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Effects of organic matter on sulphur oxidation in soil and influence of sulphur oxidation on soil nitrification

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Summary The effects of wheat straw and pressed sugar beet pulp on sulphur oxidation were determined in a loam soil amended with 1% (w/w) elemental sulphur. Wheat straw stimulated the oxidation of elemental sulphur over the first 2 to 3 weeks of the incubation period, resulting in an increase in LiCl-extractable sulphate. After 4 to 7 weeks incubation however, the only significant increase in soil sulphate followed the 1% straw addition, while at week 7 sulphate concentrations in the 0.25% and 5.0% straw amended soils were lower than the control.

Pressed sugar beet pulp (1% w/w) initially stimulated the oxidation of elemental sulphur in the soil, but by weeks 3 to 7 of the incubation period rates of oxidation in pulp-amended soils were lower than the control. Towards the end of the incubation period however, sulphate concentrations in the amended soils exceeded the control values, significantly so by week 11. The concentration of thiosulphate and tetrathionate also increased in soils receiving sugar beet pulp.

Nitrification was inhibited in soils in which sulphur oxidation was actively occurring. Although possible alternatives are mentioned, such inhibition appears to result from a decrease in soil pH brought about by the oxidation of elemental sulphur to sulphuric acid.

Introduction

Sulphur deficiencies are now occurring in soils in many areas of the world including parts of Canada; Europe; the South West Pacific and the USA[18]. In arable soils of the Canadian Prairies, for example, approximately 12 million hectares are regarded as being potentially sulphur deficient[2]. The sulphur needs of crops are generally automatically met if superphosphate is used as the main phosphorus source[20]. However, reduced sources of sulphur, including elemental sulphur, are increasingly being applied as fertilizers to soils and vegetation[18]. These sulphur sources must first be oxidized to sulphate before they can be used by crop plants. The oxidation of sulphur is sub-optimal in some soils[15]. As a result it is becoming increasingly important that more is known about the factors which influence sulphur oxidation in soils.

Although the oxidation of elemental and reduced sulphur in soils has been widely studied, our knowledge of the factors which influence the process still needs to be improved[18]. For example, reports on the effects of organic matter on sulphur oxidation appear contradictory. Gleen and Quastel[5] found that organic matter stimulated sulphur oxidation in soil while Vitolins and Swaby[17] reported both stimulation and inhibition, but in most cases no effect. If organic matter does stimulate sulphur oxidation in soil then the addition of agricultural wastes, such as cereal straw, might be a useful means of increasing rates of sulphur oxidation in soils where the process is limited[15].

The effects of sulphur oxidation on the functioning of the nitrogen cycle in soil, particularly in relation to nitrification and denitrification, also warrants further study[18]. As nitrification is an acid-sensitive process[1], it is likely to be inhibited by the sulphuric acid produced by sulphur oxidation. However, although nitrification inhibition in sulphur-amended soils has been widely reported[18], it has not always been associated with a decrease in bulk-soil pH[9].

Here we describe studies on (1) the effects of wheat straw and pressed sugar beet pulp on sulphur oxidation in soil, and (2) the effect of sulphur oxidation on the nitrification of ammonium sulphate.

Materials and methods

Soil

A fertile loam soil (sieved to < 4mm) was used, with a total N and organic C content of 0.7 and 5.5% (w/w) respectively, and a pH of 7.0. Total N was determined by the macro-Kjeldahl method[7], while organic C was determined using the Walkley and Black method[7]. The soil was air dried at room temperature for 5 days prior to use.

Effect of organic matter on sulphur oxidation

Samples of the soil (100 g) were weighed into 250 ml Erlenmeyer flasks and adjusted to 20% (w/w) water content. Elemental sulphur (1% w/w, steam sterilised on 3 separate occasions) was then added and mixed thoroughly. In experiments using wheat straw the soil was then amended with filter sterilised (0.22 μm) ammonium nitrate at a concentration of $1.6\,g\,N\,100\,g^{-1}$ straw (the N concentration recommended for optimal straw decomposition[8]). Wheat straw was chopped to 5 cm lengths, the most common chop length found after combining[3], and then added to the soil to achieve a concentration range of 0 to 1% (w/w). In experiments using pressed sugar beet pulp, nitrogen (autoclaved $NaNO_3$) and phosphorus (filter sterilised, 0.22 μm, K_2HPO_4) were both added at 0.1% (w/w). Sugar beet pulp was ground to < 0.5 mm and then added at 1% (w/w).

