## **RESEARCH ARTICLE**

## Plant availability of inorganic and organic selenium fertiliser as influenced by soil organic matter content and pH

Susanne Eich-Greatorex · Trine A. Sogn · Anne Falk Øgaard · Ivar Aasen

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Abstract The objective of the present study was to investigate the influence of soil organic matter content and pH on plant availability of both inorganic and organic selenium (Se) fertilisers. Further, the risk of Se leaching after application of inorganic Se fertiliser was evaluated. A new interpretation of an older field study at different sites in Southern Norway showed that organic C was correlated with grain Se concentration in wheat, barley and oats, explaining up to 60% of the variation in Se concentration. Pot experiments with a peat soil, a loam soil and a peat/ loam soil mixture were conducted for the present study at a range of pH values between pH 5 and 7. Below pH 6, Se uptake from added Se fertiliser was higher in the soil types with high organic matter content than in the loam. The opposite occurred at a soil pH above 6, where Se uptake was higher in the loam than in the peat soil. A simple leaching experiment after one growing season confirmed the

S. Eich-Greatorex (⊠) · T. A. Sogn ·
A. F. Øgaard · I. Aasen
Department of Plant and Environmental Sciences,
Norwegian University of Life Sciences, P.O. Box 5003,
N-1432 Aas, Norway
e-mail: susanne.eich@umb.no

A. F. Øgaard

Soil and Environment Division, Norwegian Institute for Agricultural and Environmental Research, Fredrik A. Dahls vei 20, N-1432 Aas, Norway findings of the pot experiments that Se availability in the loam soil with a relatively low organic matter content increased with increasing pH, whereas it decreased in the peat soil. Neither Se yeast, nor pure Se methionine, used as organic Se fertiliser, resulted in any significant uptake of Se when added at concentrations similar to the inorganic Se applications.

Keywords Inorganic Se fertiliser ·

Organic matter content  $\cdot$  Se availability  $\cdot$  Se leaching  $\cdot$  Se methionine  $\cdot$  Se yeast

#### Introduction

The trace element selenium (Se) is of fundamental significance for both human and animal health, with its best-known function in the antioxidant enzyme family of glutathione peroxidases (Rayman 2000). Recommended Se contents in grain, used as fodder and food, are 0.2–0.3 mg Se kg<sup>-1</sup> dry matter (DM) and 0.1–0.2 mg Se kg<sup>-1</sup> DM, respectively. In parts of the world, Se availability in the soil is so low that Se contents in the crops produced there are far below recommended values. One example is Scandinavia, where the low Se content and availability in the soils are of concern (Gissel-Nielsen et al. 1984; Wu and Låg 1988). In Finland, Se-enriched mineral fertilisers have been used in plant production since 1985 to secure the recommended content in plants for forage

or human consumption (Korkman 1984; Eurola et al. 1989; Ekholm 1997).

Since the difference between Se essentiality and toxicity is very small, the application of Se via fertiliser is controversial. Usually, between 5 and 30% of Se applied by mineral fertiliser are utilised by plants (Mikkelsen et al. 1989; Yläranta 1990; Tveitnes et al. 1996; Bakken and Ruud 2000). The rest is retained in the soil, leached or to some extent also lost to the atmosphere by volatilisation. Previously applied Se that is bound in the soil may, under changed management practices or other changes in soil environmental factors, become mobile again and may then represent a potential environmental problem and health risk. Mobilisation of Se may lead to Se leaching which in turn may contaminate drinking water and/or increase Se contents in the vegetation. To ensure that applied Se is not a potential environmental risk, the factors controlling Se mobility need to be well understood, as well as possibilities of increasing the utilisation of Se from fertilisers should be further investigated.

Among the factors controlling Se mobility in soils are pH, redox conditions and organic matter content. Several studies show that an increase in soil pH increases plant Se uptake (Cary and Allaway 1969; Lindberg and Bingefors 1970; Johnsson 1991). Redox conditions and pH have an important effect on Se availability since a combination of these factors determines the Se species present in a given soil environment. For instance, selenate (SeO<sub>4</sub><sup>2-</sup>) is the predominant Se species in near-neutral pH environments under aerobic conditions whereas selenite (SeO<sub>3</sub><sup>2-</sup>) predominates at lower pH and redox potential. Selenate is much more mobile and thus plant-available in soils than selenite which is tightly bound to positively charged binding sites in soil.

