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Study on the Effect of Sulphur, Glucose, Nitrogen and Plant Residues on the Immobilization of Sulphate-S in Soil

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Abstract: In order to evaluate the relationship between sulphur (S), glucose (G), nitrogen (N) and plant residues (st), on sulphur immobilization and microbial transformation. Five soil samples from 0-30 cm of Bastam farmer's fields of Shahrood area were collected. Eleven treatments with different levels of S, G, N and plant residues (wheat straw) were applied in a randomized block design with three replications and incubated over 20, 45 and 60 days. The immobilization of SO_4^{2-} -S presented as a percentage of that added, was inversely related to its addition rate. Additions of glucose and plant residues increased with the C-to-S ratio of the added amendments, irrespective of their origins (glucose and plant residues). In the presence of C sources (glucose or plant residues). N significantly increased the immobilization of SO_4^{2-} -S, whilst the effect of N was insignificant in the absence of a C amendment. In first few days the amounts of added SO_4^{2-} -S immobilized were linearly correlated with the amounts of added S recovered in the soil microbial biomass. With further incubation the proportions of immobilized SO_4^{2-} -S remaining as biomass-S decreased. Decrease in biomass-S was thought to be due to the conversion of biomass-S into soil organic-S. Glucose addition increased the immobilization (microbial utilization and incorporation into the soil organic matter) of native soil SO_4^{2-} -S. However, N addition enhance the mineralization of soil organic-S, increasing the concentration of SO_4^{2-} -S in soil and the extent to which available-S can be immobilized is determined by both the amount of available-S and the availability of an utilizable C source.

Key words: Fumigation, extraction, microbial biomass, turnover

INTRODUCTION

Sulphur (S) immobilization in soil is the process through which mineral S is incorporated into soil organic compounds (Wu *et al.*, 1994; Eriksen, 2005). This process together with the mineralization of organic-S regulates the accumulation and cycling of S in the soil and affects S availability to plants (O'Donnell *et al.*, 1994). This immobilization is believed to be microbially mediated which includes the conversion of S into the microbial biomass (microbial utilization). However, the dynamics of the immobilization process and the mechanisms through which immobilization is associated with microbial transformations (Chowdhury *et al.*, 2000), the utilization and turnover of S remain poorly understood. Recent developments in methodologies for measuring soil microbial biomass-S (Wu *et al.*, 1994) have facilitated progress towards the quantification of S transfer (rate and magnitude) between SO_4^{2-} -S microbial biomass-S and organic-S pools in soil. Wu *et al.* (1993) have shown that S immobilization in soils amended with plant residues (barley straw or rape leaves) is highly correlated with increases in microbial biomass-C. In a separate study using six soils with different properties, it was shown that

the amounts of added SO_4^{2-} -S immobilized were comparable with those converted into the microbial biomass over the first few days (O'Donnell *et al.*, 1994; Permanen *et al.*, 2004). The subsequent incorporation of the immobilized SO_4^{2-} -S into the soil organic matter was thought to depend on the turnover of the soil microbial biomass.

Furthermore, the study of Bhupinderpal *et al.* (2006) demonstrated that the immobilization rates of SO_4^{2-} -S were influenced by soil properties such as clay and organic matter contents, the size of the microbial biomass and the available S. However, the extent to which these factors interact and affect the immobilization rates of S remained unknown.

Dynamics of the SO_4^{2-} -S immobilization using amendments containing different rates of SO_4^{2-} -S, different form of carbon (glucose, straw) and N nutrient were studied by Wu *et al.* (1993). The initial immobilization rates of added SO_4^{2-} -S were correlated with the amounts converted to soil microbial biomass-S over first few days (O'Donnell *et al.*, 1994). Properties of decomposed biomass-S converted into the soil organic-S (incorporated into the soil organic matter) were determined on further incubation. Present objective was

to establish the quantitative relationship between the immobilization and microbial transformation of S and to estimate the effects of factors such as the amounts of $\text{SO}_4^{2-}\text{-S}$, the supply of C and N on the immobilization and availability of $\text{SO}_4^{2-}\text{-S}$ in soil. These data are essential in proving the hypothesis that the incorporation of $\text{SO}_4^{2-}\text{-S}$ into the soil organic matter via the microbial biomass is the primary mechanism for the immobilization of inorganic-S in soils (Bhupinderpal *et al.*, 2006; O'Donnell *et al.*, 1994; Pennanen *et al.*, 2004).