Control flasks lacking added organic matter, but containing elemental sulphur plus the phosphorus and the nitrogen source where appropriate, were included. The flasks were covered with parafilm perforated with fine holes to allow for gaseous exchange without excessive water loss and incubated in triplicate at 25°C. The water content was then maintained at 20% (w/w) with sterile deionised water.

S-ions were determined by extracting the soil with 0.1 *M* LiCl (1:10 soil:LiCl ratio shaken for 15 minutes). After filtration, sulphate was determined turbidimetrically[7], and thiosulphate and tetrathionate by colorimetry[10].

Effect of sulphur oxidation on nitrification

Flasks were set up as before containing elemental sulphur-amended or unamended loam soil. No organic matter was added but nitrogen was supplied as filter sterilised ammonium sulphate ($0.22\,\mu g\,N\,g^{-1}$ soil). Flasks lacking ammonium sulphate but containing elemental sulphur, and control flasks which lacked all amendments, were included. All flasks were incubated in triplicate.

Nitrate was extracted using deionised water (extraction ratio 1:10 soil:water ratio shaken for 15 minutes). After filtration nitrate was determined using chromotropic acid[14], while sulphate was determined as above. Soil pH was measured using a glass electrode on a soil slurry (1:10 soil:water ratio shaken for 15 minutes).

Results and discussion

The effect of organic matter on the oxidation of elemental sulphur in the loam soil was determined by measuring the concentration of sulphate; thiosulphate ($S_2O_3^{2-}$-S) and tetrathionate ($S_4O_6^{2-}$-S)[11]. The addition of wheat straw to the soil (particularly at 1% w/w) stimulated the production of sulphate over the first 2 to 3 weeks of the incubation period (Fig. 1). However, with the exception of the 1% treatment, sulphate concentrations in the straw-amended soils were eventually lower than in the control soils lacking straw. Small, but statistically significant increases in sulphate concentration initially followed the addition of straw (Fig. 1); after three weeks, however, increases in the concentration of the ion of the order of 1000 μg^{-1} occurred in soils amended with 1.0 and 0.25% w/w straw. Similar increases then continued to occur for the full seven week incubation period in soils amended with 1% straw. Thiosulphate and tetrathionate were not detected in straw amended soils at any time during the 7 week incubation. The stimulation of sulphur oxidation in the soil following straw addition probably resulted from an increase in the activity of those sulphur-oxidising heterotrophs capable of using staw or its breakdown products as a carbon source to support sulphur oxidation. For example, the soil fungus *Trichoderma harzianum* can use straw as a carbon source to support sulphur oxidation *in vitro*[6]. It is therefore likely that fungi could

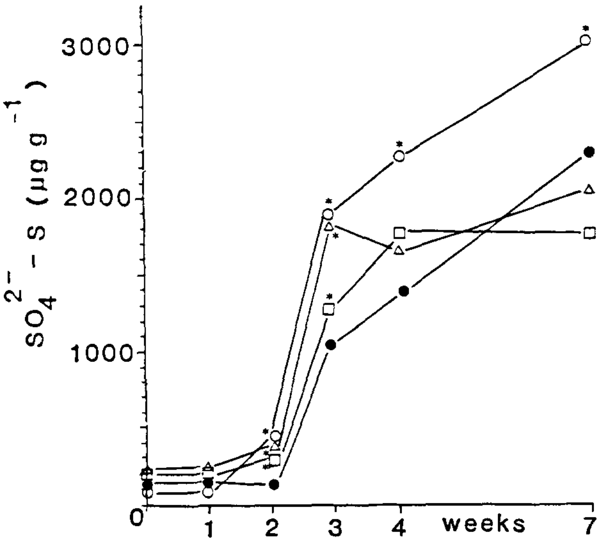

Fig. 1. Oxidation of elemental sulphur to sulphate in loam soil in the presence of 0% (●—●), 0.25% (△—△), 0.5% (□—□) and 1.0% (○—○) w/w wheat straw (* significant difference from control, $p = 0.05$).

use this resource as a carbon source to support sulphur oxidation in soil. Heterotrophic bacteria and actinomycetes[18,19] might also be able to oxidise sulphur in soil by using the breakdown products produced by straw decomposers such as *T. harzianum*. A rapid increase in the growth of heterotrophs and a consequent increase in microbial assimilation of sulphate might also explain why soil sulphate concentrations were eventually lower in the soils amended with 0.25 and 0.5% straw than in the unamended controls. Since overall microbial activity in soil, and particularly cellulose decomposition, is inhibited when sulphur is limiting[16], any sulphate produced by sulphur oxidation is likely to be rapidly immobilised. Fungi in particular appear to have a high requirement for sulphate[12].

