

Possibility of selenium biofortification of winter wheat grain

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ABSTRACT

The effect of foliar application of selenium (Se) fertilizers (sodium selenite and selenate) in two different Se doses (10 and 20 g/ha) on grain yield, Se content and mechanical and technological parameters of winter wheat grain was investigated in the field fertilization experiment. Foliar spray application of Se was applied at the growth stage of the 2nd node on the main stem (32 BBCH). Selenate foliar applications in dose 10 g Se per ha showed a significant increase of grain yield in comparison to control treatment without Se application. The mechanical and technological parameters of wheat grain were not significantly affected by both selenite and selenate foliar application. The average Se concentration was significantly lower in 2007/2008 (grain yield 8.72 t/ha) than 2006/2007 (yield 6.35 t/ha) growing season (0.133 versus 0.189 mg Se/kg dry matter). The grain Se absorption efficiency at foliar application of 10 and 20 g Se/ha of selenite and selenate were 1.35–1.45% and 13.24–15.14%, respectively.

Keywords: foliar application of selenate and selenite; grain quality; selenium content in grain

Selenium (Se) is one of the elements that determine the normal functioning of an organism; it has antioxidant properties and protects the organism against the actions of free radicals and carcinogenic factors. Selenium is an element that fulfils an important physiological function (Kieliszek and Blažejak 2013), but apart from all the elements, selenium has one of the narrowest range between dietary deficiency (< 40 µg/day) and toxic levels (> 400 µg/day) (Fordyce 2007).

While there is no evidence of Se need for higher plants, several reports show that when Se added at low concentrations, it exerts beneficial effects on plant growth. Se may act as quasi-essential micronutrient through altering different physiological and biochemical traits; thus, plants vary considerably in their physiological and biochemical response to Se (El-Ramady et al. 2016).

During the last decade, studies related with strategies for Se biofortification in food plants for human nutrition have increased significantly because this metalloid is incorporated into human metabolism mainly as a constituent of food plants (Mora et al. 2015). Biofortification is defined as the process of increasing the bioavailable concentrations of essential elements in the edible portions of cultivated plants through agricultural management (fertilization) or genetic improvement (White and Brodlay 2005).

Plant-derived foodstuffs, namely cereals, are the major dietary sources of Se in most countries throughout the world, even if Se contents are strongly dependent upon the corresponding levels in cereal-growing soils. Therefore, wheat is one of the staple crops that appears as an obvious candidate for Se biofortification, considering its

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gross-tonnage production and nutritional relevance worldwide (Lyons 2010, Galinha et al. 2013).

In each country various factors such as food importation and life-style determine the selenium intake. Due to an extremely low Se intake in the 1970's in Finland, 0.025 mg/day, an official decision was made in 1984 to supplement multi-nutrient fertilizers with Se in the chemical form of sodium selenate. The latest regulation refers to liquid fertilizers, which may be applied at an amount of 10 g Se per hectare in fertilizers applied to soil and 4 g Se per hectare applied as a foliar fertilizer since 2012. The average dietary human intake increased from 0.04 mg Se/day in 1985 to a present plateau of 0.08 mg Se/day, which is well above the current nutrition recommendations. The nationwide supplementation of fertilizers with sodium selenate is shown to be effective and safe in increasing the Se intake of the whole population (Alfthan et al. 2015). Because soil, climatic and cropping conditions will affect the efficiency of Se biofortification, experience gained in Finland and elsewhere may not be applicable to other regions.

In general, there are some Se fertilization methods including soil application, foliar application and seeds treatment with Se or seeds priming as well as *in vitro* experiments. Foliar Se application is an effective method of biofortifying food crops with Se. Both selenite and selenate have been shown to be more bioavailable to plants when applied directly to leaf surfaces opposed to soils (Kápolna et al. 2009).

Therefore, the aim of the present study was to investigate the effect of Se foliar applications in the form of selenite and selenate on wheat grain yield, Se content, grain Se uptake and Se absorption efficiency. The influence of the fertilizer and its doses on the mechanical and technological parameters of wheat grain was also evaluated.

MATERIAL AND METHODS

Small-plot field experiments were established in the first decade of October in the years 2006 and 2007 at the Breeding Station of Sládkovičovo-Nový dvor (17°34'40"E, 48°22'20"W) with winter wheat (*Triticum aestivum* L.), cv. Verita. The experiments were realized on loamy degraded Chernozem. Experimental plots with size of 10 m² (8 × 1.25 m) were arranged in block pattern and repeated four times. The seeding rate represented 5 millions

of germinating grains per hectare with the span of rows amounting to 0.125 m. Pea for grain was grown as forecrop. Agrochemical soil characteristics determined before the trial establishment are stated in Table 1.

Climatic conditions were evaluated according to Kožnarová and Klabzuba (2002). The average temperatures during the two study years were 14.2°C and 13.4°C. Both study years were extraordinary warm compared to the average of 40-year period (10.5°C). The yearly sum of precipitation in 2006/2007 (468.5 mm) was lower than in 2007/2008 (713.5 mm). In comparison to 40-year period (497.2 mm), normal year 2006/2007 was followed by extraordinary wet year 2007/2008. Rainfall during the growing period (from April to June) in 2006/2007 was lower (99.0 mm) than in 2007/2008 (196.7 mm). The average over a 40-year period was 147.5 mm.

