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Effects of selenium fertilizer on grain yield, Se uptake and distribution in common buckwheat (*Fagopyrum esculentum* Moench)

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ABSTRACT

Selenium (Se) is a significant trace element for human and livestock animals because of its physiological functions. Se in plants, especially in the crop plants, is treated as a critical dietary source. The effects of foliar spray together with soil application of Se on Se uptake, distribution in common buckwheat (*Fagopyrum esculentum* M.) plants were discussed in this study. The results showed that both foliar spray and soil application of Se increased Se uptake in common buckwheat significantly ($P < 0.05$). The highest Se content was observed in leaves (113.37–690.75 μg/kg), followed by roots (28.98–283.78 μg/kg), grains (26.49–135.89 μg/kg) and stems (23.19–86.80 μg/kg). Se content in grains had the highest correlation coefficient (0.827 and 0.845) with soil Se application treatments. Grain yield of F1 (5 g Se/ha for foliar spray) was 3.65% and 10.25% higher than that of F0 (0 g Se/ha for foliar spray) in two study years, respectively. Under soil Se application conditions, mean grain yields fluctuated from 2890.5–3058.6 kg/ha, 2966.4–3352.8 kg/ha in 2012/2013, respectively. These results indicated a significant interaction effect of foliar spray Se and soil Se application on Se accumulation in common buckwheat. Appropriate Se application might improve common buckwheat grain yield.

Keywords: essential trace element; selenium; field condition; biofortification

As a component of several major metabolic pathways, selenium (Se) plays an important role in human health (Rayman 2006, Winkel et al. 2012). It is also considered as an essential trace element for human, animals and some species of microorganisms (Stroud et al. 2010). Approximately two-thirds of the cultivated soil in China is Se-deficient, which results in a lack of Se in the human population in this region (Zhang et al. 2014). Since direct Se supplementation suffers from low bioavailability of inorganic Se and possible accidental excess of Se intake by humans, one of the effective measures to solve this situation seems to be the supplement of Se into the food chain, and agronomic biofortification is considered to be more advantageous (Hartikainen 2005).

Although no evidence has demonstrated that Se is an essential element for plants, low concentrations of Se could promote the growth of plants (Djanaguiraman et al. 2010). In order to increase Se uptake in food crops, attempts have been made in wheat (Ducsay et al. 2006, Curtin et al. 2008), maize (Chilimba et al. 2012), potato (Hlušek et al. 2005) and soybean (Yang et al. 2003). Based on our understanding, the previous studies primarily demonstrated that the application of Se increased the Se content of grains, but little focus was paid to the application methods in Se uptake among the parts of crop plants, and the interaction effect and relationship between different Se application methods.

Buckwheat (*Fagopyrum esculentum*) is grown in many countries in Asia, Europe and in certain

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other places around the world (Hsu et al. 2008). Vogrinčič et al. (2009) reported that the mass fractions of Se in all plant parts increased and were approximately 50- to 500-fold higher according to the plant part after foliar fertilization with sodium selenate, suggesting that common buckwheat is a proper food crop for Se-rich food, especially in the Se-deficiency area. However, the interaction effect and relationship of foliar and soil application of Se on crop Se uptake are still not fully understood especially in common buckwheat under field conditions. The aim of this contribution was to evaluate and discuss the role of Se foliar together with soil application in influencing Se distribution in common buckwheat plant, the interaction effect and relationship of both them on common buckwheat Se uptake and further understand the effects of Se application on the grain yield of common buckwheat.

MATERIAL AND METHODS

A two-year field experiment was performed during 2012/2013 at the Chifeng Academy of Agricultural and Animal Husbandry Sciences research farm, Chifeng, China (42°17'N, 118°52'E). The weather data during the experiment period in each year are given in Figure 1. This area is classified as a semi-arid continental monsoon climate with an annual precipitation 250–400 mm. The soil of experimental field was a sandy loam texture. The physical and chemical parameters of the experimental soil were as followed: pH 7.8, organic matter 13.4 g/kg, total N 0.55 g/kg, available N 47.3 mg/kg, available P 18.4 mg/kg, available K 101.8 mg/kg, and total Se 0.34 mg/kg. The analytical method used is described in detail elsewhere (Ray et al. 2015), except total soil Se described as Wang et al. (2012).

