

SELENIUM TREATMENT UNDER FIELD CONDITIONS AFFECTS MINERAL NUTRITION, YIELD AND ANTIOXIDANT PROPERTIES OF BULB ONION (*Allium cepa* L.)

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Abstract. The aim of the research was to find out how foliar selenium (Se) treatment in open field conditions affects plant nutrition, yield and bulb bioactive properties. 10, 50 or 100 $\mu\text{g}\cdot\text{mL}^{-1}$ Se, referred to as Se10, Se50 and Se100 treatments, was applied to onion ‘Hercules’ in 2008 and 2009. Bulb weight, total yield, content of total Se, free selenomethionine and selenomethylselenocysteine, total S, N, P, K, Ca and Mg, content of total phenolics, pungency and total antioxidant capacity (TAC) were determined. All Se treatments significantly reduced bulb S content. The Se100 treatment had a tendency to decrease bulb size and yield. The Se50 treatment increased total Se content, total phenolics and TAC and had a tendency to increase the yield. A larger proportion of total Se was converted into organic compounds in the Se50 than in the Se100 treatment. In Se 10 treatment, bulb TAC was the highest among treatments in 2009. Considering both agronomic and human health benefits, Na_2SeO_4 solution at the rate of 50 $\mu\text{g}\cdot\text{mL}^{-1}$ Se can be recommended for bulb onion.

Key words: selenoamino acids, sulfur, total phenolics, pungency, antioxidant capacity

INTRODUCTION

Selenium (Se) is an essential micronutrient needed to prevent oxidative damage and to support hormone balance in human and animal cells [Terry et al. 2000, Fairweather-Tait et al. 2011]. Low Se status in humans may increase the risk of cardiovascular diseases (CVD), cancer and other diseases, which are caused by free radicals [Rayman 2000, Fairweather-Tait et al. 2011]. CVD is the most common cause of death in most

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European countries [Rayner et al. 2009]. In Estonia, CVD is also the main reason for early loss of work capacity (at an age below 65) and death, and there has been no significant change for the better over the past 20 years [National Strategy for Prevention of Cardiovascular Diseases 2005–2020].

Plants play a key role in cycling Se from soil to animals and humans [Turakainen et al. 2005]. In some regions of the world soils are Se-deficient, which makes it impossible for plants to accumulate this element and in these regions local feed and food is also Se-deficient [White and Broadley 2009]. The region around the Baltic Sea is known for Se-deficient soils [Reimann et al. 2003]. Very low Se contents in serum and human milk in Estonia have been reported [Kantola et al. 1997]. Low Se uptake by the population due to Se-deficient soils is also a problem in other parts of the European Union [Fairweather-Tait et al. 2011], like the Slovak Republic [Ducsay and Ložek 2006], Croatia [Klapec et al. 1998], Canary islands [Romero et al. 2001] and the United Kingdom [Broadley et al. 2006]. Thus, remedies for increasing Se intake by the human population in many regions in Europe are required.

In Finland, systematic supplementation of agricultural fertilizers with Se has been carried out since 1984 and it has proven to be a safe and effective means of increasing the Se intake of both animals and humans [Aro et al. 1995, Broadley et al. 2006]. Allium species have the capability to accumulate high Se levels [Shah et al. 2004]. Se bioavailability depends on the conversion of absorbed Se into a biologically active form [Tapiero et al. 2003]. Major Se species found in selenized onions are the methylated forms of selenium, like Se-methylselenocysteine and γ -glutamyl-Se-methylselenocysteine [Kápolna and Fodor 2006]. Methylated Se species enter the methylated pool of Se directly by being transformed into methylselenol (CH_3SeH) by β -lyase. As methylselenol has been recognized as an anti-cancer compound, the consumption of foodstuff containing methylselenocysteine (MeSeCys), the main precursor of CH_3SeH , is generally recommended [Thiry et al. 2012].

Bulb onion (*Allium cepa* L.) is one of the major sources of dietary flavonoids in several countries [Hertog et al. 1993, Knekt et al. 1996]. Due to the substantial consumption and ability to accumulate Se, bulb onion could be one of the best vegetables for Se enrichment in Europe. Several studies have been performed to enrich onions with Se, but most of them have been conducted in hydroponic conditions [Kopsell and Randle 1999, Wróbel et al. 2004, Kápolna and Fodor 2006, Montes-Bayón et al. 2006]. The present research is one of the few studies to report responses of onion to Se nutrition in the field, considering the effect on both agronomic and biochemical characteristics.