The effect of organic matter in the soil on plant Se availability may vary depending on the type of organic compounds present. Several studies have shown that Se is effectively bound to soil organic matter and thus less plant available. For instance, Johnsson (1991) found the Se concentration in grain to be reduced from 1350 mg kg<sup>-1</sup> to 160 mg kg<sup>-1</sup> when the organic matter content in the plough layer increased from 1.4% to 39%. In a soil column trial with podzol, Gustafsson and Johnsson (1992) showed that 77% of the added selenite was retained in the upper 2 cm thick raw humus layer. Some organic

acids, however, may have the opposite effect on plant Se availability. Wijnja and Schulthess (2000) found that the presence of oxalate and citrate inhibited adsorption of selenate by competition for the anion binding sites, and thus increased Se availability.

In addition to the inorganic species selenate and selenite, plants are also able to take up organicallybound Se, such as Se methionine, from solution culture in a metabolically active process (Abrams et al. 1990). If plants are able to utilise Se from organic sources to a similar extent as Se from inorganic fertilisers, e.g., organic waste products with a high Se content may be used to ameliorate Se deficiency in soils.

The intention of the present study was to investigate the influence of soil organic matter content and pH on plant availability of both inorganic and organic Se fertilisers, as well as evaluate the risk of Se leaching after application of inorganic Se fertiliser. The paper includes a new interpretation of an older field study, as well as new data from pot experiments. Based on the results published by Johnsson (1991) and Gustafsson and Johnsson (1992), the hypothesis that increasing soil organic matter content results in lower plant availability of applied Se was tested.

#### Materials and methods

Application of inorganic Se in field experiments

In the period 1986–1988, a series of field experiments was conducted on mineral soils at nine different sites (Table 1), distributed over the most important wheat production districts of South-Eastern Norway (Sogn et al. 1991). The initial total Se content of the soils ranged from 0.09 to 0.39 mg  $kg^{-1}$  (determined as acid-soluble Se) and the organic C content from 1.5 to 4.1%, which represent typical values for agricultural soils in South-Eastern Norway. The Se treatments in the first experimental year consisted of an application of 0, 2.25, 4.5, 9, and 18 g Se ha<sup>-1</sup> (as Na<sub>2</sub>SeO<sub>4</sub>), respectively, given as part of a NPK fertiliser (21-4-10). The dose of the NPK fertiliser was equivalent to 100 kg N ha<sup>-1</sup>. Spring wheat (variety Runar), barley (variety Pernille), and oats (variety Svea) were grown as test crops. After the harvest, the plant material was digested by wet ashing and Se was determined by a fluorometric method (Ihnat 1974; Rygge et al. 1977).

**Table 1** Organic C content, clay content, pH, and acid-solubleSe in the plough layer (0 to 20 cm) of the nine experimentalsites at the start of the field experiments (from Sogn et al.1991)

Site No. (as in Sogn et al.)	Org. C (%)	Clay content (%)	pН	Acid-soluble Se ( $\mu$ g kg <sup>-1</sup> )
6	1.5	15	5.9	110
2	2.1	26	6.3	120
1	2.4	9	6.1	140
4	2.4	14	6.0	170
5	2.4	19	7.2	130
9	2.7	17	6.0	90
3	2.8	27	6.1	190
7	3.2	26	6.0	280
8	4.1	20	6.8	390

At each site, the top layer of the soil (0-20 cm) was sampled for analysis at the start of the experimental period (Table 1). Acid-soluble Se in the soil samples was determined by extracting the soil with concentrated HNO<sub>3</sub> and detection of Se in the extract by hydride generation atomic absorption spectroscopy using a dry-ashing procedure (Øien et al. 1988). For the present study, the data from the field experiments was used to investigate correlations between grain Se content and pH, clay content, and organic C content in the soils at the respective sites.

