# Evaluating of soil sulfur forms changes under different fertilizing systems during long-term field experiments

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#### ABSTRACT

The aim of this work was to evaluate the changes of different soil sulfur forms during long-term field experiments supplied with sewage sludge (SS), farmyard manure (FYM) or mineral fertilizers. The experiment was established at three different sites with the crop rotation of potatoes, wheat and barley. Fertilizing system was based on the same nitrogen dose per the whole crop rotation. Soil samples from the beginning of the experiment (1996) and at the end of each crop rotation were analysed after all three crops harvest. Similar tendencies of changes in water extractable ( $S_w$ ), adsorbed ( $S_{ads}$ ), 1 mol/L HCl estimated ( $S_{HCL}$ ) sulfur and their sum of ( $\Sigma$ S) related to the fertilizing systems were observed among all studied sites. A decreasing tendency of  $\Sigma$ S in soil was observed during the experiment (except for the year 2005). The biggest decrease was always visible compared to control treatment. Mineral fertilizers (S added with superphosphate) appeared to be the best source of  $S_{HCL}$ . Application of SS increased the  $S_w$  content in soil samples after potatoes harvest, whereas a decreasing tendency among the following crops was visible. Contents of other S forms ( $S_w$  at control, FYM and mineral fertilizers included) were usually balanced during the crop rotation.

Keywords: plant nutrient; fertilization; sulfur fractions; sulfur deficiency; essential element

For the future, it is important to further understand the influence of fertilization on the dynamics of plant nutrients in the soil as well as its influence on the plant growth. The content of bioavailable nutrients in soils varies not only depending on the fertilizer used, but also on the seasonal influence, soil properties and many other factors. Therefore, the data of the long-term field experiments provide valuable information for optimizing fertilization applications (Johnston 1997, Scherer 2001, Hejcman et al. 2012, Neugschwandtner et al. 2014).

Mineral sulfur (S) deficiency is an actual problem in many sites (Eriksen 2005, Lehmann et al. 2008, Balík et al. 2009, Scherer 2009 and others). The content of mineral sulfur forms in soil rapidly decreased in last 25 years (Balík et al. 2009, Scherer 2009). Although sulfur has been known as an essential element for plant growth for over two centuries, intentionally

S fertilizing was not realized before 1980's. At that time the most of the cultivated plants did not show evidence of S deficiency, because of air pollution from industry, intensive application of mineral fertilizers containing S as the ballast substance, application of manure and cropping of plant cultivars with lower S uptake in crop rotation (Zhao et al. 1999, Scherer 2001, Blair 2002).

From the mentioned reasons it may be expected that sulfur becomes a limiting factor for yield formation and product quality (Eriksen et al. 2004). Therefore, it is necessary to pay attention to the sulfur fertilization possibilities under different soil-climatic conditions.

The total soil sulfur appears in two basic forms: (i) organic and (ii) inorganic. Inorganic sulfur, on which is mainly focused this manuscript, represents usually less than 10% from total S (Tisdale et al.

1993, Wang et al. 2006, Förster et al. 2012) and appears in different forms. The most important form for S uptake by plants is bioavailable sulfate ( $SO_4^{2-}$ ) in the soil solution. This form represents usually only about 1% of total S (Knauff 2000). Another form is sulfate adsorbed to weak binding sites on the surface of soil particles. It directly influences the perseveration of the sulfate balance with the soil solution (McLaren and Cameron 1996). The most stable mineral S form represents sulfur co-precipitated with calcium and magnesium carbonates (Tisdale et al. 1993, Chen et al. 1997). Mentioned forms are possible to estimate using different sequential extraction methods, for example after Shan et al. (1992).

Therefore, the aim of this work is to evaluate the changes of water extractable ( $S_w$ ), adsorbed ( $S_{ads}$ ) and 1 mol/L HCl estimated ( $S_{HCL}$ ) sulfur forms under different fertilizing systems (sewage sludge (SS), farmyard manure (FYM) and mineral fertilizer) compared to the zero control during the long-term field experiments.

### MATERIAL AND METHODS

The long-term field experiment was established at experimental fields of the Czech University of Life Sciences in Prague and Crop Research Institute in Prague-Ruzyně (Czech Republic). The basic characteristics of the sites are given in Table 1.

Potatoes, winter wheat and spring barley were cropped in crop rotation on small plots (60 m<sup>2</sup>). All three crops were cropped parallel each year of the experiment. Organic fertilizers were applied only to the potatoes in crop rotation. The total amounts of N, P, K, Ca, Mg and S added per one

crop rotation in different fertilizing treatments are summarized in Table 2.