The experiment described here was carried out in order to test the following hypotheses: 1) Se – content of onion bulbs can be significantly increased using sodium selenate (Na_2SeO_4) foliar fertilization in open field conditions, 2) Se – enrichment could affect plant nutrition, yield, content of bioactive compounds and antioxidant capacity of bulb onion.

MATERIAL AND METHODS

Onion cultivation and Se treatments. Experiments were carried out in 2008 and 2009 in East Estonia on a Glosic Hapludalf soil. In both years pungent yellow onion

cv. 'Hercules' F1 was planted from sets in the first week of May (03. May 2008, 07. May 2009) in two lines per furrow with plant distance within the row 6 cm, the space between lines was 10 cm and between furrows 65 cm (51 sets per m²). Basic fertilization was performed in the last week of April with NPK fertilizer (50 kg·ha⁻¹ N, 22 kg·ha⁻¹ P, 83 kg·ha⁻¹ K) and followed by top dressing in the beginning of June with ammonium saltpeter (ammonium nitrate) (60 kg·ha⁻¹ N). No irrigation was used in experimental fields during the growing seasons.

When onion plants had 7–8 leaves (20 June 2008 and 24 June 2009) Na₂SeO₄ solution was sprayed at concentrations of 0, 10, 50 or 100 µg Se·mL⁻¹ at a rate of 50 mL·m⁻². The trial was set up in a randomized complete block design with four replicate plots per treatment, where each plot had an area of 20 m². Na₂SeO₄ solution was sprayed with a hand sprayer. For each plot the sprayer was filled with 1 L of solution, which was evenly sprayed over the area. For determination of yield characteristics onion bulbs from the middle part of each treatment plot were harvested in the last week of August, considering that the right harvest time is when 70–80% of onions have tops down. The harvested area was 6 m² and the number of harvested bulbs ranged from 275 to 290. After drying and curing in a well ventilated room at 20–25°C for two weeks, total yield and bulb weight were determined.

Soil and weather conditions. Soil P, K, Ca and Mg were determined by the ammonium lactate (AL) method [Van Erp et al. 2001] and the Kjeldahl method was used for N determination. Plant available S was determined as SO₄²⁻-S nephelometrically from water extracts [Van Ranst et al. 1999]. Total Se content of soil was determined by the hydride-generation atomic absorption spectrometric (HG-AAS) method [Malbe et al. 2010]. The phosphorus content of the upper soil layer was very high, and the potassium, magnesium, calcium and sulfur contents were medium (tab. 1). Se content in soil from the experimental area ranged from 45–51 µg·kg⁻¹. According to Tan et al. [2002] the marginal concentration of Se in soil is 123–175 µg·kg⁻¹, which indicates that the Se concentration in our experimental area was very low.

Table 1. The content of total organic N and plant available mineral nutrients in the 0–30 cm soil layer of the onion experimental field in Estonia

Soil properties	2008	2009
pH _{KCl}	5.8	6.1
Nitrogen, %	0.16	0.20
Phosphorus, mg·kg ⁻¹	172	193
Potassium, mg·kg ⁻¹	197	175
Calcium, mg·kg ⁻¹	977	1039
Magnesium, mg·kg ⁻¹	110	116
Sulfur, mg·kg ⁻¹	19.5	22.2
Selenium, µg·kg ⁻¹	45.5	50.8

Generally, the weather conditions were suitable for onion production in both experimental years, although in 2008 July and August had relatively high temperatures with rain, which favored spread of downy mildew (*Peronospora destructor*) and Botrytis-mould, while in the first half of the 2009 growing period the monthly precipitation was only 40% of the 30 year average.

Onion sample preparation and determination of mineral elements. Ten onion bulbs were randomly selected from every replicate plot. Altogether 40 bulbs per treatment were used. For the Se analysis a 0.2–0.5 g homogenized sample of onion bulbs was mineralized by 5 mL of concentrated HNO₃ and 2 mL of concentrated H₂O₂ in PTFE tubes in a microwave oven at temperatures of up to 180°C for 30 min. Total Se content from mineralized plant material was determined by the electrothermal- (ET-AAS) method [Viitak and Volynsky 2006]. The bulb N concentration of air-dried samples was determined by the Kjeldahl method [Benton 2001]. P, Mg and Ca concentrations were measured spectrometrically, and K was determined flame-photometrically [Moor et al. 2006]. Total S from plant material was determined by the Dumas combustion method [Benton 2001].