# Application of inorganic Se in greenhouse experiments

In order to further study the effect of soil organic matter content, as well as pH, on utilisation of inorganic Se applied via fertiliser, greenhouse pot experiments with different soil types and wheat as a test crop were conducted. The soil types included a commercial growth peat, a loam soil, and a mixture consisting of 30% peat and 70% loam soil on a volume basis. The loam was passed through a 5-mm mesh size prior to being filled into the pots. Some chemical characteristics of the peat and loam soil are given in Table 2. The values for the peat/loam soil mixture are calculated from the ratio of peat and loam soil in the mixture and their respective analyses. All pH determinations were conducted in H<sub>2</sub>O with a soil to solution ratio of 1:2.5. Organic C was determined by dry combustion (Nelson and Sommers 1982). Total N was determined according to the Dumas method (Bremner and Mulvaney 1982). P, Ca, K, Mg, and Na were extracted in ammonium acetate lactate solution (0.1 M ammonium lactate and 0.4 M acetic acid) at pH 3.75 with a soil to solution ratio of 1:20 (Egnér et al. 1960). Total Se was determined by hydride generation atomic absorption spectrophotometry (Perkin Elmer 1100-B) after digestion in HNO<sub>3</sub> and Mg(NO<sub>3</sub>)<sub>2</sub>, ashed at 450°C and resolved in HCl (Øien et al. 1988).

The effect of organic matter content and pH on Se availability was tested in three independent pot experiments in 2003, 2004 and 2005, respectively. In all three experiments, inorganic Se was added as Na<sub>2</sub>SeO<sub>4</sub>, in the form of a selenate-enriched NPK fertiliser (NPK 21-3-8 with 12 mg Se per kg fertiliser) when added at the same time as the NPK fertilisation, or as pure Na<sub>2</sub>SeO<sub>4</sub>. All treatments, including the control, received the same amount of N, P, K, Mg, and micronutrients with an N application equivalent to 157.5 kg  $ha^{-1}$  in the first two experiments and 240 kg ha<sup>-1</sup> in the third experiment (2005). In the first two experiments, NPK fertilisation, and thus also Se fertilisation, was divided into two, with a main dose at the start of the experiment and a smaller dose at the beginning of the shooting stage (Zadoks 31; Zadoks et al. 1974). In the third experiment, only one dose of NPK fertilisation was given, but the application of Se was divided in the same way as in the first two experiments. In all three experiments, the Se dose was equivalent to 9 g Se  $ha^{-1}$  (0.03 mg per pot), divided into the main Se dose equivalent to 8 g Se  $ha^{-1}$  and the dose at the beginning of the shooting stage equivalent to 1 g Se  $ha^{-1}$ .

Each of the three pot experiments was conducted at two pH levels. In the first experiment, the amount of CaCO<sub>3</sub> needed to obtain a desired pH was calculated on the basis of soil type and the original pH of the soil. Further soil samples for pH determination were taken after each growing season. In the following experiments, adjustments were made based on the pH values found in the previous experiment(s) so that measured pH values at the end of each experiment varied between soil types, growing seasons, and pH treatment. Each pH/Se-treatment had 3 replicates (n = 3).

Within each soil type, the results from the three different experiments (three times two pH levels) were grouped into pairs with similar pH so that the

Soil type	pН	Org. C (g kg <sup><math>-1</math></sup> )	$\frac{N}{(g \ kg^{-1})}$	P (mg kg <sup>-1</sup> )	K (mg kg <sup>-1</sup> )	Mg (mg kg <sup>-1</sup> )	Ca (mg kg <sup>-1</sup> )	Na (mg kg <sup>-1</sup> )	Se (mg kg <sup>-1</sup> )
Loam	4.5	22	2	59	120	57	499	18	0.26
Peat	4.2	463	11	22	158	537	1080	92	0.26
Peat/loam	4.4	62	3	56	123	101	552	25	0.26

Table 2 Selected chemical characteristics of the soils used in the greenhouse experiments

pH values in Table 4 and Fig. 2 are averages of six replicates. For the loam soil, these averages were pH 5.2, pH 5.9, and pH 6.4. For the peat soil, the respective averages were pH 5.3, pH 5.9 and pH 6.8. Since the peat/loam soil mixture was only used in the second experiment, the pH values in Table 2, i.e., pH 5.7 and pH 6.2 are averages of only three replicates. The measured pH values in the single experiments differed up to 0.3 units from these averages.