Foliar spray application of increasing doses of Se was realized at the growth stage of 2nd node on the main stem (32 BBCH) by motorized backpack sprayer on April 19, 2007 and April 21, 2008. Selenium in the form of sodium selenite (Na₂SeO₃ · 5 H₂O) and sodium selenate (Na₂SeO₄) in doses 0, 10, 20 g Se/ha was applied in both experimental years. Spraying volume per hectare was 400 L.

The harvesting of the crop was performed by small-plot combine machine at maturity (91 BBCH) on July 9, 2007 and July 18, 2008. Soil analyses and grain parameters (volume weight of grain, portion of the first class grain, thousand kernel weight, wet gluten content, sedimentation value according to Zeleny and falling number) were determined by common methods. Se content in wheat grain was determined by HG-AAS method using Varian Spectraa 300A VGA-76 apparatus (Varian, Australia).

Table 1. Agrochemical characteristics of soil (to the depth of 0.3 m) before trial establishment

Soil analyses	2006/2007	2007/2008
pH _{KCl}	7.16	7.70
N _{min} (NO ₃ ⁻ -N and NH ₄ ⁺ -N forms) (mg/kg)	18.1	24.7
P _{Mehlich III} (mg/kg)	112.9	105.2
K _{Mehlich III} (mg/kg)	388.0	357.0
Se-total content (HF + HNO ₃ + HCl) (mg/kg)	0.27	0.26
Content of C _{ox} Tiurin (%)	2.81	2.78

Table 2. Effect of different doses of both selenite and selenate foliar applications on winter wheat grain yield

Se form	Se dose (g/ha)	Grain yield (t/ha)			Relatively (%) 1 = 100
		2006/2007	2007/2008	2-year average	
0		6.03 ^a	8.61 ^a	7.32 ^a	100.0
Selenite	10	6.30 ^a	8.73 ^{ab}	7.52 ^{ab}	102.7
	20	6.43 ^a	8.75 ^{ab}	7.59 ^{ab}	103.7
Selenate	10	6.58 ^a	8.72 ^{ab}	7.65 ^b	104.5
	20	6.40 ^a	8.79 ^b	7.59 ^{ab}	103.7
Average for year		6.35 ^a	8.72 ^b		

Means followed by different letters indicate significant differences between treatments at $P < 0.05$

Grain selenium absorption efficiency (%) was calculated as: $100 \times [(mg \text{ Se absorbed on Se treatments} - mg \text{ Se absorbed on control treatments}) / mg \text{ of applied Se}]$. Grain Se uptake (mg/ha) was calculated as grain Se content (mg/kg) \times grain yield (kg/ha).

The obtained data were analysed using the Statgraphics Plus statistical software (Rockville, USA). A multifactor ANOVA model was used for individual treatment comparisons at $P = 0.05$, with separation of the means by the *LSD* multiple-range test.

RESULTS AND DISCUSSION

Two-year average results of selenite foliar applications at the growth stage of 2nd node on the main stem (32 BBCH) showed no significant effect on winter wheat grain yields, while selenate supply (10 g Se/ha) resulted in a significant increase of grain yield (Table 2). Average yield of grain fluctuated from 6.30–6.58 t/ha in 2006/2007, and from 8.72–8.79 t/ha in 2007/2008. Different weather conditions significantly influenced the wheat grain yield in the respective experimental years, when the average of all treatments was 6.35 t/ha

in 2006/2007, and 8.72 t/ha in 2007/2008. Several field experiments reported no significant effect of Se application on the yield in winter wheat (Broadley et al. 2010), yield of rapeseed (Seppänen et al. 2010) and maize grain yield (Chilimba et al. 2012, Wang et al. 2013). However, several studies showed that Se application positively affected the plant. Se application significantly increased wheat grain yield at regreening-jointing, jointing-heading, and heading-blooming stages, however, yield was not significantly affected by Se application at blooming-filling stage (Chu et al. 2013). A positive effect of 5 g Se/ha for foliar spray on grain yield of common buckwheat was reported (Jiang et al. 2015). Selenium at low concentrations acts as a growth stimulant whereas at high concentrations, it reduces root elongation and biomass production of wheat plants (Guerrero et al. 2014).