MATERIALS AND METHODS

Composite samples from 0-30 cm depth of the five farmer's field were collected, air dried and passed through 2 mm sieve and kept for 10 days in 25°C and 100% humidity. Soil physical and chemical properties were done by standard methods and are presented in Table 1.

Sixty gram of dried soil were weighed into 125 mL jars and amended with S, N, glucose and plant residues as shown in Table 2. Controls were left unamended. Sulphur as K_2SO_4 (S_{10} and S_{25} $\mu\text{g g}^{-1}$), N as KNO_3 and appropriate amounts of glucose and wheat straw were added to soil. For each treatment, a solution was prepared by dissolving the appropriate amounts of glucose, KNO_3 and K_2SO_4 in 100 mL distilled water. An aliquot of the solution (4 mL) was mixed with each soil portion. Before the addition of the solutions, plant residues (0.3 g) which had been dried (35°C) and ground were mixed with the soil portions as required. The control soil was treated with 4 mL distilled water to maintain equivalent moisture content to that of the amended soils. Following amendment, the soils were placed in 2.5 glass bottles, sealed and kept at 25°C and 100% humidity.

The contents of $\text{SO}_4^{2-}\text{-S}$ and the microbial biomass-S were determined after 20, 45 and 60 days of incubation. At each sampling, one portion of soil from each treatment was removed and subdivided by weighing (6×10 g) into centrifuge tubes (45 mL). Three of the sub-samples were fumigated for 24 h in CHCl_3 vapor (Wu *et al.*, 1994). The remaining three sub-samples were used as the controls and left unfumigated. All of the sub-samples were extracted in 10 mM CaCl_2 (20 mL) by shaking for 60 min at 400 rev. min^{-1} on an end-over-end shaker. Extracts were filtered through Whatman No. 42 filter paper and stored at -18°C prior to analysis.

Soil microbial biomass-S was measured by the procedures described by Wu *et al.* (1994). Total extractable-S was determined using Massoumi and Cornfield (1963) following conversion of organic-S in the extract into $\text{SO}_4^{2-}\text{-S}$. This was done by digesting an aliquot of the extracts (5 mL) in a 10 mL graduated glass tube for 24 h in a sand bath (160°C), using H_2O_2 (AR grade, 30% v/v, 1.5 mL). Total biomass-S was calculated from the relationship $B_s = F_s/K_s$, where F_s is the difference between total extractable-S in the fumigated soil and that in the control soil, K_s with the conversion factor (0.31), determined by Wu *et al.* (1994). Analysis of variance (ANOVA) was performed on all data sets. Data from all treatments were combined in correlation coefficient analysis. The statistical package excel were used and least significant differences (LSD, $p = 0.05$) were calculated using ANOVA.

RESULTS AND DISCUSSION

Immobilization of added sulphate-S: The amounts of added $\text{SO}_4^{2-}\text{-S}$ recovered from all of the treatments using

Table 1: Physical and chemical properties of the soils

Site	Clay (%)	OC (%)	pH	EC (mS cm^{-1})	Total S (mg kg^{-1})	Available S (mg kg^{-1})
1	33	0.88	7.65	0.74	278	1.4
2	42	1.05	7.44	0.59	311	3.4
3	31	0.79	7.26	0.61	252	3.2
4	37	0.74	7.64	0.83	195	2.6
5	44	0.88	7.48	0.79	205	3.6

Table 2: Description of the treatments

Treatments	$\text{SO}_4^{2-}\text{-S}$ ($\mu\text{g g}^{-1}$ soil)	$\text{NO}_3^-\text{-N}$ ($\mu\text{g g}^{-1}$ soil)	Glucose ($\mu\text{g g}^{-1}$ soil)	Straw- C ($\mu\text{g g}^{-1}$ soil)
Control	---	---	---	---
S_{10}	10	---	---	---
S_{25}	25	---	---	---
$\text{S}_{10}+\text{N}$	10	100	---	---
$\text{S}_{25}+\text{N}$	25	100	---	---
$\text{S}_{10}+\text{G}$	10	---	2500	---
$\text{S}_{25}+\text{G}$	25	---	2500	---
$\text{S}_{10}+\text{G}+\text{N}$	10	100	2500	---
$\text{S}_{25}+\text{G}+\text{N}$	25	100	2500	---
$\text{S}_{10}+\text{St}+\text{N}$	10	100	---	2500
$\text{S}_{25}+\text{St}+\text{N}$	25	100	---	2500