The pots contained a soil volume of 6.7 l, and were sown with 30 seeds of spring wheat (variety "Avle C2"). The plants were thinned to a total of 20 pot<sup>-1</sup> approximately one week after germination. Moisture content in the soil was maintained at 60% of field capacity by irrigation with deionised water according to weight loss. After the harvest, two pF cylinders (100 cm<sup>3</sup>, 3.7 cm high) were inserted into each soil type to take undisturbed soil samples. Air-filled pore volume in the samples was determined with the help of an air pyknometer. Water-filled pore volume was calculated from the weight difference between wet and dry soil.

The crop was harvested at maturity, and grain and straw divided for separate analysis. Grain and straw Se concentrations were measured by high-resolution inductively coupled plasma mass spectrometry (HR-ICP-MS, Thermo Finnigan Element 2, Waltham/MA, USA) after microwave digestion in aqua regia (0.25 g in 2.5 ml) and subsequent dilution (to 25 ml).

Selenium leaching in the greenhouse experiments with inorganic Se application

After the harvest of the first experiment with inorganic Se application, a simple leaching experiment was conducted in order to assess the risk of Se loss from the soil due to, e.g., heavy rainfall in the autumn and whether there were any differences in leaching depending on soil organic matter content and pH. For that purpose, pots filled with peat soil at pH 5.4 and pH 6.8, and pots filled with loam soil at pH 4.9 and pH 6.3, respectively, were used. Two pots per treatment and pH level were kept at a moisture content equivalent to 60% field capacity for some weeks after the harvest until the start of the leaching experiment. They were then irrigated to field capacity before 1 1 H<sub>2</sub>O was added to the soil surface in four units of 250 ml each during the course of a few hours. The leachate was collected in acid-washed bottles. Subsamples of 100 ml were transferred into smaller, acid-washed bottles and conserved with 0.5 ml suprapure HNO<sub>3</sub> (65%). Se content in the solution was determined by HR-ICP-MS.

Application of organic Se in a greenhouse experiment

The effect of soil organic matter content and pH on plant availability of organic Se supplements was also tested in one of the pot experiments. Organicallybound Se was added either as organic Se yeast (O.S.Y. 2000X—mineral enriched yeast, Ultra Bio-Logics Inc., Québec, Canada) or as pure Se methionine (Merck KGaA, Darmstadt, Germany). The organic Se yeast contained 2150  $\mu$ g Se g<sup>-1</sup> in the form of Se methionine. Also in these pots, the application of Se was divided into an initial Se dose equivalent to 8 g Se ha<sup>-1</sup> and a dose at the beginning of the shooting stage equivalent to 1 g Se ha<sup>-1</sup> to allow comparison to the inorganic Se fertilisation treatments. Nutrients were supplied in the same amounts as described above.

## Statistical analysis

The effects of inorganic Se fertilization on yield, Se concentration and Se uptake in the greenhouse experiments were tested statistically by analysis of variance (General Linear Model). The t-test with the LSD procedure was performed to identify different

means. Results with P < 0.05 were considered significant. The statistical analysis was carried out using SAS (SAS Institute Inc.).

### Results

Application of inorganic Se in field experiments

Selenium concentrations in the grain after the first growing season increased with the amount of Se added in the first year of the field experiments. With the lowest rate of 2.25 g Se  $ha^{-1}$ , Se concentrations in wheat grain ranged between 0.03 and 0.09 mg kg<sup>-1</sup>. An application of 4.5 g Se ha<sup>-1</sup> resulted in Se concentrations between 0.07 and 0.18 mg kg<sup>-1</sup>. An application of 9 g Se ha<sup>-1</sup> increased the Se concentration to 0.17-0.49 mg kg<sup>-1</sup>, whereas 18 g Se ha<sup>-1</sup> led to Se concentrations well above the recommended values for human consumption and animal forage, up to  $0.9 \text{ mg kg}^{-1}$  (Sogn 1987). The average yield for wheat was 2760 kg DM ha<sup>-1</sup> and no significant differences in yield among Se treatments were found (unpublished data). Se concentrations in the grain of barley and oats were in a similar range whereas the average yield for barley was 3240 kg DM  $ha^{-1}$  and for oats of 3360 kg DM  $ha^{-1}$  (unpublished data).

