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Influence of nitrogen injection application on zinc and iron uptake by winter wheat and spring barley

Vliv injektážní aplikace dusíku na příjem zinku a železa ozimou pšenicí a jarním ječmenem

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Abstract

Influence of CULTAN method (Controlled Uptake Long Term Ammonium Nutrition) on the iron and zinc uptake by winter wheat (*Triticum aestivum* L.) and spring barley (*Hordeum vulgare* L.) was observed at four-year small-plot field experiments under conditions of the Czech Republic. No significant differences in iron and zinc concentration in grain of winter wheat as well as spring barley between conventional and CULTAN treatment were found. Neither increased supply of nitrogen fertilizer nor sulphur containing fertilizer resulted in significant differences in iron and zinc concentration in grain of winter wheat as well as spring barley at both nitrogen nutrition systems. No significant differences in sulphur concentration in aboveground biomass of winter wheat and spring barley between conventional and CULTAN treatment were recorded. Iron and zinc harvest index were not influenced by the CULTAN system. Because of no significant differences in iron and zinc concentration in grain of winter wheat as well as spring barley between conventional and CULTAN treatment, it can be assumed that nitrogen is taken up by CULTAN-treated plants in nitrate form.

Keywords: ammonium, CULTAN, iron, sulphur, zinc

Abstrakt

Ve čtyřletém přesném polním pokusu prováděném v polních podmínkách České republiky byl zkoumán vliv metody CULTAN (Controlled Uptake Long Term Ammonium Nutrition) na příjem zinku a železa ozimou pšenicí (*Triticum aestivum* L.) a jarním ječmenem (*Hordeum vulgare* L.). Nebyly pozorovány statisticky významné rozdíly v koncentraci zinku a železa v zrnu ozimé pšenice a jarního ječmene mezi konvenčním způsobem dusíkaté výživy a hnojením metodou CULTAN. Zvýšení dávky dusíkatého hnojiva ani přídavek síry v hnojivu nevedly ke statisticky významným změnám v koncentraci zinku a železa v zrnu obou plodin. Hnojení metodou CULTAN statisticky významně neovlivnilo koncentraci síry v nadzemní biomase ozimé pšenice ani jarního ječmene. Sklizňový index zinku a železa nebyl statisticky významně ovlivněn metodou CULTAN. Vzhledem k statisticky nevýznamným rozdílům v koncentraci zinku a železa v zrnu obou plodin lze předpokládat, že při hnojení metodou CULTAN je dusík rostlinami přijímán v nitrátové formě.

Klíčová slova: amonný dusík, CULTAN, síra, zinek, železo

Introduction

Zinc and iron deficiencies are well-documented public health issue (Gibson, 2012; Khush, et al., 2012). Management strategies targeting grain zinc and iron densities are urgently needed to fight zinc and iron deficiency in human populations (Govindaraj, et al., 2011; Cakmak et Hoffland, 2012). Iron and zinc are currently the trace minerals of greatest concern when considering the nutritional value of vegetarian diets. With elimination of meat and increased intake of phytate-containing legumes and whole grains, the absorption of both iron and zinc is lower with vegetarian than with nonvegetarian diets (Hunt, 2003).

Nitrogen fertilization increases iron and zinc concentration in wheat grain (Aciksoz, et al., 2011a; Xue, et al., 2012). However, excessive nitrogen fertilization results in greater nitrogen losses from arable soils (e.g. Haberle et Káš, 2012) and thus serious environmental problems (e.g. Olivier, et al., 2011; Zaehle et Dalmonech, 2011).

The CULTAN (Controlled Uptake Long Term Ammonium Nutrition) system consists in injection of fertilizer with a significant ratio of nitrogen in ammonium form into the root space of plants; the place of fertilizer application in soil is called „depot". Positive charge of ammonium and high concentration of fertilizer in these depots cause higher stability of fertilizer in soil resulting in ecological and economical benefits.