Determination of selenoamino acids. Free selenoamino acids (SeMet and Se-MeSeCys) were determined in the second experimental year (2009) at the Institute of Chemistry, University of Tartu. The content of SeMet and Se-MeSeCys in onions was determined by LC-ESI-MS/MS following pre-column derivatization with Deemm as described by Rebane et al. [2011]. Average recoveries were 84.6% for SeMet and 101.7% for Se-MeSeCys.

Determination of bioactive compounds and antioxidant capacity. The onion bulbs were peeled, cut into small pieces and the different extraction solutions were added immediately. For determination of total polyphenols and antioxidant capacity, a water extract 1:10 (w/v) was prepared, 5 g sample material was weighed into a plastic jar with snap cap and 5 mL water was added. The mixture was homogenized with a Polytron homogenizer (model PT 1600 E, Kinematica AG, Lucern, Switzerland) for 2 minutes at 30000 rpm, 45 mL water was added and the suspension was shaken on a reciprocating shaker for 30 min. Filtered extract was used for determination of total polyphenols and antioxidant capacity.

Total polyphenol content was determined using the Folin-Ciocalteu method with some modification. Antioxidant capacity determination was based on the ABTS method with slight modifications. The pungency was determined according Schwimmer and Weston [1961] and reported as pyruvic acid content. 10 g of fresh onion was sliced and crushed into 20 mL water for enzymatic pyruvic acid content and into 20 mL 20% (w/v) trichloroacetic acid solution for background pyruvic acid content determination and homogenized. Detailed descriptions of analytical procedures have been reported previously by Põldma et al. [2011].

Statistical analysis. All measurements were carried out on three or four parallel samples in each repeat. The data were evaluated by two-way analysis of variance (ANOVA) and the means were compared by a least significant difference (LSD) test at a 5% probability level using Statistica for Windows version 10.0 (StatSoft, Inc., Tulsa, OK, USA).

RESULTS AND DISCUSSION

Total selenium content. In the current experiment the total Se content of onion bulbs ranged from 4 to 127 $\mu\text{g}\cdot\text{kg}^{-1}$ FW (fig. 1). The average Se content of control onions was 18 $\mu\text{g}\cdot\text{kg}^{-1}$ FW. Earlier experiments in Tenerife have shown that in six onion cultivars grown without Se enrichment, the Se content ranged from 0.44 to 0.97 $\mu\text{g}\cdot\text{kg}^{-1}$ FW [Galdón et al. 2008]. Thus, the Se content in our control onions was higher than in onions from Tenerife. The Canary Islands have also been reported to have Se-deficient soils [Romero et al. 2001].