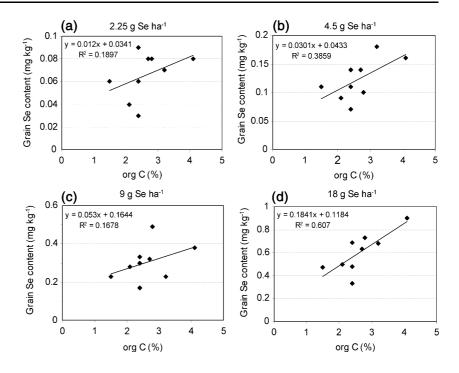
The Se concentrations in wheat, barley and oats found in the field experiments are in our study related to different soil parameters at the respective site (as given in Table 1), i.e., pH, clay content, organic C, and Se content of the soil. Among these factors, organic C was the one best correlated with grain Se concentration (Table 3). Up to 60% of the variation in grain Se concentration was explained by the variation in soil organic C content. Yield, on the other hand, was not correlated with organic C ( $R^2 = 0.10$  for all cereal types combined; data not shown).

Since figures were similar for the three crops, Fig. 1 shows only the relationship between organic C in the soil and grain Se content for wheat. The correlation was strongest at an application of 18 g Se ha<sup>-1</sup> in wheat ( $R^2 = 0.60$ ), but a higher content of organic C was correlated to a higher content of Se in the grain also at the other applications, especially at an application of 4.5 g Se ha<sup>-1</sup> ( $R^2 = 0.39$ ). Acidsoluble Se in the soil was positively correlated to grain Se concentration (Table 3), but also strongly correlated to the soil organic C content ( $R^2 = 0.77$ ; data not shown). Clay content was not correlated with Se concentration in grain whereas pH and Se concentration were weakly correlated in some cases.

**Table 3** Relationship ( $R^2$ ) between grain Se concentration [mg kg<sup>-1</sup> dry matter] in wheat, barley or oats grain after the first growing season and some soil factors in the field experiments, based on data from Sogn et al. (1991) and Sogn (1987)

Soil characteristics		Se application (g Se $ha^{-1}$ )					
		2.25	4.5	9	18		
рН	Wheat	0.23	0.11	0.05	0.02		
	Barley	0.04	0.08	0.18	0.01		
	Oats	0.14	0.17	0.11	0.16		
Clay content	Wheat	0.06	0.00	0.04	0.03		
	Barley	0.01	0.01	0.06	0.05		
	Oats	0.12	0.06	0.08	0.13		
Organic C	Wheat	0.19	0.39	0.17	0.60		
	Barley	0.29	0.42	0.29	0.42		
	Oats	0.10	0.21	0.13	0.44		
Acid-soluble Se	Wheat	0.11	0.37	0.09	0.52		
	Barley	0.21	0.31	0.25	0.40		
	Oats	0.01	0.10	0.02	0.24		

**Fig. 1** Correlation between grain Se content in wheat and soil organic C content at different levels of Se application in the field experiments after the first growing season (1986)



Application of inorganic Se in greenhouse experiments

In the greenhouse experiments, plant growth was clearly poorer in the pots filled with loam soil than in the pots filled with the peat/loam soil mixture or with peat soil alone (Table 4). Yields in the loam were close to half the amounts of those in the peat or peat/ loam mixture. Not even the increased N supply in the third experiment increased yields in the loam significantly. This poor plant growth was most likely due to less favourable air conditions in the loam soil that developed in the course of irrigation. The air volume in the loam soil was approximately 33% after the growing season as opposed to 67% in the peat soil, and the total pore volume 57 and 96%, respectively.