Because of the nitrification in soils it is suggested, that irrespectively of the applied N-form (ammonium, nitrate or urea), nitrogen will be available to the plants as nitrate. However, in the CULTAN-system, where ammonium is applied locally, ammonium is available to the plants and taken up and assimilated as a good compatible and quick responding N-form according to the intensity of plant growth (Sommer et Scherer, 2009). Ammonium supply enhances uptake of micronutrients compared to nitrate (Robin, et al., 2008; Elgharably, et al., 2010).

The aim of this study was to investigate an ability of nitrogen fertilizer injection to increase zinc and iron concentration in grain of winter wheat and spring barley resulting in higher nutritional value for human food.

Material and Methods

Small-plot field experiments with winter wheat (*Triticum aestivum* L.) cultivar Sulamit and spring barley (*Hordeum vulgare* L.) cultivar Jersey were run during 2007 – 2010 at three sites with different soil-climatic conditions in the Czech Republic (Central Europe): Hněvčeves (50°18'46.269"N, 15°42'51.552"E), Humpolec (49°32'49.604"N, 15°21'6.405"E) and Ivanovice na Hané (49°18'34.209"N, 17°5'18.753"E). Characteristics of the experimental sites are given in Table 1 and in Dvořák, et al. (2003) and Pavlíková, et al. (2002). In the trial, conventional treatment was compared to CULTAN treatment in which all the fertilizer was applied in one dose during vegetation period at the growth stage of BBCH 22 and BBCH 29-30, respectively. The impact of higher dose of nitrogen fertilizer and sulphur containing fertilizer was further determined (Tables 2 and 3). Detailed descriptions of methodology are given by Kozlovský, et al. (2009) and Sedlář, et al. (2011).

Table 1. Characteristics of experimental sites

Experimental site	Hněvčeves	Humpolec	Ivanovice na Hané
Altitude (m)	265	525	225
Precipitation amount (mm)	597	667	548
Air temperature (°C)	8.1	6.5	9.2
Soil type	haplic luvisol	cambisol	chernozem
Soil characteristic	clay loam	sandy loam	loam
pH/ $CaCl_2$	6.3	6.6	7.3
Total Zn content (mg*kg^{-1})	75	127	97
Bioavailable S content (mg*kg^{-1})	2.0	4.2	4.9
Ca $_{\text{Mehlich III}}$ content (mg*kg^{-1})	2522	2217	4458
Mg $_{\text{Mehlich III}}$ content (mg*kg^{-1})	158	183	287
K $_{\text{Mehlich III}}$ content (mg*kg^{-1})	291	197	390
P $_{\text{Mehlich III}}$ content (mg*kg^{-1})	89	120	142

Table 2. Fertilizer treatment, nitrogen amounts and timing – winter wheat.

Treatment	Dosage of added N per ha (fertilizer form)			Total N dosage (kg*ha^{-1})
	BBCH 22 (29)*	BBCH 33	BBCH 52	
conv.I	43 kg (CAN)	87 kg (CAN)	20 kg (CAN)	150
CULTAN I	150 kg (UAN)			150
conv.II	60 kg (CAN)	90 kg (CAN)	50 kg (CAN)	200
CULTAN II	200 kg (UAN)			200
conv.S	43 kg (AS)	87 kg (CAN)	20 kg (CAN)	150
CULTAN S	150 kg (UAS)			150

* CULTAN application in years 2007 and 2008 was carried out at the BBCH 29 growth stage
CAN – calcium ammonium nitrate (27 % N); UAN – urea ammonium nitrate (30 % N); AS – ammonium sulphate (20.5 % N, 24 % S); UAS – urea ammonium sulphate (19 % N, 5 % S)

Table 3. Fertilizer treatment, nitrogen amounts and timing – spring barley.