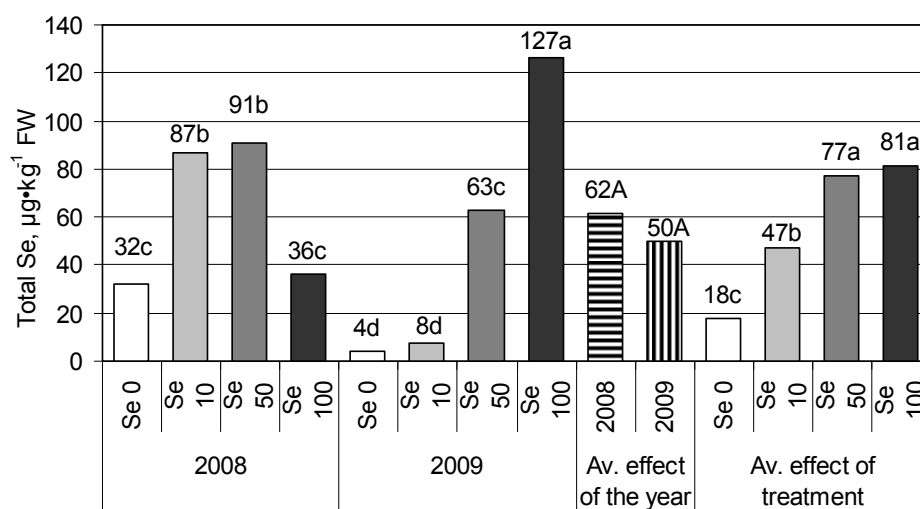


Fig. 1. Total selenium content ($\mu\text{g}\cdot\text{kg}^{-1}$ FW) of onion 'Hercules' bulbs as affected by selenium treatments in 2008 and 2009 in Estonia. Na_2SeO_4 solution was sprayed at concentrations of 0, 10, 50 or 100 $\mu\text{g}\cdot\text{mL}^{-1}$ Se at a rate of 50 $\text{mL}\cdot\text{m}^{-2}$. Mean values followed by the same letter are not significantly different at $P \leq 0.05$

In 2008, the onion bulb total Se content was significantly increased by Se10 and Se50 treatments and in 2009 by Se50 and Se100 treatments. The average effect of the Se treatment showed that all Se rates significantly increased the content of total Se compared to the control, and Se50 and Se100 were more effective than the Se10 treatment.

We were able to increase onion bulb total Se content three-fold in 2008 and by 30-fold in 2009. If we aim to supply humans with Se only by consumption of selenized onions, the Se content in our experiment is still very low. Considering that for adults the recommended daily intake of Se is 50–70 μg , it would be unrealistic to achieve this amount only by eating Se-enriched onions. Kopsell and Randle [1997] reported that onion bulb Se accumulation differed with Na_2SeO_4 application rate and among cultivars.

Thus, selecting cultivars with higher Se accumulation ability would be one solution to the problem. Another theoretical possibility would be increasing Se rates in the fertilizer solution. Wróbel et al. [2004] conducted an experiment, in which onions were grown hydroponically and Se was added to the nutrient solution to increase Se content in bulbs to $51 \pm 1.8 \mu\text{g}\cdot\text{g}^{-1}$ DM, which is 50 times higher than in our experiment. It indicates that hydroponically grown onions might be able to assimilate larger amounts of Se than by one-time foliar fertilization. However, we have to bear in mind that Se can cause toxicity in plants [Terry et al. 2000, White et al. 2004]. In an earlier mentioned study [Wróbel et al. 2004] it was observed that Se present in the growing medium caused inhibition of the root growth and growth of leaves was also inhibited by about 50% with respect to the control plants. Total yield was not recorded in the mentioned study, but most probably the yield was also negatively affected. If we aim to apply Se fertilizers in the field, the effect of Se enrichment on onion yield and its quality is important, since farmers need convincing arguments for justifying extra work and expenses.

Onion yield and bulb weight. In 2008 the average total yield in our experiment was $3.5 \text{ kg}\cdot\text{m}^{-2}$, which was significantly higher than in the following year ($3.2 \text{ kg}\cdot\text{m}^{-2}$) (tab. 2). The effect of Se treatment on the yield was not significant, although the Se50 treatment had a notable tendency to increase the yield. As an average of two experimental years, the Se50 treatment resulted in a significantly higher yield compared to the control. The Se50 treatment also increased bulb weight significantly in 2008 (tab. 2). Even though the differences in bulb size and yield from the Se50 and Se100 treatments were not statistically significant, the clear tendency towards bulb size and yield decrease in the Se100 treatment was evident compared to the Se50 treatment. This tendency could be interpreted as the first signs of Se toxicity. Therefore using higher rates of Se in foliar fertilization could be counter productive. Earlier studies have shown that only at low concentrations can Se promote growth of lettuce plants [Hartikainen et al. 1997] and rice seedlings [Wang et al. 2012].

Table 2. Total yield ($\text{kg}\cdot\text{m}^{-2}$) and fresh weight (g) of onion ‘Hercules’ as affected by selenium treatments in 2008 and 2009 in Estonia

		Selenium treatment ^a				mean
		Se0	Se10	Se50	Se100	
Total yield	2008	3.4 ab	3.5 ab	3.7 a	3.5 ab	3.5 A
	2009	3.0 b	3.2 ab	3.5 ab	3.2 ab	3.2 B
	mean	3.2 b	3.4 ab	3.6 a	3.4 ab	
Average bulb weight	2008	70 b	76 ab	85 a	79 ab	78 A
	2009	68 b	74 ab	77 ab	74 ab	73 A
	mean	69 b	75 ab	81 a	77 ab	