## Effect of organic matter on Se uptake

Measured pH after each experiment varied to some extent depending on soil type and experiment (see Materials and methods). However, sorting the data for the peat and loam soil treatments with respect to pH levels allowed a comparison of the effect of organic matter on Se uptake and utilisation. In general, plants grown in the soils with higher organic matter content, i.e. peat and peat/loam took up significantly (P < 0.05) more fertiliser Se than those grown in the loam at a pH below 6 (Table 4). At the highest pH level, no significant differences were found in plant Se uptake among those different soil types. Selenium concentration in grain and straw did not differ significantly between the soil types. Depending on the treatment, the average utilisation of Se by wheat plants at the different pH levels varied between 10 and 28% of the Se applied in the three experiments (data not shown; the data in Table 5 is based on the results of the first year only). The lowest Se utilisation was observed in the loam soil at pH 5.2 and 5.9, while the highest was observed in the peat soil at low pH (pH 5.3). When evaluating total Se uptake it must be kept in mind that plant growth in the loam was very poor compared to the other soil types (Table 4).

## Effect of pH on Se uptake

The differences in Se concentration in the grain between the pH levels were not significant, except for the loam soil (Table 4). Here, the Se concentration in the grain at pH 5.2 was nearly twice the respective concentration at pH 5.9. The total Se uptake per pot decreased significantly in the peat soil with increasing pH whereas it increased in the loam soil. Since Se

Soil type	рН	Yield g $pot^{-1}$		Se concentration <sup>a</sup> $\mu g$ Se $g^{-1}$		Se uptake <sup>a</sup> µg pot <sup>-1</sup>	
		Grain	Straw	Grain	Straw		
Peat	5.3	16.3ab	15.6a	0.26a	0.26a	8.2a	
	5.9	19.4a	16.7a	0.18a	0.13a	6.0ab	
	6.8	17.0a	15.8a	0.19a	0.15a	4.6bc	
Peat/loam	5.7	15.3abc	12.7ab	0.26a	0.23a	6.9ab	
	6.2	16.8ab	14.0a	0.24a	0.22a	7.1ab	
Loam	5.2	7.3d	5.3c	0.37a	0.23a	3.3c	
	5.9	9.8cd	7.2cd	0.21a	0.17a	3.2c	
	6.4	11.3bd	9.2bd	0.26a	0.31a	5.9b	

Table 4 Yield, Se concentration and Se uptake in the greenhouse experiment

Averages followed by different letters are significantly different (LSD-test, P < 0.05)

<sup>a</sup> The original data is corrected for the effect of pH on the release of soil-borne Se by subtracting the control values from the Se treatment values

 Table 5
 Plant utilisation and leaching losses of inorganic Se

 added in the greenhouse experiment in the first growing season

Soil type	рН	Plant Se utilisation (% of Se added)	Leaching loss (% of Se added)		
Peat	5.4	31.6	9.2		
	6.8	13.1	7.8		
Loam	4.9	7.5	1.3		
	6.3	26.6	15.8		

uptake was strongly influenced by the growth of the wheat plants, it was generally a lot higher in the peat and peat/loam mixture than in the loam. At the highest pH level, however, total Se uptake was higher in the loam (5.9  $\mu$ g pot<sup>-1</sup>) than in the peat (4.6  $\mu$ g pot<sup>-1</sup>) due to increasing yield in the loam and decreasing Se uptake in the peat. No differences in Se uptake were found in the peat/loam mixture where the difference between the two pH levels was relatively small.

#### Selenium leaching in the greenhouse experiment

The simple leaching experiment conducted with some of the pots from the first experiment showed similar leaching losses of Se in the peat soil at pH 5.4 compared to pH 6.8, whereas in the loam soil, losses were clearly higher at high pH (Table 5). Hardly any leaching occurred at the low pH level in the loam (pH

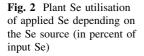
4.9) whereas leaching losses amounted to almost 16% of the Se applied at pH 6.3. Combining plant Se uptake and leaching losses shows that between 60 (peat at low pH and loam at high pH) and 90% (loam at low pH) of added Se is tightly bound in the soil or lost to the atmosphere.