For this experiment, SS from a wastewater treatment plant in Prague-Troja was used. The whole system of fertilizing is based on the uniform amount of 330 kg N/ha per 3 years (expect of unfertilized control).

Archive air-dried soil samples (< 2 mm) from the beginning (year 1996) of the experiment and from the end of each crop rotation (years 1999, 2002, 2005, 2008, 2011 and 2014) were chosen for analysis. All samples were taken after harvest of each single crop in crop rotation and before fertilizers application. Each sample was analysed in two replications. The samples were extracted to obtain water extractable, 0.032 mol/L NaH<sub>2</sub>PO<sub>4</sub> extractable adsorbed and 1 mol/L HCl extractable sulfur fractions (Morche 2008, Kulhánek et al. 2011). All extracts were measured using inductively coupled plasma-optical emission spectroscopy (ICP-OES, Varian VistaPro, Mulgrave, Australia). The content of total estimated sulfur ( $\Sigma S$ ) was calculated like a sum of S<sub>w</sub>, S<sub>ads</sub> and S<sub>HCL</sub>.

For the basic statistical evaluation and excluding the outlaying observations, Statistica software was used (StatSoft Inc. 2015). Thereafter, the tendencies in sulfur fractions changes were evaluated in Microsoft Office Excel (Excel 2007).

## RESULTS AND DISCUSSION

The contents of different sulfur forms at the beginning of the experiment (year 1996) are mentioned in Table 3. The values of  $S_w$  were very similar among the studied sites (about 11 mg  $S_w/kg$ ). The amounts of  $S_{ads}$  ranged between 5.97 and 9.44 mg

Table 1. Characteristics of experimental fields

	Humpolec	Hněvčeves	Suchdol
Latitude	49°33'15"N	50°18'46''N	50°07'40''N
Longitude	15°21'02"E	15°43'3"E	14°22'33"E
Altitude (m a.s.l.)	525	265	286
Mean yearly temperature (°C)	7.0	8.2	9.1
Mean yearly rainfall (mm)	665	573	495
Soil type	Cambisol	Luvisol	Chernozem
Soil texture	sandy loam	clayey loam	sandy loam
$pH^1$	5.1	5.9	7.5

<sup>&</sup>lt;sup>1</sup>Estimated in 0.01 mol/L CaCl<sub>2</sub>, 1:10 w/v

Table 2. Application rates of macronutrients (kg/ha) – 3-year cycle

Treatment		Potatoes	Wheat	Barley
Control	N, P, K, Ca, Mg, S	0, 0, 0, 0, 0, 0	0, 0, 0, 0, 0, 0	0, 0, 0, 0, 0, 0
Sewage sludge	N	$330^{1}$	0	0
	P	$201^{1}$	0	0
	K	$55^{1}$	0	0
	Ca	$268^{1}$	0	0
	Mg	$70^{1}$	0	0
	S	$81^{1}$	0	0
Farmyard manure	N	$330^{1}$	0	0
	P	$70^{1}$	0	0
	K	$352^{1}$	0	0
	Ca	$250^{1}$	0	0
	Mg	$30^{1}$	0	0
	S	$73^{1}$	0	0
Mineral fertilizers <sup>2</sup>	N	120	140	70
	P	30	30	30
	K	100	100	100
	Ca, Mg	36, 0	42, 0	21, 0
	S	6.5	6.5	6.5

<sup>1</sup>P, K, Ca, Mg and S dose was calculated based on the average nutrient content in organic fertilizers. N application rate was based on the fertilizers analyses. <sup>2</sup>Mineral fertilizers used: N – calcium ammonium nitrate (27% N); P – triple superphosphate (21% P, 3% S); K – potassium chloride (50% K)

 $S_{ads}/kg$ . The biggest differences were found in the  $S_{HCL}$  contents, where the values reached from 5.35–22.8 mg  $S_{HCL}/kg$ . The highest  $S_{HCL}$  contents (Suchdol and Hněvčeves sites) are also reflected in the differences among the  $\Sigma S$  contents. Due to the used ICP-OES method to estimate different sulfur forms, it has to be taken into account that not only inorganic sulfur forms, but also some organic S compounds were estimated (Kowalenko and Grimmett 2007). For this reason, the better way is to use ion chromatography like e.g. in Förster et al. (2012). Anyway, the ICP-OES method was used, because for our purposes it can be successfully used as well (Kowalenko and Grimmett 2007, Morche 2008, Kulhánek et al. 2011).