^a Na_2SeO_4 solution was sprayed at concentrations of 0, 10, 50 or $100 \mu\text{g}\cdot\text{mL}^{-1}$ Se at a rate of $50 \text{ mL}\cdot\text{m}^{-2}$. Mean values followed by the same letter are not significantly different at $P \leq 0.05$

Seleno-organic compounds. In 2009, the content of selenoamino acids in the control and Se10 treatment was below the limit of detection. In the Se50 and Se100 treatments both SeMet and Se-MeSeCys were detected in onion bulbs, but the content of mentioned organic compounds was not affected by the Se treatment. In Se50 treatment the content of SeMet was $20.9 \mu\text{g}\cdot\text{kg}^{-1}$ FW and the content of Se-MeSeCys was $89.8 \mu\text{g}\cdot\text{kg}^{-1}$ FW (tab. 3). Although the content of total Se was almost double in Se100 compared to Se50 treatment, the amount of organic Se compounds was quite similar: SeMet 18.5 and Se-MeSeCys $91.1 \mu\text{g}\cdot\text{kg}^{-1}$ FW. Thus, in the Se50 treatment 74.6% and in the Se100 treatment 36.7% of total Se was incorporated into SeMet and Se-MeSeCys.

Table 3. The content of total Se and two free selenoamino acids [selenomethionine (SeMet) and selenomethylselenocysteine (Se-MeSeCys)] in onion 'Hercules' bulbs as affected by selenium treatments in 2009 in Estonia

	Selenium treatment ^a	
	Se50	Se100
Total Se, $\mu\text{g}\cdot\text{kg}^{-1}$ FW	63 b	127 a
SeMet, $\mu\text{g}\cdot\text{kg}^{-1}$ FW	20.9 a	18.5 a
Se-MeSeCys, $\mu\text{g}\cdot\text{kg}^{-1}$ FW	89.8 a	91.1 a
% of Se incorporated in SeMet and Se-MeSeCys	74.6 a	36.7 b

^aNote: see table 2

Substitution of Se-amino acids into S-proteins can alter the tertiary structure of Se-substituted proteins and cause changes in catalytic activity [Brown and Shrift 1982]. These catalytic changes are considered to be a cause of Se toxicity in plants [Kopsell and Randle 1999, White et al. 2004]. It seems like onion plants might protect themselves against toxicity by limiting the amount of Se incorporated into amino acids.

In both treatments the content of Se-MeSeCys was about five to six times higher compared to the content of SeMet. Tapiero et al. [2003] have stated that plants convert Se mainly into Se-methionine and incorporate it into protein in place of methionine. SeMet can account for > 50% of the total Se content of the plant, whereas selenocysteine, methylselenocysteine and γ -glutamyl-Se-methyl-cysteine are not significantly incorporated into plant proteins. Major Se species found in selenized onions are Se-methylselenocysteine and γ -glutamyl-Se-methylselenocysteine [Kápolna and Fodor 2006]. The speciation results obtained in Se-enriched garlic indicated that the contribution of Se-MeSeCys increased with the increasing total Se content [Ip et al. 2000]. Our results indicated that the response of selenized onions could be similar: in onions treated with Se50, the Se-Met: Se-MeSeCys ratio was 1:4 and in Se100 treatment 1:5. From the point of view of human health it makes selenized Alliums especially valuable plants. It has been reported by Ip and Ganther [1990] that Se-Met is excellent for increasing the Se concentrations in tissues, but is relatively ineffective for suppression of carcinogene-

sis. In contrast, partially methylated forms of selenium may be directly involved in the anticarcinogenic action of selenium.

Concentration of macronutrients in onion bulbs. Concentration of all elements measured were comparable with previously published data. Onion N content in our experiment was similar to that in onion ‘Giza 20’ [Abd El-Samad et al. 2011], P content was similar to that in onions ‘Guayonje’ and ‘Carrizal Bajo’ [Galdón et al. 2008], K and Mg content to that in onion ‘Ailsa Craig’ [Chope and Terry 2009]. The Ca concentration in our onions was higher than reported by Galdón et al. [2008] and Rodrigues et al. [2003], but comparable to the values sited in the USDA database for unspecified onion cultivars [US Department of Agriculture 2011].