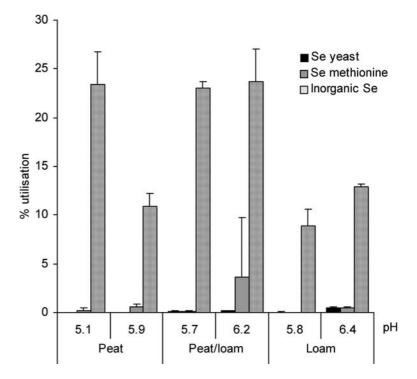
## Application of organic Se in a greenhouse experiment

Figure 2 shows plant Se utilisation from different Se sources in the greenhouse experiment, in percent of the Se added. Plant Se utilisation was calculated by the formula

[(Plant Se uptake<sub>Se-treated</sub> - Plant Se uptake<sub>control</sub>) /Se added]  $\times 100$ 

For either of the organic Se sources, Se yeast or pure Se methionine, plant Se utilisation was generally below 1%, independent of soil type and pH (Fig. 2). In the peat/loam soil mixture at pH 6.2, plant Se utilisation was slightly higher (3.6%) but it was not significantly different from the Se utilisation in the other soils. In contrast, plant Se utilisation with the inorganic Se fertiliser varied between 10 and 24% in this experiment. Neither pH treatments nor soil organic matter contents had any effect on plant utilisation of Se from organic Se fertiliser.





## Discussion

Effect of organic matter and pH on Se availability

The differences in grain Se concentration at the same Se application rate in the field experiment are mainly caused by differences in Se availability in the soil since neither Se application nor soil organic matter content had any significant effect on grain yield. Up to 60% of the variation in Se content in the field experiment could be explained by differences in organic matter content. Generally, higher soil organic C and thus organic matter content resulted in higher grain Se concentration (Table 3). This result is in contrast to what was found by Johnsson (1991) and Gustafsson and Johnsson (1992) and opposite to the effect hypothesised. The soil organic matter contents at the different trial sites varied between approximately 3 and 8% (1.5-4.1% organic C, Table 1). Although not emphasised as a main result, also Johnsson (1991) actually found that a relatively small increase of organic matter from 4.9 to 6.9% in a clay soil increased plant Se uptake. The soil organic matter contents of the soils in the field experiment were in the same range as those of the loam and the peat/loam (approximately 4.4 and 12.4%, respectively; or 2.2 and 6.2% organic C, Table 2) used in the greenhouse experiment.

In the greenhouse experiments, we found similar correlations when considering these two soil types (data not shown). However, in the peat soil without inorganic binding places, Se uptake was no longer positively correlated with organic matter content. Also in Johnsson's (1991) study, Se uptake in wheat plants decreased clearly with increasing amounts of peat in pots containing sand/peat mixtures at two different pH levels (approximately pH 5 and 6.7).

The effect of pH on plant Se uptake seems to be dependent on the soil content of organic matter. In the greenhouse experiments, plants grown in the loam soil with its relatively low organic matter content, did not take up more Se when pH increased from pH 5.2 to pH 5.9, whereas they took up significantly more Se with a pH change from 5.9 to 6.4 (Table 4). This is in accordance with several studies conducted in mineral soils with a relatively low organic matter content (e.g., Cary and Allaway 1969; Lindberg and Bingefors 1970; Johnsson 1991). The effect is expected since selenate is present to a greater extent at near-neutral pH values and is less effectively bound in the soil compared to selenite (Gissel-Nielsen et al. 1984). Even if—as in our experiments—selenate is added, it

will be reduced to selenite in a more acid environment. Selenite is effectively bound to hydrous sesquioxides and to pH-dependent charges on clay minerals. With increasing pH, the adsorption of Se in soil generally decreases due to a decreasing amount of positive charges on edges of clay minerals and sesquioxides. Thus a higher plant availability of Se can generally be expected in mineral soils with increasing pH.