Treatment	Dosage of added N per ha (fertilizer form)			Total N dosage ($kg*ha^{-1}$)
	Before sowing	BBCH 28–29	BBCH 29–30	
conv.I	80 kg (CAN)			80
CULTAN I			80 kg (UAN)	80
conv.II	80 kg (CAN)	50 kg (CAN)		130
CULTAN II			130 kg (UAN)	130
conv.S	23 kg (AS) + 57 kg (CAN)			80
CULTAN S			80 kg (UAS)	80

CAN – calcium ammonium nitrate (27 % N); UAN – urea ammonium nitrate (30 % N); AS – ammonium sulphate (20.5 % N, 24 % S); UAS – urea ammonium sulphate (19 % N, 5 % S)

Total nitrogen concentration in grain was determined by the Kjeldahl method using the Vapodest 50s (Gerhardt, Königswinter, Germany). To express protein content in grain of winter wheat and spring barley, the nitrogen concentration in grain was multiplied by the 5.7 (ČSN 46 1011-18) and 6.25 coefficient (ČSN 46 1100-5), respectively. Iron, zinc and sulphur concentrations in grain and aboveground biomass at the BBCH 28, 45 and 51 growth stages were determined by inductively coupled plasma optical emission spectroscopy ICP-OES VistaPro (Varian, Mulgrave, Australia). Nutrient harvest index was calculated as the percent of grain micronutrient content in shoot micronutrient content. Water extractable (bioavailable) sulphur in soil was determined according to Kulhánek, et al. (2011).

Statistical analysis of data was carried out using the Statistica version 9.0 (StatSoft, Tulsa, USA). Data are presented in the results at six treatments of nitrogen fertilization across the three sites and four years. Standard analysis of variance (ANOVA) procedures with the Fisher LSD test were used to calculate significant differences between individual treatments of nitrogen fertilization. A probability value of 0.05 or less ($P \leq 0.05$) was taken to be statistically significant.

Linear regression and the resulting correlation coefficients were calculated by compiling the data obtained using the all nitrogen fertilization treatments. Studied traits were analysed two by two using all possible combinations. Coefficients of correlation (r) between assessed traits are presented in correlation matrix.

Results and Discussion

No significant effect of nitrogen treatment on iron and zinc concentration in spring barley grain and on zinc concentration in winter wheat grain was recorded (Table 4). No significant differences in iron concentration in winter wheat grain between conventional and CULTAN treatment were found. Neither increased supply of nitrogen fertilizer nor sulphur containing fertilizer resulted in significant differences in iron and zinc concentration in grain of winter wheat as well as spring barley at both nitrogen nutrition systems.

Table 4. Zinc and iron concentration in grain (mg*kg^{-1}).

		conv.I	CULTAN I	conv.II	CULTAN II	conv.S	CULTAN S
Winter wheat	Fe	35.7^{ab}	33.1^{a}	38.4^{b}	34.6^{ab}	34.7^{ab}	33.8^{a}
	Zn	17.9^{a}	17.2^{a}	19.1^{a}	17.5^{a}	17.8^{a}	17.7^{a}
Spring barley	Fe	48.0^{a}	48.0^{a}	51.4^{a}	42.1^{a}	48.4^{a}	50.7^{a}
	Zn	12.4^{a}	11.8^{a}	13.4^{a}	11.3^{a}	13.0^{a}	11.9^{a}

Plant roots induce acidification of rhizosphere when ammonium as nitrogen source is applied (Ogut, et al., 2011). Thus, ammonium supply can enhance uptake of micronutrients compared to nitrate in neutral and calcareous soils (Balík, et al., 2008; Robin, et al., 2008; Elgharably, et al., 2010).

