym stopniu zwiększało zawartość w liściach chlorofilu a+b, karotenoidów, a także ich przewodność szparkową i tempo przebiegu fotosyntezy i transpiracji. Natomiast wpływ chelatów Fe, a zwłaszcza Fe-EDTA na wymienione cechy roślin nie był jednoznaczny. Największą zawartość barwników asymilacyjnych i najbardziej intensywny przebieg wymiany gazowej wykazywały liście roślin traktowanych $Fe(NO_3)_3$. Aplikacja nieorganicznych soli żelaza i Fe-Cytr. wpłynęła na istotny wzrost liczby brodawek wiązanych na korzeniach fasoli i ich masy. Najwięcej brodawek wytworzyły na korzeniach rośliny traktowane Fe-Cytr., a brodawek o największej masie – $Fe(NO_3)_3$. Dodatek mocznika do roztworów żelaza wpłynął na wzrost wartości wskaźnika transportu żelaza, a spadek zawartości żelaza, karotenoidów, przebiegu wymiany gazowej w liściach oraz liczby i masy brodawek korzeniowych.

Słowa kluczowe: dokarmianie dolistne, chelaty żelaza, żelazo nieorganiczne, wymiana gazowa, chlorofil, brodawki

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