The plant Se uptake in the peat/loam soil mixture did not change with increasing pH. However, the two pH levels tested for this soil type differed only little (pH 5.7 and 6.2). In the peat soil, Se uptake decreased significantly with increasing pH. This effect has also been described by Johnsson (1991) for soils high in organic matter. In a peat soil (39% organic matter), Se uptake was smaller at the high than at the low pH, whereas the opposite occurred in soils with organic matter contents between 1.4 and 6.3%. The effect was at least partly attributed to differences in plant growth since the yield at low pH in the unmixed peat soil was only approximately half of that achieved at high pH (Johnsson 1991). Still our results show a similar trend. In general, anions mainly bind to mineral surfaces and only to a little extent to surfaces of organic matter (McBride 1994). However, Gustafsson and Johnsson (1992) found that added mineral Se was quickly bound to the O<sub>i</sub> horizon of a forest floor.

In another study, Gustafsson and Johnsson (1994) suggested that the process of Se retention in organic matter might be primarily due to microbially-mediated reductive incorporation, in the course of which the Se anions are reduced to low valence states and then incorporated into low-molecular-weight humic substance fractions. This may explain why an increase in pH does not have the same effect in an organic soil type such as peat as in a mineral soil type. With increasing pH, selenite bound by positive charges on sesquioxides or clay particles in a mineral soil will become more available, whereas Se incorporation into organic matter may increase due to higher microbial activity. Raising the pH may therefore not be a management practise improving Se availability in soils rich in organic matter.

## Leaching

The leaching experiment conducted in the greenhouse showed similar tendencies with respect to Se mobility as plant Se uptake. In the peat soil, leaching losses differed only little between the two pH levels, with a slightly higher loss occurring at low pH. This may confirm that Se was more effectively incorporated into soil organic matter at high pH. The opposite trend was found in the loam soil. In addition to a higher plant Se uptake in the growing season, more Se was leached from the pots after the growing season at pH 6.3, showing that Se was more easily available than at lower pH. In the leaching experiment, the lower pH level was quite low in the loam soil (pH 4.9), which may explain why only 1.3% of the applied Se was leached. Since most of the Se is present as selenite at lower pH in aerobic soils and the amount of binding sites for selenite increases with decreasing pH, little selenite is available in solution. At high pH, more Se is present as selenate, which is not bound strongly in the soil and therefore leached to a larger extent (16% of applied Se).

Inorganic versus organic Se

Application of organic Se sources did not contribute to higher Se availability for the plants in our greenhouse experiment. No differences in Se uptake were found between the control (no Se) and the organic Se treatments. Not even pure Se methionine, which is considered an easily available Se form, showed any effect on Se content in wheat on our experiments. A study by Abrams et al. (1990), on the other hand, showed that wheat plants can acquire Se methionine from a nutrient solution and that this acquisition is under metabolic control.

An earlier study with the same experimental set-up as described in this paper, but with a Se methionine dose 1000 times higher than in the experiments described above, resulted in considerably higher Se contents in wheat plants (data not shown). This suggests that plants are able to take up Se methionine also from the soil but the dose necessary to have an effect on plant Se concentration is much higher than for inorganic Se. Selenium methionine may represent an easily available C source for microorganisms. It is possible that Se from the low amount of Se methionine given in our experiments is quickly inserted into microbial proteins or lost to the atmosphere in the course of microbial decomposition before plant roots can make use of it.

## Conclusions

In accordance with earlier findings, an increase in pH from pH 5.9 to 6.4 in a loam soil with a relatively low organic matter content, led to an increase in Se availability. Further, it was found that in the same soil, an increase in soil organic matter content also increased plant availability of inorganic fertiliser Se.

In a soil rich in organic matter, lower pH resulted in higher plant Se availability. The reason for this different effect lies most likely in the type of binding of Se in the different soil types. In mineral soil, an increase in pH decreases the amount of pH-dependent binding sites. At the same time, a more neutral environment increases the amount of selenate at the expense of selenite. In an organic soil, Se may quickly be incorporated in rather than bound to organic matter and thus the amount of Se retained may be dependent on conditions for microbial activity. Raising the pH does not seem to be a management practise improving Se availability in soils rich in organic matter.

Selenium methionine as an organic Se source that is considered to be easily available, had no positive effect on Se concentrations in wheat plants, at least when applied in similar amounts as inorganic Se, most likely due to competition from soil microorganisms for these compounds.

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