The addition of pressed sugar beet pulp to the soil led initially to a slight stimulation in the oxidation of elemental sulphur to sulphate, which was followed by a depressive effect which lasted between weeks 3 and 7 (Fig. 2c). Sulphate concentrations then increased in the pulp-amended soils to significantly exceed the control by the eleventh week. Sugar beet pulp also stimulated thiosulphate and tetrathionate production, especially between weeks 1 and 3, and 3 and 5 respectively (Fig. 2a, b). However, sulphate concentrations over this period remained relatively low compared to those found in the wheat straw experiment (Fig. 1). This form of organic matter appears therefore to favour the microbial oxidation (presumably largely by heterotrophs) of elemental sulphur to sulphate via thiosulphate and tetrathionate, which accounts for the finding that sulphate concentrations were low in soils containing high concentrations of thiosulphate and tetrathionate (Fig. 2c). As sulphate concentrations in pulp amended soils did not exceed the controls until the seventh week it appears that sugar beet application would favour a slow, long-term release of sulphate into soil, while wheat straw addition would be useful where a more rapid release (*i.e.* 3 weeks after application) of the ion is required. The complex carbon sources used here are widely available and relatively cheap, and as a result might be economically applied with elemental sulphur to increase sulphur oxidation in soils in which the process is limited. However, the effects of these carbon sources would need to be carefully monitored in the field, otherwise a large proportion of sulphate released on sulphur oxidation may be assimilated by heterotrophs, leading to a reduction in the short term availability of the ion to crops.

The effect of sulphur oxidation on soil nitrification is shown in Fig. 3. Sulphate production increased steadily over the first three weeks of the incubation period and then increased more rapidly in soil amended with only elemental sulphur (Fig. 3b). However, the production of the ion was

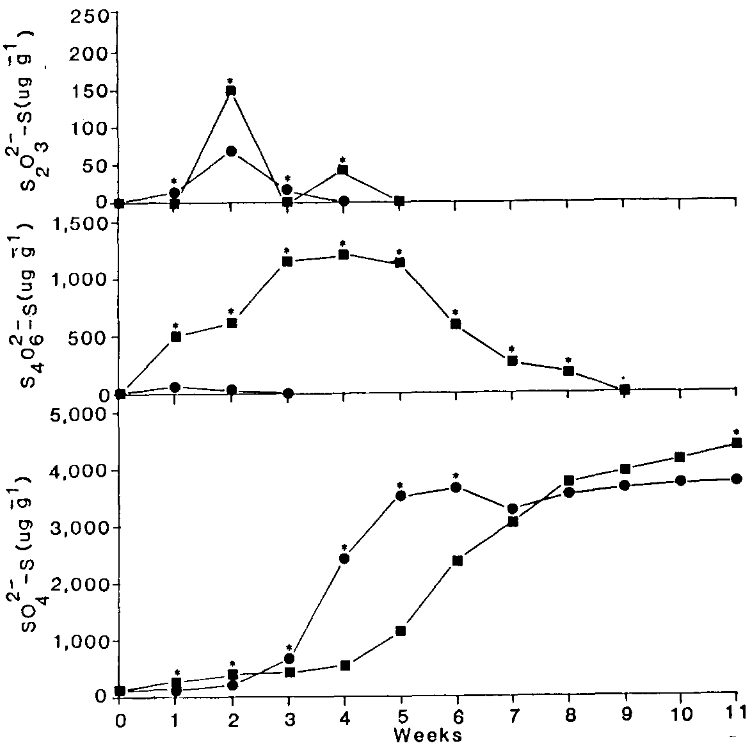

Fig. 2. Oxidation of elemental sulphur (1% w/w) to thiosulphate, tetrathionate and sulphate in loam soil in the presence of (■—■) and absence (●—●) of pressed sugar beet pulp (1% w/w) (* significant difference from control, $p = 0.05$).