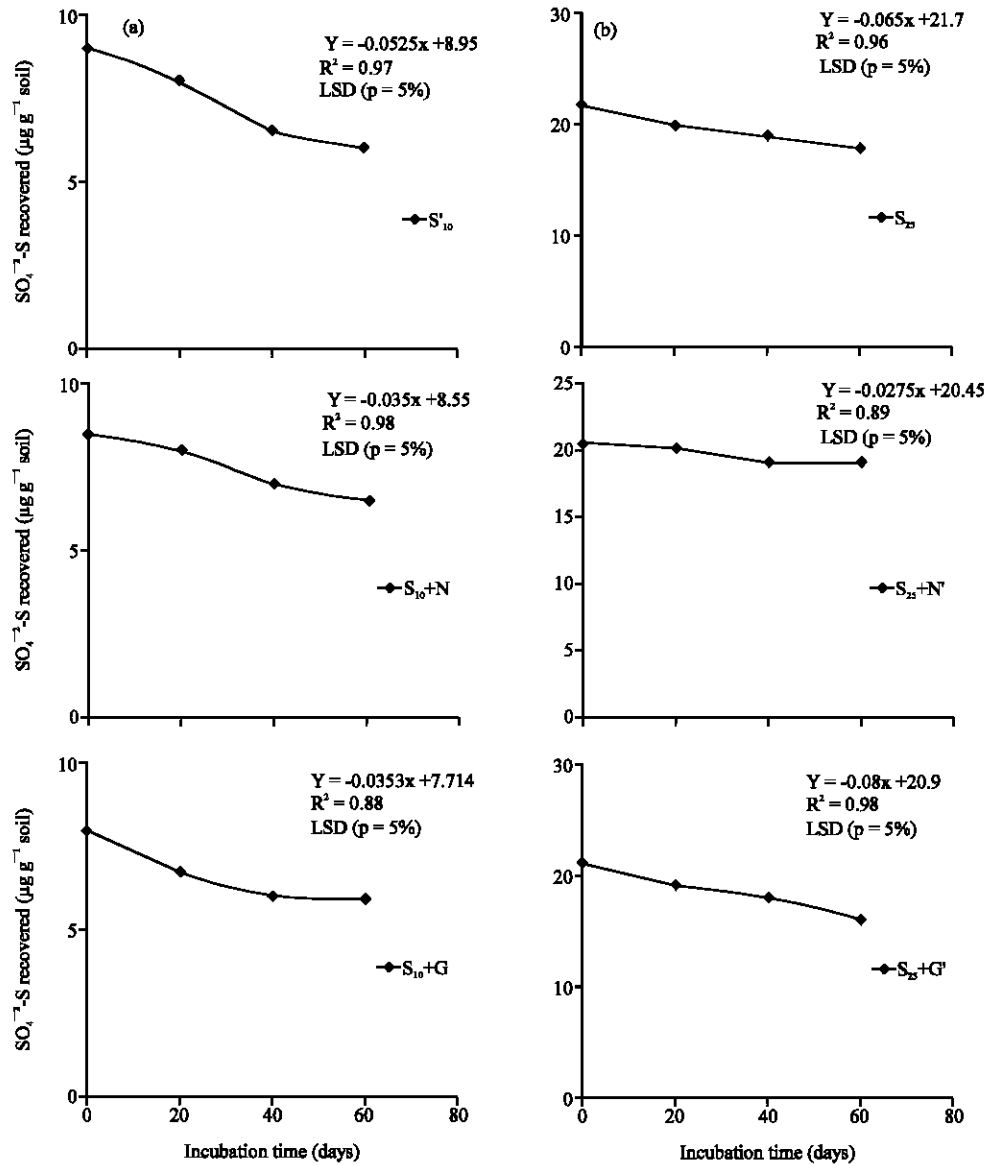


Fig. 1: Recovery of added $\text{SO}_4^{2-}\text{-S}$ (a) treatments of S_{10} , N and glucose (b) treatments of S_{25} , N and glucose