High ammonium concentration in CULTAN depots leads to inhibition of nitrification. Thus, urea, nitrate or ammonium in mineral fertilizers after their application in combination with the CULTAN system are taken up and assimilated by plants in the respective form (Sommer et Scherer, 2009). In contrast, Pfab, et al. (2012) assumed that even though ammonium concentrations in CULTAN depots are high, toxicity is insufficient for a complete inhibition of microbial activity in the surrounding of the depots. Flisch, et al. (2013) also did not confirm long-term plant nutrition intended by CULTAN system. It can be assumed that nitrate developed through nitrification under CULTAN fertilization is continuously taken up by plants, which is not contrary to the findings of Menge-Hartmann et Schittenhelm (2008) and Sedlář, et al. (2013). This can also explain no antagonism between ammonium and inorganic cations uptake by CULTAN-treated winter rape and spring barley reported by Peklová, et al. (2012) and Sedlář, et al. (2012), respectively.

Contrary to findings of Kutman, et al. (2011) and Xue, et al. (2012), a significantly positive relationship was not found between grain zinc as well as grain iron and nitrogen concentration in grain (Table 8). Sedlář, et al. (2011) and Kozlovský, et al. (2009) recorded lower protein content in grain of CULTAN-treated spring barley and winter wheat, respectively, compared to conventionally treated ones. However, this decrease in grain protein content at CULTAN treatment was not sufficient to reduction in neither zinc nor iron concentration compared to conventional treatment (Table 4).

Increase in nitrogen fertilizer supply to 200 kg*ha^{-1} at winter wheat and 130 kg N*ha^{-1} at spring barley resulted in nonsignificant differences in iron and zinc concentration which is in accordance with the findings of Xue, et al. (2012.)

No significant differences in sulphur concentration in aboveground biomass of winter wheat and spring barley between conventional and CULTAN treatment were recorded except for spring barley at the BBCH 28 growth stage where significantly higher sulphur concentration in spring barley aboveground biomass was found at conventional treatment when S-containing fertilizer was applied (Table 5).

Leaf concentration of S-containing amino acid methionine positively relates to iron acquisition (Lemanceau, et al., 2009; Aciksoz, et al., 2011b). However, no significant correlation between iron content in grain and sulphur concentration in aboveground biomass was recorded (Table 8).

Furthermore, since increased supply of nitrogen fertilizer did not lead to decrease in iron and zinc concentration in grain and because the application of sulphur containing fertilizer had no effect on zinc and iron concentration in grain either, it can be assumed that sulphur is not deficient for winter wheat and spring barley grown under conditions of the experimental sites which is in accordance with the findings of Kulhánek, et al. (2011).

Table 5. Sulphur concentration in aboveground biomass at the BBCH 28, BBCH 45 and BBCH 51 growth stages (mg*kg^{-1}).

		conv.I	CULTAN I	conv.II	CULTAN II	conv.S	CULTAN S
Winter wheat	BBCH 28	2,655^{a}	2,589^{a}	2,812^{ab}	2,919^{ab}	3,511^{c}	3,225^{bc}
	BBCH 45	1,697^{ab}	1,545^{ab}	1,747^{ab}	1,371^{a}	1,795^{b}	1,546^{ab}
	BBCH 51	1,568^{a}	1,405^{a}	1,729^{ab}	1,495^{a}	2,069^{b}	1,676^{ab}
Spring barley	BBCH 28	2,590^{a}	2,661^{a}	2,798^{a}	2,592^{a}	3,193^{b}	2,734^{a}
	BBCH 45	1,515^{a}	1,435^{a}	1,571^{a}	1,472^{a}	1,864^{b}	1,632^{ab}
	BBCH 51	1,395^{abc}	1,259^{a}	1,448^{bc}	1,350^{ab}	1,576^{c}	1,410^{abc}

Correlation coefficients between grain yield (Table 6) and grain concentration of zinc as well as iron were not significant (Table 8) which is in accordance with the findings of Murphy, et al. (2008), who state that iron and zinc are the only minerals studied whose contents are not negatively correlated with yield. Correlation coefficient between grain yield of winter wheat and zinc concentration in winter wheat grain, however, was close to level of statistical significance (Table 8).

