SOIL HEALTH : A MULTIFACETTED APPROACH TO UNDERSTANDING THE MICROBIOLOGY OF SOIL

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Background

While the importance of biodiversity in ecosystem function has been accepted for some time (Tilman & Downing 1994), only recently has the relative importance of the diversity of functional characteristics of the biota been recognised (Grime 1997). Studies of above-ground diversity predominate in the literature (Loreau *et al.* 2001). Little is known of diversity in soil. Soil is more complex and biologically diverse than above ground ecosystems (Wardle & Giller 1997). Thus it might be argued that the loss of a small number of taxonomic groups from a complex substrate such as soil will have little impact because many different microbes would contribute to the functions of soil and therefore, many species would be functionally redundant. However, research has found that high biodiversity may be considerably more important in complex ecosystems, such as soil, than in simple systems (Grime 1997) especially when changes over time and space affect the system (Loreau et al. 2001). Soil is the location for mineral cycling, decomposition of organic materials, and flow of energy. Understanding the processes regulating these functions, and their affect on plant growth, justifies understanding the biological diversity housed in soil.

To date, only a small number of studies have examined the effects of microbial biodiversity on plant productivity and diversity. Van der Heijden *et al* (1998) proposed that diversity of species of arbuscular mycorrhizal (AM) fungi directly correlated with total plant biomass, numbers of plant species that establish in soil from seed, and total plant uptake of phosphorus from soil. The interpretation of data from this study was debated (Wardle, 1999; Van der Heijden 1999; Loreau *et al.* 2001). AM fungi play a role in uptake of minerals by plants, especially zinc and phosphorus. In addition, AM fungi modify plant response to the environment, including pathogens, and are associated with formation of microaggregates and are therefore central to soil structure and stability. Two recent studies have demonstrated that in gnotobiotic culture experiments, plant productivity may be increased with increasing species richness of ectomycorrhizal fungi (Baxter & Dighton, 2001; Jonsson *et al.*, 2001). These studies suggest that microbial biodiversity plays an important role in boosting plant productivity, maintaining the plant habitat, and regulating plant community diversity.

Our research addresses three questions:
1. Is soil biodiversity reduced in agricultural soil compared with undisturbed soils?
2. Does cropping affect which groups of fungi are present in agricultural soils?
3. Do microbes increase carbon stored in soil?

1. Is soil biodiversity reduced in agricultural soil compared with undisturbed soils?

Disturbance is a general term that describes activities associated with cropping, or natural phenomena. The first question is mainly being addressed using molecular measures of diversity. DNA is extracted from soil. Markers specific to particular groups of microbes (Archaea, bacteria and fungi, and groups within each) are identified and counted in a process called terminal restriction fragment length polymorphism (TRFLP). TRFLP is also being used to determine whether functional diversity is affected by cropping by examining specific enzymes found within bacteria and fungi. While useful indicators of functional diversity in bacterial communities are available, similar markers are unavailable (or unsuitable) for assessment of functional diversity of soil fungi. As such primers may have to be developed *de novo* for assessment of functional diversity in fungi in cotton growing soils.

Our preliminary data on archaeal, bacterial and fungal biodiversity in cotton cropping, natural and "semi-disturbed" soils suggest that above ground plant communities and soil management practices are correlated with the belowground structure of microbial communities. In addition, early analysis suggests that relatively small variations in agricultural practice such as crop rotation may have a significant effect on microbial biodiversity in soil. The functional effects of these shifts in biodiversity and community structure are still being examined. Further work will provide a better understanding of these processes.

In a small project, we are also examining where propagules of AM fungi survive in cropping soil at the end of severe drought. This has become possible because the recent drought has provided us with a site where we can compare various means to increase the population of AM fungi in soil.

Peter McGee and David Nehl found that even after three years of sustained drought, a few propagules of AM fungi survive in the top soil (less than 10 per hundred gram soil). This is important because earlier research showed that AM fungi recover rapidly under cotton crops (McGee et al 1999), such that propagules at depth might be enough to enable a crop to produce a satisfactory harvest (Nehl et al 1999). David is now determining mechanisms to maximise recovery of AM fungi, and clarify the use of Linseed as a bio-indicator of the size of populations of AM fungi.