In the further evaluation of the experiment (in order to eliminate the plots variability) the data from the year 1996 (before fertilizing) were converted to 100%. The data from the following years were then compared with the year 1996. For the better transparency, the average contents of studied sulfur forms were calculated for each evaluated year. The changes of estimated soil sulfur forms related to different crops in crop rotation and terms of the fertilizers application are mentioned further.

Figure 1 shows the changes in estimated sulfur fractions at Suchdol site. The  $S_{\rm w}$  content was always the highest in the SS treatment. This treatment is followed by fertilizing with FYM. It was probably

Table 3. The contents of estimated sulfur forms in the year 1996 (mg S/kg soil)

	S <sub>w</sub>	$S_{ads}$	S <sub>HCL</sub>	ΣS
Suchdol	10.40 ± 0.98	$7.46 \pm 0.72$	22.8 ± 4.64	40.7 ± 5.00
Humpolec	$12.3 \pm 0.77$	$9.44 \pm 0.72$	$15.6 \pm 1.03$	$37.3 \pm 1.75$
Hněvčeves	$10.5 \pm 0.76$	$5.97 \pm 0.38$	$5.35 \pm 0.54$	$21.8 \pm 1.19$

 $S_w^{}$  – water extractable;  $S_{ads}^{}$  – adsorbed;  $S_{HCL}^{}$  – 1 mol/L HCl estimated;  $\Sigma S$  – total estimated sulfur

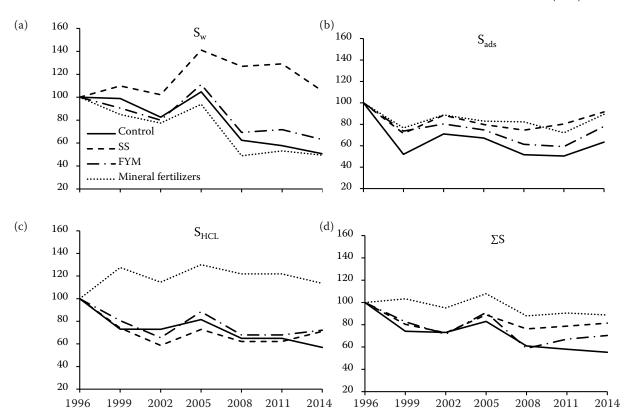


Figure 1. Changes of sulfur contents (%) at Suchdol site between 1996 and 2014.  $S_w$  – water extractable;  $S_{ads}$  – adsorbed;  $S_{HCL}$  – 1 mol/L HCl estimated;  $\Sigma S$  – total estimated sulfur

due to the slightly higher S dose applied using SS and better SS mineralization described further.

The lowest contents of S<sub>w</sub> were always found in the treatment fertilized with mineral fertilizers. It is the treatment where the highest yields of all studied crops were reported (Černý et al. 2010). Therefore, the higher yields probably resulted in the higher sulfur uptake by plants and lower contents of S<sub>w</sub> in comparison to the control treatment. Furthermore it is possible to assume that sulfate applied with mineral fertilizers is more prone to leaching and that S sequestration depends on both the fertilizer type and S application rate (Förster et al. 2012). This was confirmed at Hněvčeves site as well, but not at Humpolec site. The S<sub>w</sub> and S<sub>ads</sub> contents estimated at Humpolec site were almost always higher compared to Suchdol and Hněvčeves. Therefore the bioavailable sulfur content at Humpolec site was probably more sufficient to cover the plant uptake and unused sulfur added with mineral fertilizers caused the higher values of S<sub>w</sub> compared to control.

The contents of  $S_{ads}$  were relatively low and ranged between 3.80 and 10.6 mg  $S_{ads}/kg$ . There

were no big differences between the studied fertilizing treatments. The lowest  $\boldsymbol{S}_{ads}$  contents were always found in the control treatment without fertilizing. The rest of the studied treatments showed almost the same tendencies. It can be partially explained due to the fact, that SO<sub>4</sub><sup>2-</sup> adsorption is strongly pH-dependent and increases with decreasing pH (Förster et al. 2012). In our case, the lowest pH after 14 years of experiment was measured in the control treatment at Suchdol site. Similar results were observed at Lukavec site (soilclimatic conditions are comparable with Humpolec site) where the lowest pH values were measured at mineral fertilizers and control treatment. Only at Hněvčeves site, the highest pH decrease was observed at mineral fertilizers treatment followed by SS and control (Vašák et al. 2015). The second reason for the lowest  $S_{ads}$  contents in the control could be the lowest S input at this treatment. Location and climatic conditions of different sites play important role as well (Förster et al. 2012).