The experiment adopted a split plot design with three replications. Main plots were foliar spray Se application treatments, including 0 (F0), 5 (F1), and 10 (F2) g Se/ha. While four concentration levels of Se for soil application: (0, 6, 12 and 18 g Se/ha, hereafter referred to as C, L, M and H) were assigned to the subplots with each plot 30 m^2 (4.95 × 6.06 m). The solution of commercial fertilizer including selenite were used for foliar spray fertilizing at the period of 45 days after sowing for F1, and F2 was at the period of 45 and 65 days after sowing (each 5 g Se g/ha). The soil Se (Na_2SeO_3) application was supplied together with compound fertilizer when common buckwheat was planted.

Grain yield and components were measured by common methods. Ten plant samples for each subplot were collected randomly and separated into roots, stems, leaves and grains at the mature stage. Roots were washed three times with deionized water to remove dust and soil. Plant samples were oven dried to constant weights and ground into powder for Se analysis.

For the determination of total Se, the plant samples were digested using the method described by Wang et al. (2012) with modification. Digestion of samples in the 50 mL conical flask was carried out on a temperature control furnace, 0.5–1.0 g of sample and 10 mL of acid mixture (8 mL of HNO_3 + 2 mL of $HClO_4$) were added and kept overnight at room temperature. Then, flasks were heated at

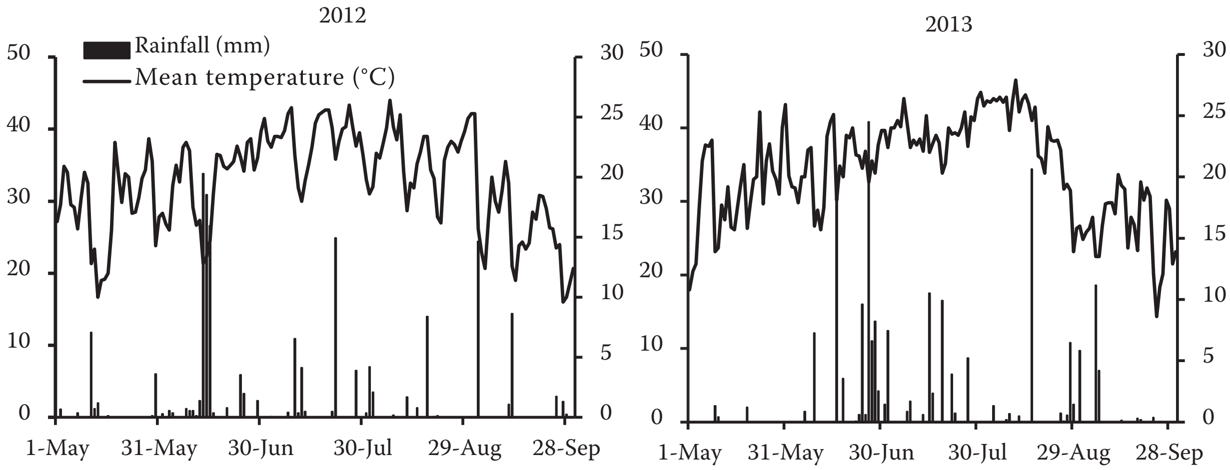

Figure 1. Daily mean air temperature and rainfall during the study period

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170°C until white smoke appeared, sample solutions were cooled to the room temperature, 5 mL of 6 mol/L HCl was added and kept heating for the reduction of Se^{6+} to Se^{4+} until the white smoke appeared again. About 2 mL residual sample solution was diluted by 10% HCl (v/v) in 25 mL volumetric flask after digestion. According to the GB/T 21729 (2008), the total Se was determined using 8220 atomic fluorescence spectrophotometer with hydride generation (Jitian, Beijing, China). The Se usability of common buckwheat grain (SeU) was calculated as follows:

$$SeU = (c_{Se} \times Y_{Se} - c_{control} \times Y_{control})/m_{Se}$$

Where: c_{Se} and Y_{Se} – Se concentration and yield of grain in the plots with Se applied; $c_{control}$ and $Y_{control}$ – Se concentration and yield of grain in the plots without Se applied; m_{Se} – mass of Se applied in the plots.

The data were subjected to ANOVA using GLM of SAS 9.2 (SAS Institute, 2002). The least significant difference (*LSD*) was used to compare means of traits at $P = 0.05$.