2. Does cropping affect which groups of fungi are present in agricultural soils?

The second question is being addressed using three groups of fungi with different functions: arbuscular mycorrhizal (AM) fungi, Trichocomaceae (including *Penicillium* and *Aspergillus*), and chytrids.

AM fungi are recognised within the Cotton Industry as being crucial to growth of plants. They are also fungi with relatively long life cycles, relatively large reproductive units (spores), have slow rates of reproduction, and are relatively susceptibility to disturbance. To date, we have shown that hyphal length and spore numbers in soil from cotton fields are less than from undisturbed soils from other regions (McGee et al 1997, McGee unpublished). Current work supports this earlier finding. Further, species of fungi with small spores are much more common in cotton soils. The reasons for the differences are unclear, but probably relate to the impact of cultivation of survival of fungi that form large spores (takes longer to fill a large spore) and the competitive interactions between fungi in roots and soil. AM fungi have been characterised according to spore morphology. Molecular tools are now being developed (Loke and Saleeba, unpublished) to allow us to determine which fungi colonise roots, the competitive parameters that enable colonisation of the roots of cotton, and the impact of specific management on survival of AM fungi.

Trichocomaceae are important because many members produce highly toxic compounds that enable producers to compete successfully with other organisms. Thus a single fungus will occupy part of a root surface where it protects itself and the section of root from invasion by a competitor. This becomes important if fungi with combative characteristics are lost because of disturbance, and are replaced by fungi that lack combative characteristics.

High diversity of combative fungi is elsewhere associated with lack of disturbance (Markovina unpublished). It is now clear that removal of vegetation changes the diversity of Trichocomaceae. Indeed, fungi specifically associated with individual species appear to be lost as that host species is lost. Specific fungi are mostly found within soil. Above ground fungi appear mostly to be generalist in function. However, the effect of replacement of specific fungi by generalists in soil has not yet been examined in any detail.

Chytrids are fungi that have an extremely short life cycle, small reproductive units, and massive changes in the size of their populations. Thus we might expect them to increase in diversity under disturbance because of the reduction in competitive interactions due to the reduction in the number of competitors. At present, however, their functional characteristics remain for the most part unknown and therefore current generalised hypotheses about chytrids may be incorrect.

It is now clear that soils used for cropping have fewer species of chytrids than either relatively undisturbed soils nearby (eg river banks and roadsides), and significantly fewer than soils of the east coast of NSW. This is odd. Even soils kept fallow for 7 years have some chytrids, indicating that some may not be associated with plant growth at all. This has been tested in experiments (Commandeur et al, in press), and a picture of complex interactions is emerging. We are pursuing a better understanding of the source of nutrients used by chytrids, survival characteristics of some common species, and competitive interactions of chytrids in soil.

3. Do microbes increase carbon stored in soil?

The third area of research is associated with the need of cropping industries to reduce the loss of carbon from soil. Further, soil is an important store of carbon, which on release to the atmosphere increases heating of the planet. The consequences of increasing greenhouse gases are still unclear, though a change in the environment for crop growth is inevitable. Several microbes transfer carbon from plant to soil. AM fungi not only exist for long periods in soil, but they are now known to produce a glycoprotein called glomalin, especially in soils of fine texture (Rillig & Steinberg 2002). Glomalin may exist for long periods in the absence of the fungus (Steinberg & Rillig 2003). Glomalin is important because it contains carbon and thus may be a carbon store. Glomalin also attaches to the surface of and binds microaggregates (Wright & Upadhyaya 1998). Thus it also plays a secondary role in soil structure. We are attempting to understand the role of glomalin in cotton soils leading to a means of increasing the store of carbon in soil, and possibly an improvement in soil structure.

Preliminary work indicates that biomass of AM fungi is influenced by cropping practice. Current work is determining glomalin and total protein levels in cropping and undisturbed soils (Whiffen, unpublished). The next stage of this project aims to clarify whether levels of microbial carbon can be manipulated by farming practices.

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