Table 4. Onion ‘Hercules’ bulb elemental composition (mg 100 g⁻¹ FW) as affected by selenium treatments in 2008 and 2009 in Estonia

		Selenium treatment ^a				mean
		Se0	Se10	Se50	Se100	
N	2008	202 b	198 b	218 a	194 b	203 A
	2009	195 b	193 b	209 a	192 b	197 A
	mean	198 b	195 b	213 a	193 b	
P	2008	35.8 b	35.8 b	37.4 ab	35.4 b	36.1 B
	2009	38.6 a	37.8 ab	36.8 ab	37.4 ab	37.7 A
	mean	37.2 a	36.8 a	37.1 a	36.4 a	
K	2008	211 ab	200 b	212 a	207 ab	207 A
	2009	175 c	173 c	173 c	181 c	175 B
	mean	193 ab	186 b	192 ab	194 a	
Ca	2008	37 a	33 b	35 ab	34 b	35 A
	2009	21 cd	16 e	19 d	22 c	19 B
	mean	29 a	25 c	27 b	28 ab	
Mg	2008	8.0 d	8.6 cd	9.3 c	8.0 d	8.8 B
	2009	17.6 b	17.5 b	18.8 a	16.8 b	17.7 A
	mean	12.8 b	13.1 b	14.0 a	12.4 b	

^aNote: see table 2

Onion bulb mineral composition was somewhat affected by Se treatments, but to a much lesser extent than Se fertilization affected garlic in our earlier experiments [Põldma et al. 2011]. In contrast to the study on garlic, no consistent decreasing effect of Se fertilization on bulb macronutrient concentration was found. Only the content of Ca was decreased by the Se10 and Se100 treatments in 2008 and by the Se10 treatment in 2009. Galdón et al. [2008] found that Ca had a negative, although insignificant correlation with Se in onion bulbs. The same authors found a positive correlation between Se and Na, K, Mg and Fe content in onions. In our experiment, the content of P and K was

affected by experimental year rather than by Se fertilization, however, N and Mg content was significantly affected by Se-fertilization. In both years bulbs from Se50 treatment had the highest content of N and Mg compared to other treatments (tab. 4). N and Mg are extremely important in chlorophyll formation and photosynthesis. Moldovan et al. [2009] have found that Se fertilization increased chlorophyll a, chlorophyll b and total chlorophyll content in onion leaves. These findings indicate that at certain rates, Se might be beneficial for photosynthesis, which was probably related to the yield and bulb size increase in the Se50 treatment in our experiment. Germ et al. [2007] found that in potato, Se induced higher terminal electron transport system activity 4 weeks after treatment. The same authors have argued that this was due to Se increasing plant metabolism, or plants needing energy to repair damage caused by Se compounds. Since in our experiment the yield and bulb weight tended to be highest in the Se50 treatment, and bulb N and Mg content was significantly increased in Se50 treatment, we support the theory that in optimal rates Se stimulates plant metabolism. Ekelund and Danilov [2001] showed that Se had no effect on *Euglena* photosynthesis and respiration when Se was the only factor, but a 24-h exposure to both Se and UV resulted in an increase in both processes. It was concluded that Se was involved in the formation and/or activation of protective compounds that increased the rate of photosynthesis.

Sulfur content and bulb pungency. Onion bulb sulfur content in our experiment ranged from 15.8 to 24.0 mg·100 g⁻¹ FW (tab. 5). Rodrigues et al. [2003] have reported bulb S content to be 66.42 mg·100 g⁻¹ FW in red onions and 40.89 mg·100 g⁻¹ FW in white onions. Randle et al. [1999] studied onion bulb S accumulation in response to increasing S-fertility, according to mentioned results, the total S content of 'Southport White Globe' onions at the lowest S fertility was 0.18% dry matter (DM). Considering that onion DM content in the mentioned study ranged from 12 to 14%, it gives us S content to be 20–25 mg·100 g⁻¹ FW. Thus, the S content in our experimental onions was similar to the onions grown in low S fertility.

In the first experimental year all three Se treatments significantly reduced onion bulb S content (tab. 5). In the second experimental year Se50 treatment had no effect on S content, but Se10 and Se100 treatments had a decreasing effect on bulb S. As an average of two years, all three Se treatments significantly reduced bulb S content. Similar results were obtained with Se-enriched garlic, where Se treatment also decreased bulb S content [Pöldma et al. 2011]. The close chemical similarity of S and Se allow for antagonistic behavior between the two elements [Kopsell and Randle 1997, White et al. 2004, 2007]. Onions are primarily consumed for their flavor, which is dominated by S compounds [Kopsell and Randle 1997]. This leads to the question, whether the decrease in S content had a negative effect on onion flavor.