RESULTS AND DISCUSSION

The effects of Se application on Se uptake in root, stem, leaf, and grain of common buckwheat were showed in Table 1. Both the experimental treatments significantly affected Se concentration in the root, stem, leaf, and grain of common buckwheat.

Many studies have reported that Se uptake of crop could be increased by Se application in wheat (Germ et al. 2013), maize (Chilimba et al. 2012), rice (Li et al. 2010) and other crops (Altansuvd et al. 2014). Single effects of foliar spray or application via root of Se on grain Se concentration and yield have been recognized in buckwheat under controlled conditions (Vogrinčič et al. 2009). The present study, in a field condition, firstly reported significant effects of foliar spray, soil application Se and their interaction in common buckwheat. It indicated that the different pathway of Se uptake would affect total Se accumulation in common buckwheat, because Se was absorbed by leaf and root under our experimental treatments, respectively. Consequently, the mechanism of Se uptake and translocation in plants need further studies.

Both foliar spray and soil application of Se had a significant effect on Se accumulation in the parts of common buckwheat ($P < 0.05$). The highest Se content among different parts of common buckwheat was observed in F2 and H treatments, F1 and F2 had higher Se accumulations in the parts of common buckwheat plant in comparison to F0 treatment, respectively (Table 2). Chilimba et al. (2012) found that maize grain Se concentration increased by 11–33 μg Se/kg for each g Se/ha soil applied (0, 1.5, 3.0, 4.5 and 6.0 g Se/ha) under field condition, Vogrinčič et al. (2009) also suggested that common buckwheat have a high Se-rich potential. In our study, Se content of the common buckwheat plant increased with the rising Se rates of soil application. The Se content was the highest in the leaves (113.37–690.75 μg/kg), followed by the roots (28.98–283.78 μg/kg), grains (26.49–135.89 μg/kg)

Table 1. Variation of significance in foliar spray and soil application of selenium (Se) for Se concentration in parts of common buckwheat

Year	Variation source	Root	Stem	Leaf	Grain
2012	foliar spray (V_1)	**	***	***	***
	soil application (V_2)	***	***	***	***
	$V_1 \times V_2$	***	***	**	**
	CV (%)	27.153	11.704	20.153	17.877
	$P > F$	< 0.0001	< 0.0001	< 0.0001	< 0.0001
2013	foliar spray	**	***	***	**
	soil application	***	***	***	***
	$V_1 \times V_2$	***	***	**	**
	CV (%)	37.659	20.322	18.999	16.752
	$P > F$	< 0.0001	< 0.0001	< 0.0001	< 0.0001

*0.01 < P < 0.05; **0.001 < P < 0.01; ***P < 0.001; ns – not significantly different; *CV* – coefficient of variation; *F* – *F-value*

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Table 2. Effects of foliar spray and soil application of selenium (Se) on Se concentration in parts of common buckwheat (μg/kg)

Year	Factor	Treatment	Root	Stem	Leaf	Grain
2012	foliar spray	F0	118.7[c]	32.8[c]	128.2[c]	43.3[b]
		F1	161.7[b]	42.9[b]	334.4[b]	81.7[a]
		F2	183.7[a]	67.6[a]	690.8[a]	88.3[a]
	soil application	C	28.9[b]	23.2[d]	215.6[d]	26.5[d]
		L	54.8[b]	41.1[c]	321.8[c]	55.1[c]
		M	251.3[a]	52.3[b]	441.2[b]	94.1[b]
		H	283.8[a]	74.7[a]	559.3[a]	108.6[a]
2013	foliar spray	F0	104.2[c]	36.9[c]	113.4[c]	64.8[b]
		F1	150.9[b]	50.8[b]	317.2[b]	102.3[ab]
		F2	209.8[a]	82.3[a]	597.5[a]	111.5[a]
	soil application	C	26.3[b]	23.6[c]	204.8[c]	55.1[c]
		L	74.7[b]	52.8[b]	314.2[b]	67.7[c]
		M	247.2[a]	63.5[b]	329.6[b]	112.7[b]
		H	271.6[a]	86.8[a]	522.1[a]	135.9[a]

Means followed by different letters indicate significant differences between treatments at $P < 0.05$. Foliar spray Se application: F0 – 0, F1 – 5, F2 – 10 g Se/ha; soil application: C – 0, L – 6, M – 12, H – 18 g Se/ha

and the stems (23.19–86.80 μg/kg). In addition, the soil factors of our experimental area and the interaction effect between soil and foliar Se application resulted in a higher Se-rich in grain probably.