The  $S_{HCL}$  form in soil showed interesting tendencies. The amounts of  $S_{HCL}$  among the FYM, SS and control treatment were very similar dur-

ing the studied period, but the highest content was always found in the treatment fertilized with mineral fertilizers. This is probably due to the fact that almost whole sulfur added in the form of superphosphate (6.5 kg S/ha/year) was in the calcium sulfate form. This tendency was confirmed at all studied sites. Suchdol site has about 2.5% of carbonates; sulfur estimated using 1 mol/L HCl was probably mostly in Ca and Mg coprecipitated form. However, the results of Morche (2008) show that soils with less than 1% CaCO<sub>3</sub> provide evidence that most of the S extracted with 1 mol/L HCl is organically bound sulfur. Moreover, some other HCl soluble minerals (e.g. Fe and Al oxides and sulfates), mainly in non-calcareous, acidic soils can contribute to this S fraction as indicated by Chen et al. (1997).

The sum of estimated sulfur forms was influenced by the high amount of  $S_{HCL}$  in the mineral fertilizers treatment. There is a visible decreasing tendency of  $\Sigma S$  content in soil during the experiment, especially in the control treatment. It can be explained by the fact that the sulfur inputs to the soil strongly decreased at the end of  $20^{th}$  century (Balík et al. 2009, Scherer 2009).

Figure 2 shows the tendencies in mineral soil sulfur changes at Humpolec site during the experiment. The highest  $S_w$  contents were found in the SS treatment. This treatment was again followed by FYM. Compared to Suchdol site, the lowest  $S_w$  content was always obtained in the control treatment; probable reasons are discussed above.

SS also appeared as a good source of  $S_{ads}$ . Only in this treatment, the level of  $S_{ads}$  was higher in the end of the experiment compared to its beginning. The differences among following fertilizing treatments (control, FYM and mineral fertilizers) were not clearly visible during the whole experiment.

The average content of  $S_{HCL}$  (calculated for control) during the whole experiment was 10.5 mg  $S_{HCL}$ /kg. It is the value about 7.5 mg  $S_{HCL}$ /kg lower compared to Suchdol site. The soil from Humpolec site contains less than 0.35% of carbonates. As discussed before, estimated sulfur was probably mainly in organic form (Morche 2008). Nevertheless, the  $S_{HCL}$  contents were always the highest in mineral fertilizers treatment in each studied site and year. Therefore, it is assumed that the differences between this treatment and others were mainly caused

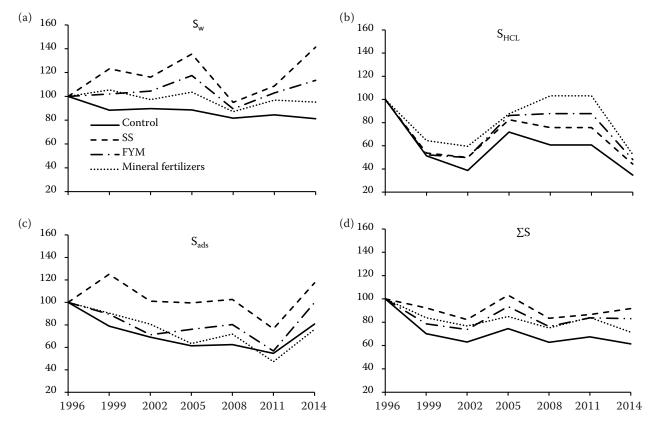


Figure 2. Changes of sulfur contents (%) at Humpolec site between 1996 and 2014.  $S_w$  – water extractable;  $S_{ads}$  – adsorbed;  $S_{HCL}$  – 1 mol/L HCl estimated;  $\Sigma S$  – total estimated sulfur

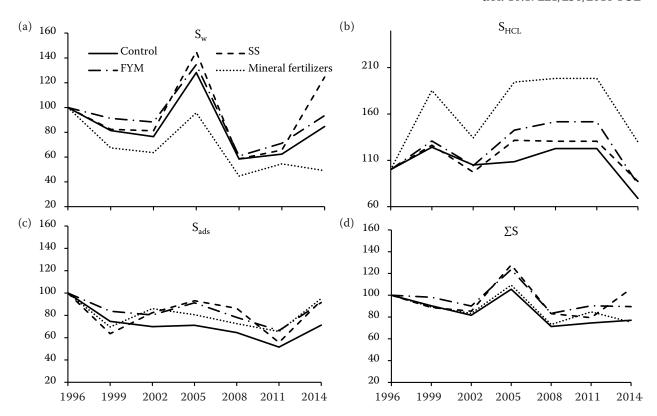


Figure 3. Changes of sulfur contents (%) at Hněvčeves site between 1996 and 2014.  $S_w$  – water extractable;  $S_{ads}$  – adsorbed;  $S_{HCL}$  – 1 mol/L HCl estimated;  $\Sigma S_{HCL}$  – total estimated sulfur

due to the calcium-bound sulfates contained in superphosphate estimated with 1 mol/L HCl as well. This was also confirmed at Hněvčeves site, which has about 0.7% of carbonates. This fact partially prove the statement of Kulhánek et al. (2011) who estimated the increasing content of occluded sulfur in soil after calcium sulfate S application at three different sites in three year experiment.