reduced for 4 weeks when ammonium sulphate was added as the nitrification substrate, suggesting that ammonium inhibits the process in some way. The addition of ammonium sulphate led to a rapid increase in the concentration of nitrate in the soils (Fig. 3c) and to a gradual reduction in soil pH (Fig. 3a), *i.e.* the normal changes occurring in soils following nitrification[1]. Elemental sulphur inhibited nitrification, presumably because the sulphuric acid produced lowered the pH of the soil to such an extent that this acid sensitive process was inhibited (Fig. 3a). However, Maftoun and Banihashemi[9] reported that nitrification was inhibited by sulphur oxidation, without a substantial decrease in bulk soil pH occurring. They suggested that volatile sulphur compounds might be the inhibitory agents involved, but concluded that localized reductions in pH occurring in soil micro-sites were responsible. Another possibility, suggested by Saleh[13], is that thiobacilli compete with Nitrosomonas, thereby inhibiting ammonium oxidation. The fact that nitrification occurred in our experiments when soils were amended with

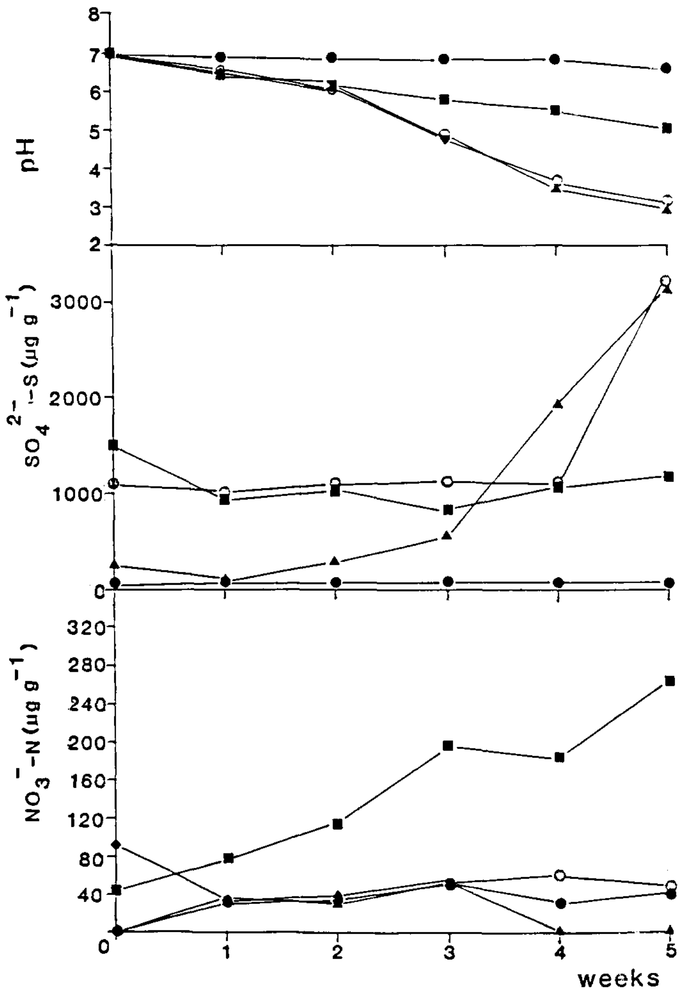

Fig. 3. Change in (a) pH, (b) sulphate concentration and (c) nitrate concentration of loam soil when amended with 1% w/w elemental sulphur (▲—▲), 1% w/w elemental sulphur plus ammonium sulphate (○—○) ammonium sulphate alone (■—■) or left unamended (●—●). Significant difference ($p = 0.05$) occurred in nitrate concentration between soils amended with elemental sulphur and elemental sulphur plus ammonium sulphate from week 2 onwards.

ammonium sulphate in which sulphate concentrations exceeded 1000 μg S g^{-1}, but where the pH remained above pH 5.0 suggests that the reduction in soil pH following sulphur oxidation is the most important factor leading to the observed nitrification inhibition. Nitrification was stimulated in the unamended control soil on incubation (Fig. 3c), while nitrate concentrations in soils containing only elemental sulphur increased but only for the first three weeks of the incubation period. Thereafter the nitrate concentrations in these soils fell, corresponding to a rapid increase in the production of sulphate and a fall in soil pH below pH 4.0. These results again indicate that the inhibition of nitrification following sulphur oxidation is due to the production of acid.

The study of sulphur oxidation in soil has received less attention than nitrification, its equivalent process in the nitrogen cycle. However, it is clear that more information is needed about the factors which influence sulphur oxidation in soils, especially since sulphur deficiencies are occurring in soils throughout the world, the correction of which is likely to lead to a continued increase in the use of reduced forms of sulphur as fertilizers.

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