Two-year average results of volume weight of grain, portion of the first class grain, thousand kernel weight, wet gluten content, Zeleny sedimentation value and falling number of winter wheat grain was not significantly affected by different doses (10 and 20 g Se/ha) of either selenite or selenate foliar application (Tables 3 and 4). No significant

Table 3. Effect of different doses of both selenite and selenate foliar applications on volume weight of grain, portion of the first class grain and thousand kernel weight (TKW) of winter wheat grain (averages of the years 2006/2007 and 2007/2008)

Se form	Se dose (g/ha)	Volume weight of grain (g/L)	Portion of the first class grain (%)	Thousand kernel weight (g)
0		773.0 ^a	87.1 ^a	44.3 ^a
Selenite	10	778.7 ^a	88.7 ^a	45.1 ^a
	20	776.0 ^a	88.4 ^a	45.7 ^a
Selenate	10	775.6 ^a	88.8 ^a	45.6 ^a
	20	774.1 ^a	89.7 ^a	45.2 ^a

Means in the column marked by the same letters do not differ significantly at $P < 0.05$

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Table 4. Effect of different doses of both selenite and selenate foliar applications on wet gluten content, Zeleny sedimentation value and falling number of winter wheat grain (averages of the years 2006/2007 and 2007/2008)

Se form	Se dose (g/ha)	Wet gluten (%)	Zeleny sedimentation value (mL)	Falling number (s)
0		30.8 ^a	34.3 ^a	288 ^a
Selenite	10	31.7 ^a	33.8 ^a	281 ^a
	20	30.5 ^a	36.3 ^a	268 ^a
Selenate	10	28.1 ^a	34.5 ^a	286 ^a
	20	28.3 ^a	35.5 ^a	242 ^a

Means in the column marked by the same letters do not differ significantly at $P < 0.05$

effect of increasing doses of foliar-applied selenite and selenate on grain protein content and hectolitre weight of durum wheat recorded Poblaciones et al. (2014), but on the other hand, they found a significant lower thousand kernel weight after application of 20 and 40 g of selenium per hectare in comparison to a treatment without application of selenium.

The total Se contained in grain was significantly affected by all the three factors; year, Se form and Se dose (Table 5). The Se concentration in grain was much greater in growing season 2006/2007 than in 2007/2008 (on average 0.189 and 0.133 mg/kg dry matter, respectively). It may be possible that the dilution effect was responsible for the higher yield in 2007/2008. The average Se concentration was significantly lower in irrigated (grain yield 9.1 t/ha) than dryland (yield 7.4 t/ha) wheat (0.17 versus 0.25 mg Se/kg), possibly owing to yield dilution (Curtin et al. 2008).

The Se content in the wheat grain in 2006/2007 ranged between 0.030 mg/kg dry matter at dose 0 and 0.092 or 0.540 mg/kg dry matter at 20 g/ha of selenite and selenate, respectively. In 2007/2008 the range was between 0.033 mg/kg and 0.052 or 0.350 mg/kg dry matter, respectively. Results show that foliar selenite and selenate additions can increase Se contents in mature wheat grains up to 3 and 18 times, compared to selenium non-supplemented crops. As Curtin et al. (2006) found, selenium content in wheat grain increased from 0.03 mg/kg in the control samples to 0.45 mg/kg when Se was added as selenate at 20 g/ha by using foliar spray.

Although the Se uptake was linear for both Se forms, the increases were much greater at sodium selenate treatment than at sodium selenite treatment. The grain Se absorption efficiency in wheat at foliar application doses of 10 and 20 g Se/ha as selenite and selenate were 1.35% or 1.45% and 13.24% or 15.14%, respectively (Table 6). If 1 g of Se/ha as selenite was applied, 9.3% of this amount was transited to wheat grain. This transition represented 2.4% and 2.9% of total applied Se when the doses of 10 and 20 g Se/ha, were applied, respectively (Ducsay and Ložek 2006).

The present study indicates that application of Se fertilizer at growth stage 32 BBCH was effective in increasing wheat Se concentrations. Se accumulation in wheat grain was closely associated with the application dose and form of Se. The appropriate Se application in wheat might prompt grain yield improvement. It is important to state that sodium selenate was much more effective than sodium selenite. It can be expected that for the fully-soluble Se source (sodium selenate), an application rate of 5 g Se/ha should raise grain Se content to 0.1 mg/kg dry matter.

Table 5. Effect of different doses of both selenite and selenate foliar applications on the selenium (Se) content in winter wheat grain

Se form	Se dose (g/ha)	Se content in wheat grain (mg/kg dry matter)			Relatively (%) 1 = 100
		2006/2007	2007/2008	2-year average	
0		0.030 ^a	0.033 ^a	0.032 ^a	100.0
Selenite	10	0.064 ^a	0.038 ^a	0.051 ^a	159.4
	20	0.092 ^a	0.052 ^a	0.072 ^a	225.0
Selenate	10	0.220 ^b	0.190 ^b	0.205 ^b	640.6
	20	0.540 ^c	0.350 ^c	0.445 ^c	1390.6
Average for year		0.189 ^a	0.133 ^b		

Means followed by different letters indicate significant differences between treatments at $P < 0.05$

Table 6. Effect of different doses of both selenite and selenate foliar applications on the selenium (Se) uptake and absorption efficiency by winter wheat grain (average of the years 2006/2007 and 2007/2008)

Se form	Se dose (g/ha)	Grain Se uptake (mg/ha)	Grain Se absorption efficiency (%)
0		234 ^a	
Selenite	10	369 ^a	1.35 ^a
	20	523 ^a	1.45 ^a
Selenate	10	1558 ^b	13.24 ^b
	20	3262 ^c	15.14 ^b

Means followed by different letters indicate significant differences between treatments at $P < 0.05$

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