Elemental sulphur oxidation in Australian cropping soils

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Abstract

Sulphur (S) is one of the essential nutrients for plant growth. Over the last few decades, soil S deficiency has become more common in many countries primarily due to the application of high analysis S-free fertilisers and stricter regulations for industrial S dioxide emissions. To ameliorate soil S deficiency, elemental S (ES) as a fertiliser source is of great interest as ES is cost-effective and less susceptible to leaching than sulphate sources. However, ES has to be oxidised to sulphate to become available for plants. Sulphur oxidation is a biological process and depends on many factors affecting the size (genetic potential) and activity of the microbial population, but the predictability of ES oxidation by S-oxidising organisms has not been studied in soil. This work aimed to 1) examine the relationship between the genetic potential of a soil to oxidise ES and the oxidation rate of ES; and 2) investigate causes for the slower oxidation of granular ES compared to powdered ES.

The relationship between the oxidation rate of ES, and soil physico-chemical properties and microbial populations (indicated by gene abundances) was investigated in ten Australian cropping soils covering a wide range of soil physico-chemical properties in a laboratory incubation experiment. The oxidation rate of ES, estimated from decreases in ES concentrations, varied greatly from 5.1 to 51.7 $\mu g\ cm^{-2}\ d^{-1}$ across soils and was positively correlated with the initial soil pH ($R^2 = 0.54$, $P < 0.05$). A regression equation including pH and organic C content as independent variables explained 79% of the variation in the oxidation rate ($P < 0.01$).
The copies numbers of a functional gene *soxB* was quantified to indicate the abundance of S-oxidising bacteria, and 16S ribosomal ribonucleic acid (16S rRNA) and 18S ribosomal

ribonucleic acid (18S rRNA) to indicate the abundance of total bacteria and fungi, respectively. The abundances of *soxB* and 16S rRNA were positively correlated ($P < 0.05$) with ES oxidation rate ($R^2 = 0.67$ for *soxB* and 0.66 for16S rRNA), but no significant correlation was observed between the oxidation rate and 18S rRNA abundance. This suggests that ES oxidation is dependent primarily on bacterial populations in soils. A combination of bacterial gene abundance (*soxB* or 16S rRNA) and soil pH could explain more than 80% of the variation in ES oxidation rate ($P < 0.01$). A distribution of *soxB* gene across diverse taxonomic and physiological bacterial groups was observed in the soils, which explains the strong relationship between *soxB* and 16S rRNA abundances ($R^2 = 0.99$, $P < 0.01$).

Elemental S is often combined with macronutrient fertilisers and this is generally found to reduce the ES oxidation rate as compared to ES in powdered form. We hypothesised that this reduction may be due to 1) acidification in the soil around the granule (in addition to ES oxidation, acidification can also be induced by monoammonium phosphate with which ES is often co-granulated); or 2) increased ionic strength of the soil solution in the vicinity of the granule from water-soluble fertilisers. Therefore, the effect of increases in acidity or ionic strength on ES oxidation in a sandy soil was studied. Interestingly, neither increases in acidity nor in ionic strength significantly affected ES oxidation in this soil, even though significant shifts in bacterial abundance and community composition were observed due to these changes. An additional experiment carried out at two ES application rates with two different soils showed similar results. This indicates that changes in bacterial abundance and community composition brought about by temporary changes in pH and ionic strength do not necessarily affect ES oxidation. The lack of agreement between bacterial population and ES oxidation might be due to the measurement of the total populations of bacteria, including dormant ones. The consistent ES oxidation (%ES oxidised) across treatments in this experiment suggests that

there were sufficient active populations of S-oxidisers even at high acidity and ionic strength levels. Furthermore, while soil pH related to ES oxidation rate across soils as indicated by our previous study, no relationship was found in the soil acidified for < 15 weeks. This inconsistent effect of pH on the oxidation of ES (across soils *versus* within a soil) can be reconciled by the fact that pH differences across soils are associated with differences in many soil chemical and biological properties, which is not the case for short-term acidification.

As the slower oxidation for co-granulated ES, compared to powdered ES, is likely not related to the chemical changes (pH, ionic strength) around the granule, we speculated that the slower oxidation is due to a reduction in the surface area of ES exposed to S-oxidisers in soil. To test this hypothesis, an experiment was conducted in which ES oxidation, soil chemical properties and bacterial abundance and community composition were compared between powdered (mixed through soil) and granular fertiliser (diammonium phosphate +10% ES). Soil in the vicinity of the granule including the granule was sampled for analysis. Oxidation of the co-granulated ES was much slower than for the powdered ES, with 36% oxidised for the former and 95% for the latter by the end of 20 weeks incubation. This difference was not related to differences in soil pH, bacterial abundances and community composition between these two treatments. Instead, the difference in ES oxidation rate between the two treatments corresponded to the difference in surface area of the granule and that of the individual ES particles, strongly suggesting that the slower oxidation rate for co-granulated ES was due to a reduction in the effective surface area available for S-oxidisers to colonise. Hence, oxidation of ES is not limited by the population of ES-oxidising bacteria in soil, but by the amount of ES exposed to soil organisms.

This work shows that ES oxidation is influenced by both soil biological and physico-chemical properties across soils, and it could be well predicted by two variables i.e. soil pH and bacterial

gene abundance under a steady environment. However, alterations in the abundance and community composition of bacteria resulted from temporary ambient changes within a soil do not necessarily affect ES oxidation, which suggests that the slow oxidation of ES in granules is not related to chemical changes but due to the low degree of dispersion of granules in soil. Therefore, in an effort to improve the effectiveness of granular ES, it is key to improve the exposure of ES to soil microorganisms, e.g. by technically improving granule dispersion in soil or by inoculating S-oxidisers into the granule.

Declaration

I certify that this work contains no material which has been accepted for the award of any other degree or diploma in my name in any university or other tertiary institution, and to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text. In addition, I certify that no part of this work will, in the future, be used in a submission in my name for any other degree or diploma in any university or tertiary institution without the prior approval of The University of Adelaide and where applicable, any partner institution responsible for the joint award of this degree.

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Cuicui Zhao Date

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3. Zhao. C, V.V.S.R. Gupta, F. Degryse, and M.J. McLaughlin. Effects of pH and ionic strength on elemental sulphur oxidation in soil. Submitted to Soil Biology and Biochemistry.

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