In the current experiment onion bulb pungency ranged from 4.8 to 11.3 μmol of pyruvic acid·g⁻¹ FW (tab. 5). Our results are comparable to the data reported by Kopsell and Randle [1997], where pungency of 16 onion cultivars ranged from 3.8 to 11.8 μmol·g⁻¹ FW.

In the first experimental year all three Se treatments increased onion pungency compared to the control. In the following year no significant increase of pungency due to Se fertilization was observed. Kopsell and Randle [1999] have demonstrated that Se treatment caused changes in the flavor precursor profile in onions, however, among four

studied cultivars, pungency decrease was significant only in ‘Sweet Success’ onions. In our experiment, Se treatments reduced bulb S content in most of the treatments, but S decrease did not correlate with pungency decrease. Randle et al. [1995] have demonstrated that onion flavor precursor biosynthesis was a strong sink for sulfur even in sulfur deficiency. Se treatment in our experiment decreased bulb S content, but probably not to such an extent to influence bulb pungency. The methodological aspects should also be considered at this point. Onion pungency is estimated by amount of pyruvic acid, which is produced stoichiometrically from flavor precursor S-alk(en)yl-L-cystein [Anthon and Barrett 2003]. In case of substitution of S with Se in alk(en)yl-L-cystein, the amount of produced pyruvic acid from one molecule of alk(en)yl-L-cystein will be the same. Therefore, the changes in pungency may be dependent only on the content of alk(en)yl-L-cystein in onion, but not from the ratio of S alk(en)yl-L-cystein and Se alk(en)yl-L-cystein in onion.

Table 5. Onion ‘Hercules’ bulb content of sulfur ($\text{mg} \cdot 100 \text{ g}^{-1}$ FW), total phenolics (mg of gallic acid (GA) 100 g^{-1} FW), pungency (μmol of pyruvic acid $\cdot \text{g}^{-1}$ FW) and antioxidant capacity (mg Trolox equivalent (TE) g^{-1}) as affected by selenium treatments in 2008 and 2009 in Estonia

		Selenium treatment ^a				mean
		Se0	Se10	Se50	Se100	
Sulfur $\text{mg} \cdot 100 \text{ g}^{-1}$ FW	2008	24.0 a	20.1 de	20.8 cd	22.2 bc	21.8 A
	2009	23.1 ab	19.0 e	23.6 ab	15.8 f	20.4 B
	mean	23.6 a	19.6 c	22.2 b	19.0 c	
Total phenolics mg of GA $\cdot 100 \text{ g}^{-1}$ FW	2008	121 bc	126 bc	152 a	126 bc	131 A
	2009	114 c	138 ab	130 b	123 bc	126 A
	mean	118 c	132 ab	141 a	125 bc	
Pungency, μmol of pyruvic acid $\cdot \text{g}^{-1}$ FW	2008	7.5 c	11.1 a	11.3 a	10.1 b	10.0 A
	2009	6.0 d	6.5 d	5.1 e	4.8 e	5.6 B
	mean	6.7 c	8.8 a	8.2 a	7.4 b	
Antioxidant capacity mg TE g^{-1}	2008	73 d	67 cd	77 b	74 bc	69 B
	2009	64 d	85 a	76 b	76 b	75 A
	mean	62 b	76 a	77 a	75 a	

^aNote: see table 2

Antioxidant properties. In 2008, content of total phenolics in onion bulbs ranged from 121 to 152 mg of GA $\cdot 100 \text{ g}^{-1}$ FW and in 2009 from 114 to 138 mg of GA $\cdot 100 \text{ g}^{-1}$ FW (tab. 5). Sellappan et al. [2002] have reported total polyphenol content in white onions to be 7.3 mg of GA $\cdot \text{g}^{-1}$ of DM. Considering that average DM content of our experimental onions was 11%, the content of total phenolics in our onions was higher. Se50 treatment increased total phenolic content significantly in both years and Se10 treatment in 2009, whereas the greatest increase (26%) was caused by Se50 treatment in 2008.