Figure 3 shows the results from Hněvčeves site. The differences between studied years are the most visible here. Especially the contents of  $S_w$  sulfur in the year 2005 are higher compared to other years. In spite of this fact, the differences among the fertilizing treatments are clearly visible as well as on the other sites. The lowest contents of  $S_w$ , like at Suchdol site, were always found in the mineral fertilizers treatment. Thereafter follows the control treatment. Treatments fertilized with SS and FYM showed quite similar results, which were higher than mineral fertilizers and control.

Control treatment showed usually the lowest values of  $S_{ads}$ . Differences between other fertilizing systems were not clearly visible. Curtin et al. (2007) studied the influence of long-term applica-

tion of S in the form of single superphosphate on the content of  $S_{\rm ads}$  in long-term experiment. They found the significantly increasing tendency with both superphosphate doses (7 and  $14\,\mathrm{kg}$  S/ha/year) against zero control. In our experiments, only 6.5 kg S/ha/year was added as superphosphate. Therefore, the differences between control and mineral fertilizers were not so clearly visible, but the  $S_{\rm ads}$  contents in the mineral fertilizers treatment were almost always higher in comparison to control among all three studied sites and seasons.

The studied S forms content increase is typical for all three sites in the year 2005. It was probably caused due to the weather conditions in this year, which were optimal for the mineralization of the organic sulfur sources (higher rainfall in July and August together with higher temperatures in September) (CHMI 2016). It confirms the role of climatic conditions on the soil S fractions contents mentioned in Förster et al. (2012).

The differences between soil sulfur contents related to the crops in crop rotation were also evaluated. Almost in all cases, the highest contents of  $S_w$  were clearly visible in samples taken

up in the SS treatment after potatoes harvest. At the next crops in crop rotation (wheat and barley) a decreasing tendency follows. This was caused because SS was always applied only before the potatoes for the whole crop rotation. This tendency appeared in a lesser extent among the  $S_{ads}$  and  $\Sigma S$  as well, but not among the S<sub>HCL</sub>. A similar result was expected during FYM fertilizing, but the contents of three measured S forms did not change during the crop rotation. It is probably due to the fact that in the FYM sulfur is in more stable organic forms compared to SS. That confirms the study of Lima et al. (2009), who estimated the long-term influence of SS or FYM application and found significantly higher organic matter (especially lignin and lignin-like products) accumulation in the FYM treatment. The second reason of lower S<sub>w</sub> content among the FYM treatment can be the fact that its long-term application led to the S accumulation in the dry soil aggregates (Yang et al. 2007) whose size increased with the increasing organic fertilizers rate (Scherer et al. 2012). After Scherer et al. (2012) the soil particle size plays the key role in the S availability, where the content of plant available S decreases with the increasing soil particle size, while the influence of the kind and amount of used organic fertilizers (FYM, SS and composts) is negligible. The above-mentioned fact also partially confirms the results of Odlare et al. (2008) who compared the microbial activity in soil after SS or cattle manure application. They found slightly higher microbial activity in soil after SS application. Based on the results of Sastre et al. (1996), SS had a positive influence on microbial activity in soil and due to this process on the organic matter decomposition as well. According to Tabatabai and Chae (1991) SS has also more optimal C:S ratio for organic compounds mineralization compared to FYM. This confirmed also Förster et al. (2012) who stated that SS has a large pool of potentially mineralizable S. It was probably the reason for S<sub>w</sub> content increase in the SS treatment after potatoes harvest. As expected, the content of studied S forms did not change among control and mineral fertilizers treatments during the crop rotations.

Based on the results of our three long-term field experiments it can be concluded that a decreasing tendency of  $\Sigma S$  content is visible in the period from 1996 to 2014 (except for the year 2005). The biggest decrease is always visible in the control

treatment. SS application led to the highest  $S_{\rm w}$  contents compared to the other fertilization treatments. Mineral fertilizers treatments appeared to be the best source of  $S_{\rm HCL}$ . The contents of studied sulfur forms in soil were (especially in the year 2005) influenced by the local climatic conditions as well.

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