10 mM CaCl_2 decreased over 60 days incubation (Fig. 1). These decreased were similar in that they were initially rapid but became slow with extended incubation. This agrees with the findings of O'Donnell *et al.* (1994) who measured the immobilization of added $\text{SO}_4^{2-}\text{-S}$ in a number of soils with different properties. In this study results showed that decreases in the recovery of $\text{SO}_4^{2-}\text{-S}$ were greater in those soils receiving larger additions of amendments (S_{25} , $\text{S}_{25} + \text{G}$ and $\text{S}_{25} + \text{G} + \text{N}$ treatments, compared with S_{10} , $\text{S}_{10}+\text{G}$ and $\text{S}_{10}+\text{G} + \text{N}$ treatments (Table 2). The amount of $\text{SO}_4^{2-}\text{-S}$ immobilized (converted into soil microbial biomass-S or incorporated into soil

organic matter) in soil was, as expected, positively correlated with the addition rate. However, the extent to which $\text{SO}_4^{2-}\text{-S}$ was immobilized, as a percentage of addition, was inversely correlated to the addition rate. This is indicated clearly by the fact that the percentage of $\text{SO}_4^{2-}\text{-S}$ immobilized was smaller in those soils receiving the larger additions of $\text{SO}_4^{2-}\text{-S}$.

The addition of glucose enhances the growth of soil microbial biomass and can result in a net immobilization of soil $\text{SO}_4^{2-}\text{-S}$. In this experiment treatments with the addition of glucose markedly increased the immobilization of $\text{SO}_4^{2-}\text{-S}$, particularly over

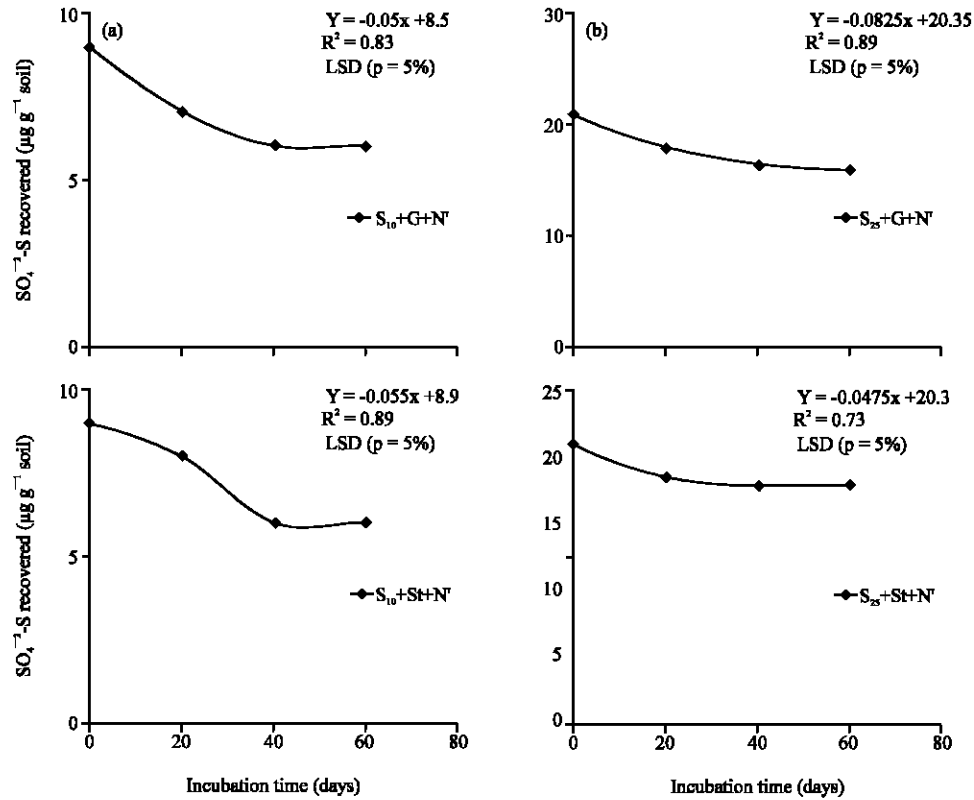


Fig. 2: Recovery of added $\text{SO}_4^{2-}\text{-S}$ (a) $\text{S}_{10} + \text{G} + \text{N}$ and $\text{S}_{10} + \text{St} + \text{N}$ treatments (b) $\text{S}_{25} + \text{G} + \text{N}$ and $\text{S}_{25} + \text{St} + \text{N}$ treatments

the first 10 days of incubation. For example, in treatment $\text{S}_{10} + \text{G} + \text{N}$ the recoveries of $\text{SO}_4^{2-}\text{-S}$ in first 10 days was 50% and by the end of incubation immobilization had decreased to over 35%. There was also an observable interaction between glucose and S in increasing the microbial utilization of $\text{SO}_4^{2-}\text{-S}$, since with increasing the S level the immobilization of S by the microbial utilization was greater. In contrast, in those treatments receiving no additional C source (S_{10} , $\text{S}_{10} + \text{N}$), the immobilization of $\text{SO}_4^{2-}\text{-S}$ was less than 15% throughout the incubation. These results were expected, as it has been shown that the addition of labial substrate such as glucose can result in a rapid increase in microbial biomass which requires more S nutrient from the soil (Ghani *et al.*, 1993; O'Donnell *et al.*, 1994; Pennanen *et al.*, 2004).