Onion bulbs fertilized with the highest rate of Se did not contain more total phenolics than unfertilized control bulbs in either of the experimental years.

Total antioxidant capacity (TAC) of onion bulbs in the current experiment was affected by Se treatment in both years (tab. 5). In 2008, the Se50 and Se100 treatments increased TAC significantly, and Se10 treatment showed a tendency to increase TAC. In 2009, onion bulb antioxidant capacity was significantly higher in all Se-treated onions, whereas bulbs with the highest TAC were obtained from Se10 treatment. It has been proposed by Turakainen et al. [2005] that as in humans and animals, Se strengthens the capacity of plants to counteract the oxidative stress caused by oxygen radicals produced by internal metabolic or external factors. Our previous study with garlic proved that foliar Se fertilization at rates of 10–50 $\mu\text{g Se}\cdot\text{mL}^{-1}$ increased bulb antioxidant capacity [Põldma et al. 2011]. It appeared from the current experiment that for onions, the largest increase in TAC also occurred when lower rates of Se were used. This finding further supports our earlier assumption, that at lower rates Se activates certain antioxidant reactions in plants, but this is not a plant's response to Se toxicity. Otherwise we would have seen the highest bulb TAC in Se100 treatment in 2009, where Se content was almost double compared to the Se50, but bulb TAC remained at the same level.

CONCLUSIONS

The current study demonstrated that it is possible to increase bulb onion Se content significantly in field conditions by foliar Se fertilization. Considering both agronomic and human health benefits, Na_2SeO_4 solution at the rate of 50 $\mu\text{g}\cdot\text{mL}^{-1}$ Se can be recommended for bulb onion. The Se50 treatment had a tendency to increase the yield and bulb size, increased bulb total antioxidant capacity in both years, also a greater amount of total Se was converted into organic compounds compared to the Se100 treatment. Higher rates of Se could be toxic to onion plants, since Se100 treatment had a clear tendency towards bulb size and yield decrease compared to the Se50 treatment, also highest Se content in the Se100 treatment was accompanied with the lowest S content, which is an unfavorable effect.

Further field experiments with other onion cultivars and with repeated foliar Se fertilization could be beneficial to find out whether it is possible achieve higher Se content in onion bulbs without adverse effects on plant metabolism, onion yield and bulb quality.

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**WPLYW ZABIEGÓW SELENEM W WRUNKACH POLOWYCH
NA ODŻYWIANIE MINERALNE, PLON I WŁAŚCIWOŚCI
ANTYOKSYDACYJNE CEBULI**

Streszczenie. Celem niniejszego badania było stwierdzenie, w jaki sposób zabieg dolistnego stosowania selenu (Se) w otwartych warunkach polowych wpływa na odżywianie roślin, plon oraz bioaktywne cechy cebul. 10, 50 lub 100 $\mu\text{g}\cdot\text{mL}^{-1}$ Se, co określano jako zabieg Se10, Se50 i Se100, i zastosowano w uprawie cebuli ‘Hercules’ w latach 2008 oraz 2009. Określono masę cebul, plon całkowity, całkowitą zawartość Se oraz wolnej selenometioniny i *selenometyloselenocysteiny*, całkowitą zawartość S, N, P, K, Ca i Mg, fenoli, ostrość oraz całkowitą zdolność antyoksydacyjną. Wszystkie zabiegi z użyciem Se znacznie ograniczyły zawartość S w cebulach. Zabieg Se 100 wykazywał tendencję do zmniejszania wielkości bulw i plonu. Zabieg Se50 zwiększał całkowitą zawartość Se, całkowitą zawartość fenoli i całkowitą zdolność antyoksydacyjną oraz powodował tendencję do zwiększania plonu. Więcej całkowitego Se przetwarzano się na związki organiczne w Se50 niż w Se100. W zabiegu Se10 całkowita zdolność antyoksydacyjna w cebulach była największa ze wszystkich zabiegów w roku 2009. Biorąc pod uwagę korzyści zarówno agronomiczne, jak i dla ludzkiego zdrowia, roztwór Na_2SeO_4 w ilości 50 $\mu\text{g}\cdot\text{mL}^{-1}$ Se można zarekomendować dla cebuli.

Słowa kluczowe: selenoamino-kwasy, siarka, całkowita zawartość fenoli, ostrość, zdolność antyoksydacyjna

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