The Se usability of common buckwheat grain dry matter was shown in Figure 2. Application methods of Se influenced the Se usability of grain significantly ($P < 0.05$). The Se usability of grain under F1 condition was much higher in comparison to that under soil Se application condition, but it was not under F2 condition. As mentioned above, the buckwheat grain had not enough time for Se offtake under F2 treatment in our study resulting probably in the Se usability decrease. The rate of Se uptake depends on the concentration and chemical form of Se in the soil solution, as well as rhizosphere conditions such as pH and the presence of sulfate and phosphate, which compete with Se uptake (Sors et al. 2005). Thus, it led to the Se usability of grain decrease under soil Se application condition in our study.

As shown in Table 3, Se concentrations in different parts of common buckwheat were significantly correlated with Se application methods, with the exception of root concentration under foliar spray Se condition ($P < 0.05$). Our results

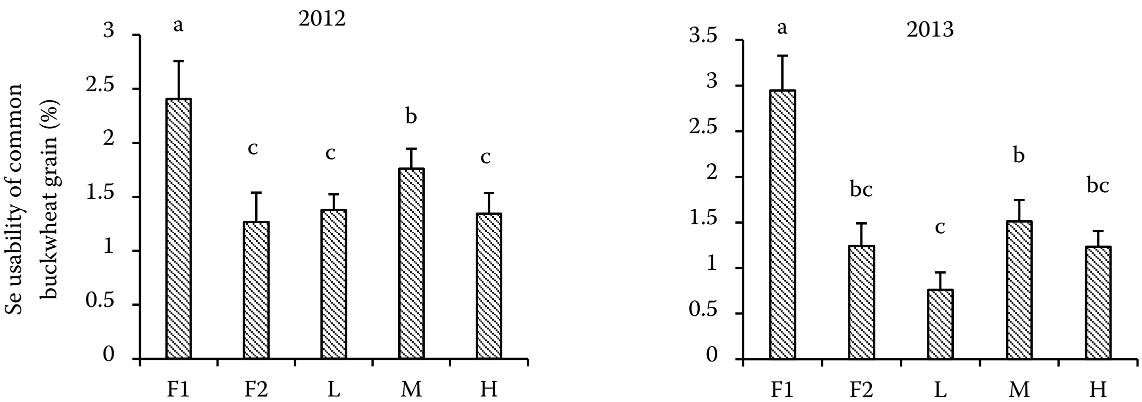

Figure 2. Selenium (Se) usability of common buckwheat grain. Means followed by different letters indicate significant differences between treatments at $P < 0.05$. Foliar spray Se application: F1 – 5, F2 – 10 g Se/ha; soil application: L – 6, M – 12, H – 18 g Se/ha

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Table 3. Correlations between selenium (Se) concentrations in various parts of common buckwheat and two Se fertilizer application methods

Year	Treatment	Root	Stem	Leaf	Grain
2012	foliar spray	–0.045	0.569**	0.809**	0.388**
	soil application	0.689**	0.742**	0.385*	0.827**
2013	foliar spray	–0. 072	0.557**	0.642**	0.471*
	soil application	0.672**	0.674**	0.322*	0.845**

*$P < 0.05$; **$P < 0.01$

show that Se concentration in grains was the highest correlation coefficient (0.827, 0.845) in soil Se treatments, but it was 0.809 and 0.642 in leaves under foliar spray conditions. These observations indicated that a better Se accumulation in grain may be via soil Se application. Se content of leaves increased greater than that in grains by foliar spray Se, probably due to the Se application period (part of F2) which was 30 days before harvest in our study, giving limited time for common buckwheat leaves to transfer Se into grains. In addition, the Se of foliar spray probably affected crop Se uptake form its roots resulting in the negative correlation (–0.045, –0.072) between foliar spray Se and the Se content of roots.