Based on the comparison of treatments S_{10} and $\text{S}_{10} + \text{N}$ and S_{25} and $\text{S}_{25} + \text{N}$, the addition of N without added C was unlikely to change significantly the immobilization of $\text{SO}_4^{2-}\text{-S}$ (Fig. 2). However, the combination of N with glucose was shown to have a positive effect on S immobilization in soil. As shown in Fig. 2, in the treatments of $\text{S}_{10} + \text{G} + \text{N}$ and $\text{S}_{25} + \text{G} + \text{N}$, the

immobilization of $\text{SO}_4^{2-}\text{-S}$ increased by 12- 15% (as a percentage of the addition rates) by 20 days of incubation, when compared with treatments $\text{S}_{10} + \text{G}$ and $\text{S}_{25} + \text{G}$. Thus, the supply of N can limit S immobilization in soil, particularly during rapid growth of the microbial biomass, as found following glucose addition confirming the finding of Pennanen *et al.* (2004) and Vong *et al.* (2008).

Additions of straw residues ($\text{S}_{25} + \text{St} + \text{N}$) increased the immobilization of $\text{SO}_4^{2-}\text{-S}$, compared with the treatment $\text{S}_{25} + \text{N}$ which provide an equivalent amount of $\text{SO}_4^{2-}\text{-S}$ and N but contained no residue amendment (Fig. 2). This was presumably a result of the rapid growth of the soil microbial biomass following the addition of plant residues (Pennanen *et al.*, 2004; Wu *et al.*, 1993). However, early in the incubation, the immobilization of $\text{SO}_4^{2-}\text{-S}$ was greater in the treatment using $\text{S}_{25} + \text{St} + \text{N}$ than in the treatment using $\text{S}_{10} + \text{St} + \text{N}$. This apparent discrepancy could be explained by the fact that the $\text{S}_{25} + \text{St} + \text{N}$ treatment has greater S content. However, the effect of plant residues on the immobilization of $\text{SO}_4^{2-}\text{-S}$ was much smaller than that of glucose, since the amounts of $\text{SO}_4^{2-}\text{-S}$ immobilized in

treatments $S_{25} + St + N$ were apparently less than those found in treatment $S_{25} + G + N$. Straw is considerably less labile and, at the same addition rate, might be expected to produce smaller increase in soil microbial biomass and S immobilization than either glucose or the straw residues (Pennanen *et al.*, 2004; Wu *et al.*, 1993). Earlier studies have shown that the incorporation of plant residues with narrow C-to-s ratio (<200:1) can increase the contents of SO_4^{-2} -S in the soil, whereas the incorporation of those plant residues which have wide C-to-S ratios (<400:1) may result in a net immobilization of soil inorganic-S, Pennanen *et al.* (2004) and Wu *et al.* (1994) have reported similar results. The extent to which S immobilization depends on microbial transformation S (microbial utilization and turnover) is reflected in the quantitative relationship between the amounts of SO_4^{-2} -S immobilized and those converted into measurements show a significant linear correlation over the 60 days incubation. Thus all of the results presented suggest that the addition of SO_4^{-2} -S has little effect on the transformation of native soil S confirming the results of O'Donnell *et al.* (1994). These results show a significant priming effect of N on the mineralization of soil S. Similar effect were also found by Ghami *et al.* (1992) and Pennanen *et al.* (2004). However, the addition of N had little effect on soil microbial biomass-S.

CONCLUSION

It can be concluded that there are close relationships between the immobilization of SO_4^{-2} -S and the microbial utilization and turnover of S. The data support the hypothesis that the primary mechanism by which SO_4^{-2} -S is incorporated into the soil organic matter is the microbial biomass. Since immobilization is dependent upon microbial utilization and turnover, the extent to which available-S (SO_4^{-2} -S) can be immobilized is determined by both the amount of available-S and the availability of an utilizable C source.

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