Table 6. Grain yield at 14% moisture (t*ha^{-1})

	conv.I	CULTAN I	conv.II	CULTAN II	conv.S	CULTAN S
Winter wheat	9.20^{a}	8.70^{a}	9.31^{a}	8.91^{a}	9.33^{a}	9.01^{a}
Spring barley	6.88^{a}	6.95^{a}	6.69^{a}	7.02^{a}	6.95^{a}	7.01^{a}

No significant effect of nitrogen treatment on micronutrient harvest indexes (Table 7) was recorded. Thus, iron and zinc concentrations in grain of winter wheat and spring barley were not significantly influenced by dilution effect (Zhang, et al., 2012) at both nitrogen nutrition systems.

Table 7. Iron (FeHI) and zinc (ZnHI) harvest index.

		conv.I	CULTAN I	conv.II	CULTAN II	conv.S	CULTAN S
Winter wheat	FeHI	0.53^{a}	0.48^{a}	0.55^{a}	0.49^{a}	0.48^{a}	0.49^{a}
	ZnHI	0.84^{a}	0.83^{a}	0.84^{a}	0.85^{a}	0.82^{a}	0.84^{a}
Spring barley	FeHI	0.57^{a}	0.62^{a}	0.60^{a}	0.59^{a}	0.60^{a}	0.62^{a}
	ZnHI	0.87^{ab}	0.88^{a}	0.84^{b}	0.85^{ab}	0.86^{ab}	0.85^{ab}

Increase in nitrogen fertilizer supply to 200 kg N*ha^{-1} at winter wheat and to 130 kg N*ha^{-1} at spring barley had no significant effect on both iron and zinc concentration in grain as well as micronutrient harvest index. Because of no significantly positive effect of the increased nitrogen fertilizer supply on grain yield of winter wheat and spring barley (Table 7) the application of these high nitrogen supplies cannot be recommended under conditions of Czech Republic.

Table 8. Trait correlation

	GY	% N grain	grain Fe	grain Zn	S at BBCH 28	S at BBCH 45
Winter wheat						
% N grain	-0.01					
grain Fe	-0.09	0.13				
grain Zn	-0.45	0.22	0.38			
S at BBCH 28	0.20	0.29	-0.02	-0.22		
S at BBCH 45	0.21	0.33	0.17	-0.16	0.40	
S at BBCH 51	0.09	0.37	0.22	0.19	0.10	0.37
Spring barley						
% N grain	-0.12					
grain Fe	0.05	-0.27				
grain Zn	-0.13	-0.23	0.42			
S at BBCH 28	0.03	0.02	-0.02	0.37		
S at BBCH 45	-0.11	0.13	0.20	0.51	0.41	
S at BBCH 51	-0.20	0.06	0.22	0.42	0.31	0.74

GY – grain yield, % N grain – nitrogen concentration in grain, grain Zn – zinc concentration in grain, S at BBCH 28 – sulphur concentration in aboveground biomass at the BBCH 28 growth stage, S at BBCH 45 – sulphur concentration in aboveground biomass at the BBCH 45 growth stage, S at BBCH 51 – sulphur concentration in aboveground biomass at the BBCH 51 growth stage

Conclusions

No significant effect of nitrogen treatment on iron and zinc concentration in spring barley grain and on zinc concentration in winter wheat grain was recorded. Neither increased supply of nitrogen fertilizer nor sulphur containing fertilizer resulted in significant differences in iron and zinc concentration in grain of winter wheat as well as spring barley at both nitrogen nutrition systems. No significant differences in sulphur concentration in aboveground biomass of winter wheat and spring barley between conventional and CULTAN treatment were recorded. Iron and zinc harvest index were not influenced by the CULTAN system.

No significant correlation coefficients were recorded in relationships: iron content in grain x sulphur concentration in aboveground biomass, grain concentration of zinc x grain yield, grain concentration of iron x grain yield, grain concentration of zinc x nitrogen concentration in grain, grain concentration of iron x nitrogen concentration in grain.

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