The effects of Se application on seeds per plant, 1000-kernel weight (TKW), and grain yield of common buckwheat were showed in Table 4. The seeds per plant were significantly affected by soil Se application in 2012/2013 ($P < 0.05$). In 2013, the grain yield was influenced by either foliar spray or soil application of Se ($P < 0.05$). Compared to F0, the grain yield of F1 was increased by 3.65% and 10.25% in 2012/2013, respectively (Table 5). Under soil Se application conditions, mean yields of buckwheat grain fluctuated between 2890.5–3058.6 kg/ha in 2012, and 2966.4–3352.8 kg/ha in 2013. The highest yield was harvested in M and L treatment in both years, respectively. Due to no significant effect of Se application on TKW, the increased seeds per plant resulted in the grain yields increasing properly, but an excessive Se application could decreased the grain yield (F2 and H). Mean grain yield in 2012 was lower than that in 2013, the difference of weather conditions (Figure 1) influenced the two-year grain yields significantly.

Selenium is an essential element for humans and livestock, although no essential function has yet been proven in vascular system of plants (Broadley et al. 2010). However, studies concerned with Se effects on crop yield gave controversial results. Some studies reported little effect of Se on crop grain yield, such as wheat (Ducsay and Ložek 2006), maize (Chilimba et al. 2012), and soybean (Yang et al. 2003), while others reported that Se improved grain yield of maize (Wang et al. 2012), wheat (Nawaz et al. 2015), and rice (Zhang et al. 2014). In this study, observations in the field condition showed an increase in yield of buckwheat under appropriate Se application levels (F1, L and M), but the grain yield of buckwheat did not increase with the amount of Se application (F2, H) (Table 5).

Table 4. Variation of significance in foliar spray and soil application of selenium (Se) fertilizer for seeds per plant, 1000-kernel weight (TKW) and grain yield

Variation source	2012			2013		
	seeds/plant	TKW (g)	grain yield (kg/ha)	seeds/plant	TKW (g)	grain yield (kg/ha)
Foliar spray (V_1)	ns	ns	ns	ns	ns	**
Soil application (V_2)	*	ns	ns	**	ns	*
$V_1 \times V_2$	ns	ns	ns	ns	ns	ns
CV (%)	17.52	3.28	12.96	8.15	3.72	17.58
$P > F$	0.283	0.015	0.730	0.081	0.901	0.042

*$0.01 < P < 0.05$; **$0.001 < P < 0.01$; ***$P < 0.001$; ns – not significantly different; *CV* – coefficient of variation; *F* – *F-value*

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Table 5. Effects of foliar spray and soil application of selenium (Se) fertilizer on seeds per plant, 1000-kernel weight (TKW) and grain yield

Factor	Treatment	2012			2013		
		seeds/plant	TKW (g)	grain yield (kg/ha)	seeds/plant	TKW (g)	grain yield (kg/ha)
Foliar spray	F0	217.4[a]	26.1[a]	2907.3[a]	248.2[a]	28.9[a]	3071.9[b]
	F1	224.3[a]	27.5[a]	3013.5[a]	251.8[a]	28.4[a]	3386.8[a]
	F2	220.4[a]	27.1[a]	2860.8[a]	246.9[a]	28.1[a]	2900.5[b]
Soil application	C	221.8[ab]	26.1[a]	2890.5[a]	239.1[b]	28.3[a]	3292.6[ab]
	L	214.7[ab]	27.1[a]	2891.9[a]	269.1[a]	28.2[a]	3352.8[a]
	M	242.1[a]	27.4[a]	3058.6[a]	255.7[ab]	28.2[a]	3220.4[ab]
	H	204.2[b]	25.9[a]	2933.7[a]	237.4[b]	28.0[a]	2966.4[b]

Means followed by different letters indicate significant differences between treatments at $P < 0.05$. Foliar spray Se application: F0 – 0, F1 – 5, F2 – 10 g Se/ha; soil application: C – 0, L – 6, M – 12, H – 18 g Se/ha

Our results correspond with the previous study (Zhang et al. 2014) on rice plants, in which they will wee the influence of Se application on rice yield. Moreover, soil texture, its physicochemical characteristics, method and time of Se application also influence its relative effectiveness (Lyons et al. 2005) in improving the yield of crops (Nawaz et al. 2015).

The present study indicated that Se accumulation in common buckwheat was closely associated with the application rate of Se. The appropriate Se application in the common buckwheat cultivation might prompt grain yield improvement. We also observed a significant interaction effect between foliar spray Se and soil basal application Se fertilizer in Se accumulation of common buckwheat. The mechanism of the interaction effect needs to give a further research for understanding the plant Se uptake well.

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